The role of waterbirds in Australia's 2022 Japanese Encephalitis outbreak - a rapid sythesis

Prepared on behalf of Wildlife Health Australia
for the Australian Government
by BirdLife Australia
June 2022

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Recommended citation:

Purnell, C (2022). The role of waterbirds in Australia's 2022 Japanese Encephalitis outbreak Unpublished – a rapid synthesis. BirdLife Australia, Carlton.

This report was prepared by BirdLife Australia on behalf of Wildlife Health Australia for the Australian Department of Agriculture, Fisheries and Forestry.

Published by BirdLife Australia, Suite 2–05, 60 Leicester Street, Carlton, Victoria 3053, Australia.

This report was prepared by Chris Purnell and distribution maps produced by Bradley Clark-Wood.

Cover photos:
Middle right- Intermediate Egrets in breeding plumage. Peter Scholer
Bottom – Pied Heron, Royal Spoonbill, Little, Intermediate and Great Egret feeding in a tropical lagoon. Chris Purnell

Contents

Executive summary

The unexpected emergence and spread of Japanese Encephalitis Virus (JEV) in Australia in 2022 prompted the Australian Department of Agriculture, Fisheries and Forestry (DAFF) and Wildlife Health Australia (WHA) to begin investigations into the wildlife origins and mechanisms of spread of the virus. Given the role of waterbirds as primary hosts of JEV, WHA facilitated a meeting of national and regional waterbird ecologists to identify data and information that could contribute to biosecurity and health planning response to the current and future JEV threats.

This report builds on the initial engagement provided by the waterbird expert workshop and further investigates the potential role of waterbirds in the 2022 outbreak.

The synthesis identifies possible avian paths for introduction of JEV into the Australo-Papuan region[1] and proposes a climate-driven timeline of events which conspired to facilitate introduction from New Guinea's Fly River floodplain to Australia's Cape York Peninsula and subsequent spread throughout Queensland, New South Wales, Victoria and South Australia.

Significant habitats and flyway components that may be used by potential waterbird hosts include:

- The Middle Fly floodplain of Papua New Guineas Western Province (a likely source of virus and zone of interaction for waterbird taxa from the Asian and Australian zoogeographic regions.)
- The Torres Strait Islands, western Cape York Peninsula and the southern Gulf of Carpentaria (a major flyway for waterbirds including dry-season migrants from the south and a zone of entry into Australia).
- The eastern Murray Darling Basin floodplains bordering the Great Dividing Range (hosts large waterbird breeding colonies, an avenue for spread, maintenance, amplification and livestock interface transmission).

Events which may implicate waterbirds in the role of the 2022 outbreak include:

- The strong El Niño event of 2015-16 that created widespread drought conditions throughout Papua New Guinea drew down the Fly River, increasing large areas of potential waterbird and mosquito habitat throughout the floodplain. The event simultaneously reduced wetland extent and quality throughout Australia. These events are likely to have triggered widescale movement of Pelecaniformes (pelicans and cormorants), Ciconiiformes (herons) and Anseriformes (ducks and geese) including significant movement from Australia into the Trans Fly region (see Figure 2).
- Prevailing dry conditions and sea level height anomalies caused extensive mangrove dieback in the Far North and likely triggered at least temporary abandonment of significant heronries on Australia's tropical rivers and subsequent dispersal in search of new opportunities. Longer-term impacts on the viability of internationally significant heronries in the Gulf Plains are likely as habitat structure and function is yet to recover. The most abundant species and potentially the greatest impacted by loss of tidally influenced colonies was Intermediate Egret – a significant potential host for JEV.

[1] Noting this report was undertaken as a RAPID SYNTHESIS and presents possible pathways for introduction and spread within the Australian context, it was recognised that there may be additional plausible avian pathways that have not been captured. Therefore, additional input from experts was sought during a 2nd expert workshop with national and regional waterbird ecologists in October 2022 and is presented in Appendix 1.

- Five consecutive years of warm winters from 2015 resulted in overall dry conditions throughout eastern and south-eastern Australia. Two years of consecutive positive Indian Ocean Dipole (PIOD) events in conjunction with the consecutive Central Pacific El Niño, resulted in cumulative depleting impact on water in the landscape and culminated in the driest spring (2019) on record across Australia. The Far North experienced three late and/or poor wet seasons culminating in the failed wet of 2019. These events reduced the extent and condition of waterbird habitat in Australia potentially triggering interseasonal immigration to New Guinea.
- In early 2020 flooding affected much of eastern Australia. A La Niña event was declared in September. The 2020-21 A La Niña increased the likelihood of rain-bearing weather patterns over eastern Australia and reduced evaporation due to increased cloudiness and reduced temperatures, replenishing soil moisture. A positive Southern Annular Mode (SAM) reinforced the impacts of La Niña and triggered above average rainfall for most of NSW. Extreme rainfall caused flooding in eastern and central Australia in March 2021 and was followed by continued wet periods in Southern Slopes of NSW to October. November 2021 was Australia's wettest on record and the coolest since 1999. These events triggered wide-scale immigration of waterbirds into the Murray Darling Basin where several megacolonies were established.
- Inundation of large areas of agricultural land in the eastern Murray Darling Basin created significant habitat for Ciconiiformes and Anseriformes breeding in the region and brought large populations of these birds into close and regular contact with breeding *Culex* mosquitoes and areas of high-density pig farming.
- During waterbird breeding events, feral pigs exploit waterbird colonies as a productive food source, preying on eggs and chicks, potentially increasing the likelihood of natural cycles of JEV maintenance, amplification and transmission.
- Recorded instances of JEV outbreaks in commercial piggeries are consistent with opportunistic migration routes identified through satellite tracking of ibis and spoonbills in eastern Australia.

This is the first time the combination of sequential PIOD and strong El Niños followed by widescale flooding and exuberant waterbird breeding has been documented in Australia and therefore presented conditions which:

1) promoted movement of Australian birds into the Trans Fly during the dry climate of 2019 exposing them to conditions conducive to JEV infection and potential amplification
2) facilitated transfer of JEV to the mainland, further amplify and spread of through the Murray Darling Basin during subsequent widescale flooding and exuberant breeding in 2021 and 2022.

Previous El Niño events (1995, 1998) have been implicated in recent incursions of JEV into Australia and exuberant waterbird breeding has also been implicated as a mechanism for spread in four major outbreaks of Murray Valley Encephalitis throughout Australia.

The erratic and sometimes gregarious dispersal of waterbirds from sites of potential infection to locations where they are likely to interact with further vectors and hosts of varying origins, creates a complex network of transmission. Acknowledging that birds may remain infection competent for a maximum of only seven days post exposure, regular and widespread opportunities for onward transmission (e.g. presence of immune naïve hosts during breeding events) must be present for rapid large-scale outbreaks into new regions as recently observed in Australia.

Australia has many large waterbird and wetland datasets which could be useful in modelling climate driven scenarios of JEV introduction and spread by waterbirds and several other One Health applications. However, many of these data exist in siloed databases, and are not dynamically accessible. Furthermore, significant temporal, spatial and research gaps preclude our ability to effectively identify waterbird based epidemiological pathways, episodic triggers and ultimately mitigation measures. These gaps are particularly evident for Australia's Far North & Torres Strait Islands and for New Guineas Trans Fly Region.

Implementation of the following recommended activities is proposed by BirdLife Australia and waterbird experts who were consulted to address these knowledge gaps:

1. Serological sampling
    1.1. Sampling of potential host species, targeting large waterbird colonies, throughout Australia.

2. Monitoring
    2.1. Regular aerial waterbird colony surveys of the Gulf Plains and western Cape York peninsula (replicating work reported by Jaensch & Richardson 2013 & Taplin 1990).
    2.2. Regular waterbird colony surveys of the Far North of Northern Territory (replicating Chatto 2000).
    2.3. Regular waterbird surveys of the Middle Fly region of PNG (Replicating Halse et al. 1995)
    2.4. Exploratory surveys of floodplain, forested freshwater swamps and intertidal wetlands of southern New Guinea.
    2.5. Continued annual aerial surveys of the Magpie Goose populations across the Top End.
    2.6. Continued annual standardised surveys of large colonies in the Murray Darling Basin.
    2.7. Regular surveys of large colonies in the Channel Country, Lake Eyre Basin, when activated.

3. Data management
    3.1. BirdLife Australia to develop its National Waterbird Monitoring Program and associated database which seeks to facilitate wide scale dynamic and referenceable data-sharing and improve context aware coverage of wetlands through standardised monitoring.
    3.2. University of NSW to develop the National Colonial Waterbird Breeding Database.

4. Tracking
    4.1. Satellite tracking of Cattle, Little, Intermediate and Great Egret, Pied Heron, Nankeen Night-Heron and Pelican from colonies in the Murray Darling Basin and Chanel Country.
    4.2. Tracking of Cattle, Little, Intermediate and Great Egret, Pied Heron, Nankeen Night-Heron, Royal Spoonbill and Australian White Ibis from heronries in the Far North.
    4.3. Tracking of Wandering Whistling-Duck and Magpie Goose from the western Cape York Peninsula and Torres Strait Islands.
    4.4. Tracking of Cattle, Little, Intermediate and Great Egret, Pied Heron and Nankeen Night-Heron from populations in the Middle Fly River region of Papua New Guinea.

5. Gene flow studies
    5.1. Gene flow studies targeting potential host species across several regions.
    5.2. Further genetic research using Feather Map samples.

6. Maintain a waterbird ecology working group associated with Wildlife Health Australia to inform health responses to viruses associated with waterbird reservoirs.

These activities will greatly contribute to knowledge of waterbird movements, population structures and dynamics and natal site fidelity and will ultimately help inform epidemiological models of host-disease dynamics in the waterbird hosts and the mechanisms causing maintenance and amplification of arboviruses in Australia.

In addition to One Health outcomes the activities listed above will contribute significantly to conservation strategies seeking to address ongoing long-term declines in the majority of Australia’s wetland dependant birds.

JEV and Waterbird dynamics

Background

Japanese Encephalitis Virus (JEV) is a zoonotic flavivirus that is a major cause of neurological disease in some parts of Asia. Previously thought to be restricted to Asia, JEV has become endemic in New Guinea and subsequently in the Torres Strait region in the late 1990's (1). Although the epidemiology of JEV is not fully understood, transmission between hosts is facilitated by *Culex* mosquitoes and cycles utilise several types of hosts to maintain, amplify and/or act as bridges for the virus within and between species, including humans (Figure 1).

The virus can infect wild populations of a wide range of animals including birds, bats, rodents, horses, cattle, macropods, snakes and pigs. However, most species are considered to be dead-end hosts, capable of being infected but not of being the source of additional infections via further interactions with mosquitoes (2). Waterbirds, in particular egrets and herons (of the family Ardeidae), are suspected not only to be the primary reservoir for the virus, but also a major disperser. While waterbirds may maintain and transport the virus across large scale landscapes evidence from endemic countries suggests they are not considered to amplify virus loads to a sufficient level to allow spillover directly into human populations. It is thought that transmission from waterbirds, via mosquitoes, into high-density pig populations greatly increases the likelihood of amplification of the virus to a level that becomes a threat for human infection (3), although some serological evidence suggests JEV transmission may continue in the absence of pig farms (4).

In February 2022 an outbreak of JEV was confirmed on Australian mainland with cases detected in New South Wales, Victoria and Queensland piggeries. The outbreak is ongoing and widespread with further infections reported west of the Great Dividing Range and west along the Murray River into South Australia.

Although there have been sporadic unrelated detections of JEV in the Torres Strait and Far North Queensland in recent history, this is the first time the virus has been detected in southern Australia. Forty-two cases have been reported in humans in 2022, resulting in five deaths to June 2022. This event represents a significant change in the virus' presence in Australia triggering the Department of Health to declare it a *Communicable Disease Incident of National Significance*.

The unexpected emergence and spread of JEV in Australia in 2022 prompted the Australian Department of Agriculture, Fisheries and Forestry (DAFF) and Wildlife Health Australia (WHA) to begin investigations into the wildlife origins and mechanisms of spread of the virus. Given the role of waterbirds as primary hosts of JEV, WHA facilitated a meeting of national and regional waterbird ecologists to identify data and information that could contribute to biosecurity and health planning response to the current and future JEV threats.

This report builds on the initial engagement provided by the waterbird expert workshop and further investigates the potential role of waterbirds in the 2022 outbreak[2].

In doing so we detail ecological requirements and phenology of waterbirds in the Australo-Papuan region in the context of known JEV reports and recent climate and weather events. We propose a timeline of events which conspired to create ideal amplification, transmission and spread of the virus from Papua New Guinea into Australia's north and then more widely into the Murray Darling Basin. We touch on hypothetical avenues of spread into livestock and humans. Finally, we identify existing datasets and further research which may better inform epidemiological pathways, episodic triggers and mitigation measures.

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[2] Given the urgency of requirement for response to the current outbreak it is acknowledged that this rapid synthesis is not an exhaustive study but introduces concepts of waterbird ecology that should be further explored in an epidemiological context.

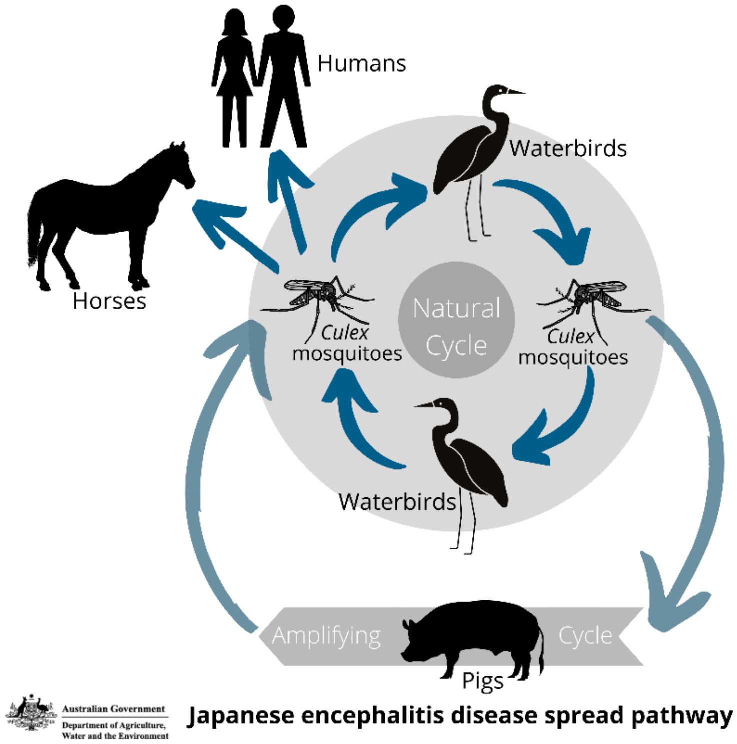


Figure 1. Known transmission cycles for JEV. Department of Agriculture, Fisheries and Forestry.

A new distribution paradigm for JEV and Australian arboviruses.

Japanese Encephalitis Virus (JEV) is a member of the flavivirus genus which contains a large number of arthropod-borne viruses (arboviruses). As with many emerging viruses in Australasia it is thought to have had its origins in the Indo-Malayan region but was not isolated and described until its emergence in Japan in 1870 (5). It has subsequently been found across most of Asia and is the most important cause of epidemic encephalitis worldwide, with 35,000-50,000 cases and 10,000 deaths worldwide identified each year

Until recently the most significant human flavivirus pathogens in Australia have been Murray Valley Encephalitis virus (MVE) and the Kunjin strain of West Nile Virus, both of which are mosquito-borne and largely maintained in mosquito-animal cycles with humans as incidental, dead-end hosts (6). JEV has previously been considered restricted to Asia, but more frequent and pronounced incursions of the virus into the Australo-Papuan region have challenged this assumption. The virus has most likely become endemic to Papua New Guinea posing a constant threat to northern Australia and the potential for further outbreaks throughout the southeast.

When considering how arboviruses are introduced, maintained, and transferred in south eastern Australia there are two main hypothesis of origin (Roshier pers com). The first hypothesis poses that viruses, like MVE, only exist in south eastern Australia during La Niña years and rely on movement from northern Australia by birds participating in major breeding events throughout temporarily inundated, inland water bodies. Once in the region the virus is maintained through natural cycles involving mosquitos and susceptible birds. Once these large congregations of birds disperse and dry conditions prevail, this theory implies that there is insufficient recruitment of susceptible birds in the region and the virus declines to extinction until the next La Niña.

The second hypothesis poses that an arbovirus may be endemic to the southeast, surviving at low-levels between outbreaks and relying on movement and exposure of new, susceptible hosts to maintain itself.

When wet conditions trigger exuberant breeding in the southeast, the virus can then locally amplify, transfer to mobile birds throughout new landscapes and spillover into human populations.

Given the 2022 JEV outbreak is the first of its type recorded outside of Australia's far north, this second hypothesis is not relevant when considering origin. However, given persistent wet conditions, ongoing waterbird recruitment and the likelihood of further La Niña events it may become an increasingly relevant scenario in the near future.

This report explores the first hypothesis to investigate potential origin, pathways and climatic triggers which may have facilitated the 2022 JEV outbreak.

History of JEV in Australia

JEV emerged for the first time in Australia's north when an outbreak of genotype 2 occurred in the Torres Strait Islands in 1995. Investigations into the outbreak identified JEV infection amongst human residents and pigs on nine outer islands. Virus isolations implicated *Culex annulirostris* mosquitoes as the major vector for the outbreak. The species had been breeding in a variety of waterbodies close to and within the communities which contained high densities of pigs, 63% of which were situated near standing water(7).

In 1996 and 1997, JE virus activity was restricted to the northern island of Saibai, about 3 km from the Papua New Guinea coast. In 1998 a geographically widespread outbreak occurred in both the Torres Strait and Cape York Peninsula on the Australian mainland which resulted in five human cases, two of which were fatal. The virus was subsequently identified in the Western Province of Papua New Guinea, and it was thought that this was the source of incursions (1). Following the 1998 outbreak all domestic pigs within the Badu community were relocated to a communal piggery about 3 km from the town to minimize JEV infection by limiting contact between mosquitoes, pigs and humans (7).

Persistent and almost annual JEV activity continued in the area until 2005, although isolation of only genotype 1 suggested that this had been a discrete incursion rather than local maintenance. The incursions of 1998 and 2004 reached Cape York Peninsula, but local transmission was not identified in these instances.

Serological surveillance of animals in the Torres Strait detects some seroreactivity to JEV (and/ or the related viruses) in most years since and there have been occasional serological detections on Cape York (Northern Australia Quarantine Strategy pers comms). No further human activity was detected in Australia until early 2021, when a resident of the Northern Territory was diagnosed with JEV. The 2022 outbreak represents the most intense and widespread incursion in Australian history.

Potential hosts

Black-crowned Night-Heron has been implicated as a major host responsible for maintenance and spread of JEV in Asia and thus herons and allies (Ardeids) have been the subject of many international seroprevalence studies (2). Globally, the ecology of Ardeids makes them inherently susceptible to regular interactions with high densities of mosquitos. Their cohabitation is perhaps the reason many Ardeids have developed a high tolerance to mosquito bites which predisposes them to develop high titres of arboviruses (8, 9).

Among the Ardeids, Little Egret, Black-crowned Night-Heron, Intermediate Egret, Great Egret, Cattle Egret and Pond Heron have been identified as having documented JEV infection and demonstrated infection competence (2, 3).

In the only flavivirus study undertaken in Australia; Nankeen Night-Heron, White-necked Heron, Little and Intermediate Egrets were successfully infected with MVE, Kunjin or JEV. JEV viraemia was detected in all but one bird several days later, but only Nankeen Night-Heron developed sufficient JE viremia to further infect a mosquito vector (2).

As Australo-Papuan fauna are assumed to be immune naïve to the virus, this synthesis does not limit its scope to the species mentioned above. Rather it explores species which have biological, ecological and phenological traits which identify them as potential agents of virus infection, maintenance, introduction to Australia, amplification, spread and/or transmission into high-density pig populations.

Good host species are thought to be typically generalists (in habitat and diet) with a high tolerance to anthropogenic pressures, spend at least part of their life cycle in large aggregations (3) (colonial breeding or high-density roosting), inhabit shallow ephemeral wetlands, semi-permanent ponds or flooded grassland and be capable of large-scale movement both between Australia and New Guinea and within Australia (on a north south orientation). Applying these criteria to advice provided by experts in Australian waterbird populations narrowed candidates to the species of the orders Pelecaniformes, Ciconiiformes and Anseriformes listed (Table 1). Migratory shorebirds were not considered as part of this synthesis given genotyping of Australian JEV isolates identified New Guinean origins and migratory shorebirds, although present, are not known to congregate in large numbers in Paua New Guinea.

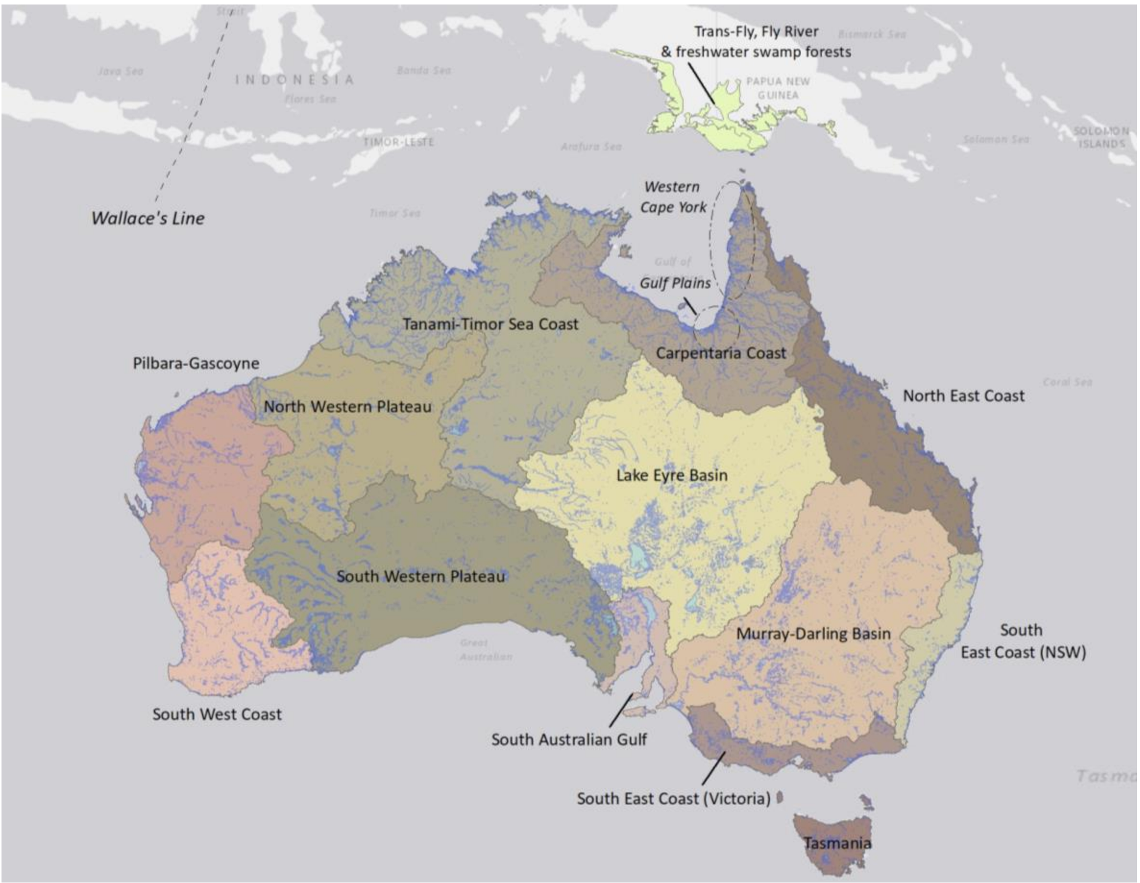
A thorough review of potential wildlife hosts has been provided by Wildlife Health Australia in response to the 2022 outbreak (2).

Table 1. Species of Australian waterbirds considered in this synthesis (10).

Order	species name	common name
Anseriformes	Anseranas semipalmata	Magpie Goose
	Dendrocygna guttata	Spotted Whistling-Duck
	Dendrocygna eytoni	Plumed Whistling-Duck
	Dendrocygna arcuata	Wandering Whistling-Duck
	Anas superciliosa	Pacific Black Duck
	Anas gracilis	Grey Teal
Ciconiiformes	Nycticorax caledonicus	Nankeen Night-Heron
	Bubulcus ibis	Cattle Egret
	Ardea alba	Great Egret
	Ardea intermedia[3]	Intermediate Egret
	Egretta picata	Pied Heron
	Egretta garzetta	Little Egret
	Threskiornis moluccus	Australian White Ibis
	Platalea regia	Royal Spoonbill
	Plegadis falcinellus	Glossy Ibis
Pelecaniformes	Pelecanus conspicillatus	Australian Pelican

[3] The BirdLife Australia working list version 3 identifies the Australian breeding population of Ïntermediate Egret as Plumed Egret *Ardea plumifera* .

Figure 2. Bioregions referred to in this report.



Waterbirds as vectors for dispersal to new geographic areas

High rate and scale mobility, shared habitat preference with mosquitoes and often gregarious habits, foster ideal conditions for arbovirus maintenance, amplification and transmission in waterbirds. To address the potential role of waterbirds in the 2022 outbreak we consider two stages of virus movement 1) potential sources and transfer to the Australian mainland and 2) spread through Queensland, New South Wales, Victoria and South Australia.

The spread of JEV throughout Asia has been attributed to a combination of factors including the expansion of pig and rice-farming into peri-urban areas, the expansion of urban areas into rural settings and the many layered effects of climate change (11). Global warming not only increases the frequency and intensity of flooding in which mosquitoes and waterbirds may congregate and breed, but intense heat and prolonged droughts can subsequently concentrate high abundances of these vectors and hosts around ever shrinking resources (12). In some cases, these dry periods may also initiate large-scale and unseasonal immigration into novel habitats or regions. Rising temperatures have been linked to *Culex* mosquitoes invading and persisting at higher latitudes and elevations and have triggered a global shift in waterbird populations towards higher latitudes where more mesic habitats persist (13).

The proliferation of farming to feed an Asian population of about 3 billion people, which has more than doubled in the last 50 years, has altered landscapes and closed the gap between large populations of humans, livestock and wildlife (14). Manipulation of floodplain hydrology to service agriculture further serves to bring potential vectors into closer proximity to livestock and an expanding urban fringe by presenting, reliable, often resource-rich water sources. In cases such as rice farming, supplemental or preferential habitat that mimics natural wetland conditions may exist over large areas throughout periods of privation in natural systems (15-18).

Seroepidemiological studies of Australian arboviruses have suggested that prevalence is largely dictated by zoogeography (3). As vertebrate hosts are required for virus cycles it stands to reason that arboviruses may be restricted by the generalized distributions of their hosts. Long-term shifts in climate or short-term anomalies may facilitate temporary or incremental shifts in the distribution of vertebrate hosts, which may increase interactions of immune naive species with new virus vectors or hosts. Below we summarise three mechanisms which may facilitate the introduction of virus into a new zoogeographic range.

Intergrading habitat and taxa distributions

In Australasia, Asian and Australian zoogeographic regions are delimited by a seemingly small gap of water known as Wallace's Line. MVE and alphaviruses Ross River virus and Barmah Forest virus are suspected to occur only on the Australian side of Wallace's Line whereas JEV originally occurred only on the Asian side. However, dispersal dynamics imply that realms should inter-grade (19) with different biotas intermixing along a geographical zone of transition. In addition, biotas need not be geographically cohesive. They could, for example, occupy patches of distinctive habitat across a region that are readily colonised by a group of species (20).

The Black-crowned Night-Heron is one species that has been implicated in the spread of JEV in Asia. The species' range, as with many Australian waterbirds, does not comply to Wallace's line, but extends east into the zoological transition zone known as Wallacea – an area which includes Sulawesi, Lombok, Sumbawa, Flores, Sumba, Timor, Halmahera, Buru, Seram, and many smaller islands of eastern Indonesia and independent Timor-Leste (21). While Wallacea doesn't constitute the core range of the Black-crowned Night-Heron it does represent an overlap in distribution with Australo-Papuan species, including the Nankeen Night-Heron (which extends from Australia to the Philippines), and a potential area of virus transmission (Appendix 1)

Range extension of host species

Cattle Egret

The Asiatic Cattle Egret *Bubulcus ibis coromandus* is another species that has been implicated in the spread of JEV in Asia. The species' range expanded dramatically across Asia in the 19th century following changing agricultural practices (16), coinciding with the evolution and spread of the more recent JEV genotypes. The species continued range expansion throughout the twentieth century, extending through the Malay Archipelago into New Guinea.

Cattle Egrets were first reported in New Guinea in 1941 and in Australia in 1948 when large flocks were observed in the Northern Territory. As a species capable of exploiting the vast tracks of pasture utilised for grazing livestock, Cattle Egrets are thought to be continuing to expand their range. Their appearance in Western Australia's isolated south-west in the 50's demonstrates that vast areas of inappropriate habitat pose no barrier to their movement (22). Indeed, Australia has been implicated as the source of the range expansion to New Zealand with banding studies identifying several resightings for birds originating in Queensland and New South Wales (16). As JEV continues to spread in India, Nepal, and Australia we may consider range expansion of host species as one hypothesis for dispersal of the virus.

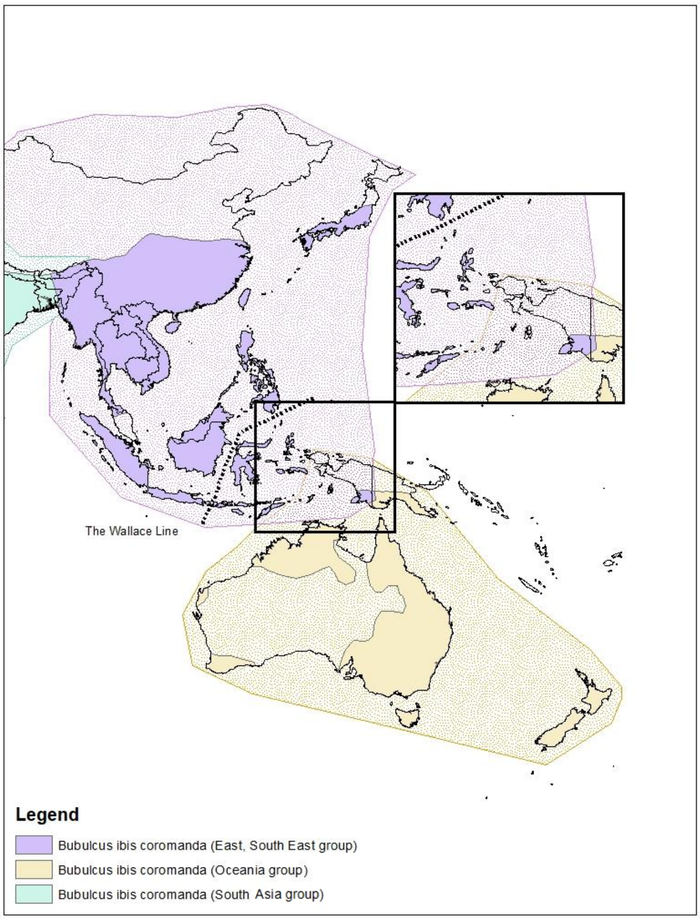


Figure 3. Species distribution of Cattle Egret taxa. Solid areas comprise known distribution. Hashed areas comprise potential distribution. Map generated using composite data from BirdLife International, BirdLife Australia and Wetlands International.

Spotted Whistling Duck and Magpie Goose

Three species of waterfowl also appear to undergoing range expansion across the Australian mainland in the last decade.

Spotted Whistling-ducks *Dendrocygna guttata* are widely distributed on the New Guinea mainland where they are resident, moving in response to resource availability (23). This species was first recorded on Cape York Peninsula in 1999 and in 2000 breeding was expected to have occurred at Chilli Beach, Kutini-Payamu National Park on the eastern Cape (24). A small flock was thought to become resident in the dry season in Weipa by the late 2000's (Birdata). In the subsequent decades records have become more numerous and widespread (Figure 4). Of the 512 records outside the estimated core range, all but four were since 2016, with 75% occurring since 2019 (24-26). Breeding was confirmed on Mungalla station (north of Townsville) in 2018-19 (27). Although Spotted Whistling-Duck appear to be coastally distributed in Australia based on records, it should be noted that likely habitat in western Cape York Peninsula and East Arnhem Land remains largely unsurveyed in recent decades.

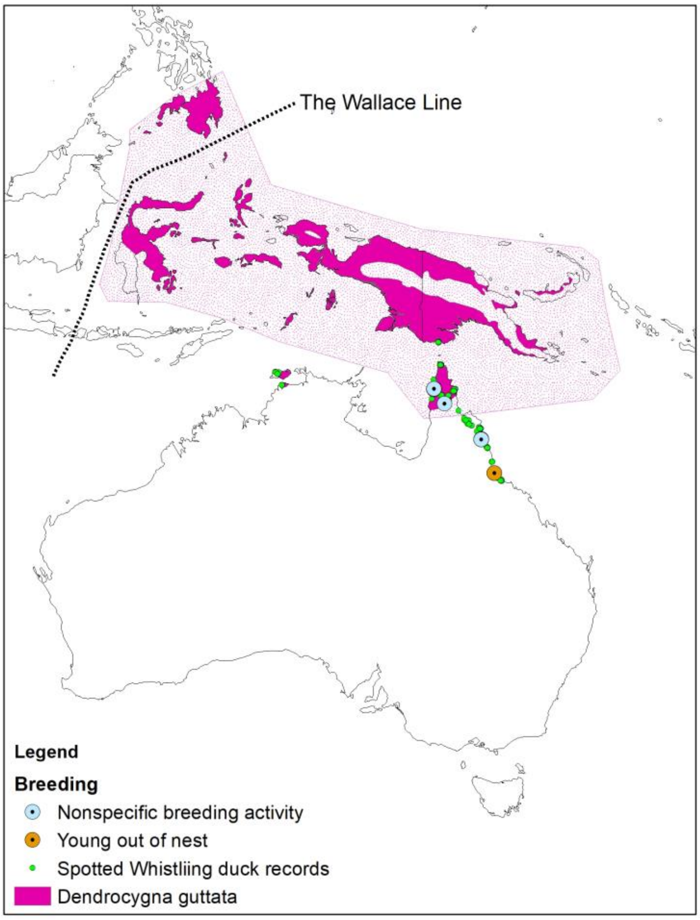


Figure 4. Species distribution for Spotted Whistling Duck including records of range extension from Birdata June 2022.

Magpie Goose A*nseranas semipalmata* presents a case of redistribution. The species underwent a drastic population and range retraction as a direct consequence of European colonisation of Australia. Birds of southern colonies including on the Clarence River, Darlington, Lower Lachlan and Murrumbidgee Rivers were either shot for food or poisoned in large numbers when they grazed in crops. Habitat modification in the Basin further reduced numbers until they were largely absent from NSW, Victoria and South Australia by 1901. Two small breeding populations remain in pockets of southwest Victoria and southeast South Australia but occurrence through the eastern Murray Darling Basin and Riverina is largely opportunistic. In recent years, allocations of eWater and large-scale floods have triggered more regular southerly movement of birds back into the eastern and southern Basin. It has also been suggested that a proliferation of feedlots for cattle along the western slope of the Great Dividing Range may have contributed to recolonsiation (Jaensch pers com). These drivers may increasingly connect birds, likely to be dry season migrants, from the north, into regular contact with southern goose colonies, other waterbird populations and agricultural and livestock settings.

Migration overshoot

While the species featured above don't undertake regular seasonal migration many Ciconiiformes and Anatidae of the northern hemisphere migrate long distances annually. Species or subspecies with defined seasonal migration patterns within the Asian zoogeographic region may overshoot and appear as vagrants on Australian wetlands. Migration overshoot or "flyway confusion" may be common in migratory shorebirds owing to their propensity to breed in overlapping distributions and travel and stage in mixed species flocks along the East Asian Australasian Flyway, however records of vagrant Anatidae and Ciconiiformes are less common for the Australian mainland.

The Northern Shoveler, *Spatula clypeata,* has been recorded in southeast Australia in ten of the last 20 years. The species, which breeds in northern Europe and across the Palearctic, winters in southern Europe, the Indian subcontinent and southern Asia. It is distinguishable from the Australasian Shoveler but is nevertheless likely to be under-reported in Australia with high proportions of positive records originating from popular wetlands often frequented by experienced birdwatchers.

Another taxon which is thought to regularly overshoot migration is the nominate subspecies of Little Egret, *Egretta garzetta garzetta*. Globally the subspecific taxonomy of the Little Egret remains unresolved. Previously, two subspecies were recognised: the nominate *E. g. garzetta* that is widespread north of Wallace's Line into Europe and Africa, and *E. g. nigripes*, that inhabits the Australian zoogeographic region including Wallacea.

Recent scrutiny into the potential occurrence of *E. g. garzetta* in Australia identified that it was recorded in the Northern Territory in 11 years from 2005 to 2017 and several times in Victoria during the same period (28). Again, these observations were made at highly frequented sites by a handful of experienced birdwatchers who were looking for the taxa. In the case of Little Egret, the similarity between Australian and Asian subspecies is not obvious and thus it is suspected that *E. g. garzetta* is largely under reported in Australia. Without clarity on true reporting rates, it is difficult to identify the extent and regularity of occurrence of *E. g. garzetta* or indeed whether its occurrence is a new phenomenon which may play a role in the spread of new and novel viruses to the Australian mainland.

Avenues for introduction - Wetlands and waterbirds of New Guinea and Australia's Far North

Research into the origins of the emergence of JEV in Australasia in the mid-1990s identified evidence of the virus being present in Papua New Guinea's Western Province as early as 1989. The magnitude and intensity of the subsequent 1998 JEV event on mainland Australia, highlighted the proximity and ease of incursion, with JEV activity recorded from southern New Guinea, throughout the Torres Strait and into western Cape York Peninsula (see breakout box on next page for additional considerations).

It has been theorised that the long-term rain deficits persisting in New Guinea from 1991 and culminating in strong and catastrophic El Niño conditions in 1997 (29) were a key mechanism in the 1998 outbreak, acting to concentrate *Culex* mosquitoes around drying, nutrient rich floodplains and triggering widespread breeding. Indeed, the same rationale can be applied to waterbird populations which may exploit resources like floodplain fish and other aquatic vertebrates and invertebrates which become isolated and concentrated in ever shrinking and accessible waterbodies. It is interesting to note that three consecutive El Niño years were recorded prior to the 1995 event and more recently in 2019/20 (Figure 5). These factors may combine to locally maintain and amplify the virus.

Transport of the virus to mainland Australia via wind-blown mosquitoes has been supported by simulations of dispersal which rely on mosquitoes carried by low pressure systems from Papua New Guinea to western Cape York Peninsula (30). The vast wind fields created by Tropical Cyclone Sid in late 1997 would have been ample to facilitate this passage and aligns with JEV activity in the mouth of the Mitchell River. Below we investigate waterbird hosts as an alternate or supplementary mechanism for introduction.

Supplementary information and additional considerations presented after completion of this synthesis.

Although this synthesis investigates one possible transmission pathway – Papua New Guinea to Cape Yorke Peninsula – this theory does not account for a fatal human case of JEV on Tiwi Islands in February 2021. Alternate pathways and mechanisms for JEV introduction were the focus of an October 2022 workshop hosted by WHA with context provided from subsequent and retrospective diagnostic testing.

A summary of additional considerations can be read in Appendix 1. The minutes of the October 2022 workshop can be requested from Wildlife Health Australia.

Published in late 2022, Mackenzie et al (2022) provides further context to the emergence of genotype IV in Australia

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Mackenzie, J.S.; Williams, D.T.; van den Hurk, A.F.; Smith, D.W.; Currie, B.J. Japanese Encephalitis Virus: The Emergence of Genotype IV in Australia and Its Potential Endemicity. *Viruses* **2022**, *14*, 2480. https://doi.org/10.3390/v14112480

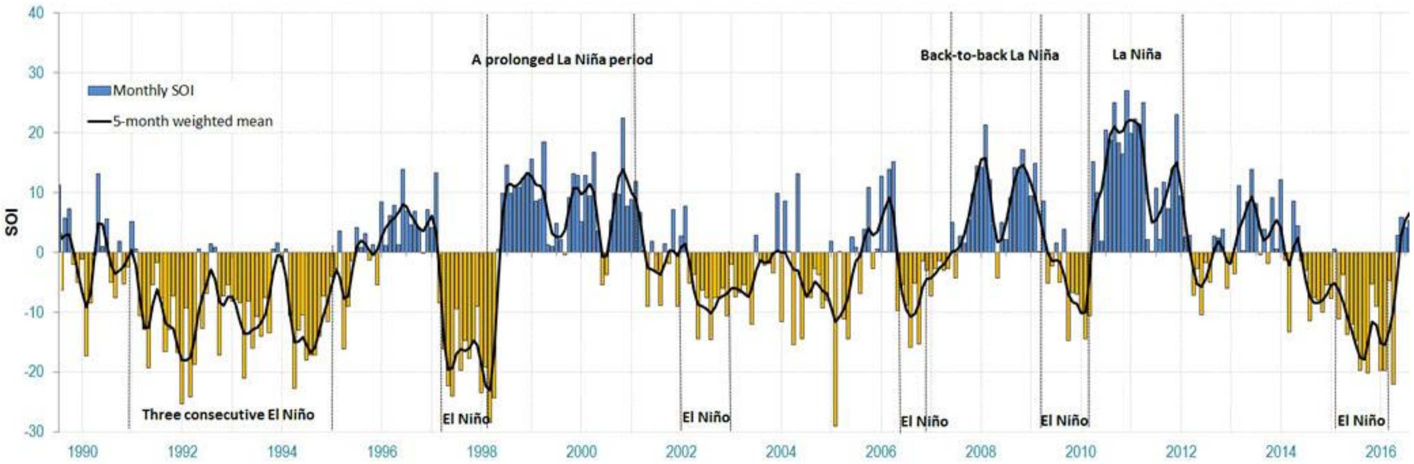

Figure 5. Sustained negative values (bottom/yellow) of the SOI below −7 may indicate El Niño, while sustained positive values above +7 may indicate La Niña. Adapted from http://www.bom.gov.au/climate/enso

Significant habitats, connectivity, movements, and triggers

The Fly River Floodplain, Papua New Guinea

The Trans Fly ornithological region in the southern lowlands of New Guinea has been likened in its biota, and size to the floodplains of the Alligator Rivers (Northern Territory) in places, with large flat areas of savannah and floodplain which are seasonally inundated by the Fly River. The grass and reed dominated floodplains of the middle reaches of the Fly have been documented to support significant populations of Australo-Papuan waterbirds. The floodplain of the region, known as the Middle Fly, bisects dense rainforest which also contains, oxbow swamps and valley lakes. The Fly floodplain can be 30 kilometres wide in places and is of value for waterbirds in the mid to late dry season (June - December). During the dry season blocked valley lakes and oxbows also draw down, exposing increased foraging opportunities for shallow water specialist and grazing birds (31).

The only waterbird censuses in the Middle Fly were conducted in the late dry of 1994 and early dry of 1995 (31). These expansive aerial and ground surveys recorded *c*.587,000 waterbirds in the late dry season. The most abundant species were egrets *c*. 231,000 individuals[4], (comprising 130,000 Intermediate, 90,000 Great and 1,500 Little Egrets), Glossy Ibis (*c*.60,300), Australian White Ibis (*c*. 50,400) and Magpie Geese *(c*.48,000). As noted above, 1994/95 was the third consecutive year of El Niño events and the floodplain may have been uncharacteristically dry but the depth and gradient of drawdown in valley lakes and oxbows provided abundant habitat. During this period researchers noted the extraordinary abundance of mosquitoes (Halse pers com). Indeed, mosquito collections in February 1998 in the Western Province were very high, with many traps catching over 10,000 mosquitoes per night (1).

Subsequent waterbird surveys of the same region shortly after the wet season returned abundances an order of magnitude lower than 5 months earlier (≈55,000 total individuals in total). The wide-scale inundation of the floodplain and oxbow lakes during the wet reduces the availability of shallow foraging habitat and triggers an exodus of wading and shallow water grazing species. The most abundant species remaining in the April surveys were Magpie Geese (c.19,000), many of which were breeding or had bred.

Unlike the floodplains of Australia's Far North, the Fly floodplain does not support large populations of mammals and with 80% of Papua New Guinea's rural communities practising subsistence farming there are no high-density populations of domestic pigs in the region (32), Storey pers com)[5]. Thus, low densities of wallabies, feral pigs and Rusa Deer may provide the only alternative bloodmeal to waterbirds for *Culex* mosquitos. With an ample availability of habitat and seasonal concentrations of waterbirds around resources and breeding events, this reliance on waterbird cycles may significantly amplify within season viremia.

In recent decades, significant volumes of waste rock and tailings from upstream mining operations have resulted in elevated levels of suspended solids and riverbed aggradation in the Fly River system, particularly in the upper reaches. Following the 1997-98 El Niño drought, there was a prolonged La Niña period (1999-2001) in which high volume flooding and mobilisation of stored sediment initiated a stepped-change in water levels across the Middle Fly region. As a result, river water level is minimum of 1mADH higher and the floodplain now only dries under extreme droughts. Although the process has reduced the overall extent of shallow floodplain habitat it has also worked to convert flooded forest into open swamps (33). Regardless, researchers active in the region have observed an apparent decline in waterbird

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[4] Interestingly only 1 Cattle Egret was recorded.

[5] Although animal product makes up less than 3% of lowland diets nationally, 60% of all households claimed pig ownership in the 1990 census with 1.8 million pigs being raised in villages. Herd sizes were typically <20 pigs and pig to human rations are higher in the highlands (1.2:1) than the lowlands (0.3:1) (18).

abundance throughout the 1995 study area. No further comprehensive waterbird surveys have been conducted of the Middle Fly and it has not been established whether a long-term decline has truly occurred or whether these waterbird populations have emigrated to new and transitioning habitats in the region.

At least part of the population displaced either seasonally from the Middle Fly during the wet season, or for longer periods (given the changes mentioned above), are likely to use the more seasonal Bensbach River (PNG) and Wasur (Indonesia) wetlands. However, these areas cannot account for the abundance of birds observed in the 1994 surveys. With little evidence of significant movement to wetlands to the east or west (Halse pers comm), the Australian mainland (200kilometers from the Fly estuary) is the most likely destination for regular, short or longer-term, migration of a large cohort of these birds.

Although there has been little research targeting movement dynamics between Australia and New Guinea, band and wing tag resights of Intermediate, Little and Great Egrets have been recorded as far as 3,000km from their natal colony (34). These include several first-year Intermediate Egrets banded in the Macquarie Marshes in NSW and resighted in the Middle Fly in December. Banded Nankeen Night-Herons have also been recovered from Papua New Guinea including several local movements from large heronries in the Torres Strait but also one bird originating 2,992 kilometers south.

Gene flow studies of Magpie Geese and Wandering Whistling-Ducks in the Australo-Papuan tropics suggest that there are complex source sink dynamics tying New Guinea to Cape York Peninsula. Interestingly, there was genetic separation of two flocks of Wandering Whistling-Duck sampled only kilometres apart on Cape York Peninsula, suggesting the two populations are derived from independent sources and dispersal events from New Guinea (35). This supports theories that the western Cape is a significant flyway for waterbirds moving not only from Australia to New Guinea but also on into Wallacea (albeit in lower numbers). The flyway and habitat of the Trans Fly may also facilitate interaction between Australian birds and those of predominantly Asian or New Guinean origin (species or sub species) which may be carrying JEV (34-36). This mechanism for reintroduction and the apparent reliance on waterbirds for bloodmeal in the absence of other large populations of mammals, may account for the virus's ability to persist in New Guinea and feed repeated incursions into the Torres Strait and Australia.

Unlike migratory shorebirds and several species of terrestrial birds, waterbirds of the Australo-Papuan region do not undertake predictable large-scale, seasonal migration beyond the Australian mainland. Instead, Australian waterbirds may be defined as partial migrants in that portions of widely distributed population may exploit habitats within the Torres Strait, New Guinea and/or the Wallacean Islands of Indonesia in response to resource availability. This may be associated with annual wet and dry cycles but is contingent on how those seasons manifest and the associated impacts on resources on annual or interannual scales.

Avenues of spread - Australian waterbird ecology: maintenance, amplification, and transmission

Torres Strait Islands

The islands of the Torres Strait provide important staging areas for waterbirds migrating to and from Papua New Guinea and mainland Australia. Indeed, the outer Islands have been the site of several JEV outbreaks in recent decades (7). Many potential JEV carrying waterbird species are annual passage migrants or dry season residents through the western group of islands.

Magpie Goose, Wandering Whistling-Duck, Australian Pelican, Australian White Ibis, Great Egret, Pacific Black Duck and Nankeen Night-Heron are widespread and occur in the Torres Straight throughout the year. Some species are observed flying to and from the New Guinea mainland to roost at dusk. Nankeen Night-Heron and Pacific Black Duck breed in the north west islands in some years (37). Magpie Geese move in large numbers throughout the western Islands at all times of year although many birds disperse to both New Guinea and Australia during the wet to breed.

Far northern Australia

Once across the Torres Strait the complex anabranching system of floodplain drainage networks on western Cape York Peninsula provide a diverse range of wetland habitats for waterbirds. The Mitchell River Catchment contains Australia's largest fluvial megafan with extensive wetlands of the Wenlock, Archer and Jardine River systems and supporting large perennial and seasonal wetlands including salt flats, dune lakes, lagoons, marshes and oxbow lakes (38). The wetlands of the region are known to support small breeding colonies of Magpie Geese, ibis, spoonbill, herons and egret however the only large-scale census was completed in 1990 when a failed wet season was assumed to be complicit in very poor breeding participation (39).

In the southeast Gulf of Carpentaria (the Gulf Plains bioregion) the floodplains of the Leichhardt, Flinders, Bynoe, Norman and Gilbert rivers and their estuarine mangrove forests support over 30 documented colonies of Ciconiiformes and other waterbirds (40) several of international significance. This region sits strategically on a significant flyway between the Torres Strait and the major floodplains of the Murray Darling and Lake Eyre Basin.

The coast and major floodplains of Australia's Far North support internationally significant numbers of colonial nesting waterbirds, particularly Magpie Goose, Intermediate, Great, Little and Cattle Egrets, Nankeen Night Heron, Australian White Ibis and Royal Spoonbill (40-44) (Figure 7).

The most significant floodplains in the Northern Territory are those of the Moyle, Daly and Finniss Rivers west of Darwin, and the Adelaide, Mary, Wildman, South Alligator, East Alligator Rivers east of Darwin into Arnhem Land. Although the Carpentarian Barrier does not impede connectivity between these floodplains and those of Queensland, there may be few incentives to leave the region with many waterbirds capable of finding dry season refuge within this large network of habitat(45). However, there must be some exchange with other bioregions to the southwest, east or far inland, because despite occurrence of large numbers of Straw-necked and Glossy Ibises in the drier months, there are no regular breeding colonies of these species in the Top End of the Territory.

The colonies of the Top End's tropical river floodplains are especially significant as they're active most years due to relatively reliable monsoonal rainfall. The colonies occur in mangrove-lined estuaries and freshwater riverine or off-channel woodland swamps. Overbank flooding creates vast areas of floodplain swamp, drawing down in the dry season to provide highly productive habitat in off-river lagoons, back-swamps and residual marshes. Several of the individual river systems mentioned above support >10,000 breeding pairs of egrets and herons which may be dispersed in several staggered colonies as large as 5km wide.

In most years the birds of these colonies may see their requirements relatively well met locally and move between more coastal and upstream freshwater river systems (spanning as much as 40kms) between wet and dry periods. Tracking studies of Magpie Goose in the Darwin region identified variability in individual responses in post dry season dispersal. Birds were recorded dispersing to local <50km and distant <550km floodplain systems in the wet season to breed. Magpie Geese returned to the same floodplain systems between wet seasons and some spent dry seasons in the same peri-urban environments (45). This illustrates both a degree of site fidelity and the ability for waterbirds to exploit anthropogenic settings in times of privation.

These Far North 'resident' populations may be supplemented by birds arriving from more variable floodplains in northwest, central or eastern Australia or New Guinea in annual or episodic periods of resource shortages. Indeed, large number of waterbirds in the tropical river systems do not breed in the area or do so relatively infrequently compared to other parts of their range. These species, including Yellow-billed Spoonbill, Pelican and Straw-necked Ibis are typically non-breeding dry-season visitors to the Far North. Some visit the area on a more or less annual basis and in considerable numbers, whilst others are more nomadic (44). Banding and tracking studies have identified several species of Egret and Ibis regularly dispersing from colonies in the Murray Darling Basin to the Far North only to return to their natal colony when sexually mature (16, 18, 46, 47). Equally, in the absence of reliable anthropogenic resources, in the Far North populations may emigrate from the region if the timing and extent of the wet season is impacted and conditions are more favourable to the south.

Situated strategically on the major waterbird flyway between New Guinea and the major floodplains of Australia's southeast, the colonies of the Gulf Plains may be highly significant to the phylogeography of waterbirds in the Australo-Papuan region for several Ciconiiformes. Even if simply considered a staging area for transient birds, the role of the Gulf Plains in the spread of JEV may be significant.

Several of the floodplains mentioned above also support high densities of communal roosting species like Wandering Whistling-Duck and Plumed Whistling-Duck. Whistling-ducks regularly form "camps" of several to tens of thousands of birds on the edges of persistent lagoons and waterholes throughout the dry season. These congregations are most abundant on the extensive lagoons of the mid reaches of the South Alligator River, on the plains of the Mary and Daly rivers and the floodplains of the Keep and Victoria Rivers (the Legune wetlands) on the border of Western Australia and the Northern Territory and in the wetlands of the Barkley Tablelands. Large dry season concentrations may also be observed in Darwin's peri urban fringes after productive breeding seasons like 2022 (Figure 6).

Owing to the year-round abundance of *Culex* mosquitoes, the zoonotic cycle between mosquitos, birds and pigs may happen continuously in these tropical areas. However, research into the diet of *Culex annulirostris* mosquitoes in the region shows a preference for wallabies and other macropods. Higher proportions of waterbird and pig infection may be evident only when these species are in high-densities. In more temperate areas, high levels of rainfall are required to increase mosquito populations and potentially allow infection to build up in large populations of waterbirds and then pigs, causing risk of JEV to humans exposed to infected mosquitoes (1, 48).

Figure 6. Plumed and Wandering Whistling-Duck roost in a roadside lagoon on the fringes of Darwin in early 2022. Photo: Gavan Keane

Southeastern and Central Australia

In Australia's temperate and arid environments, waterbirds respond to the availability of resources and habitats on varying scales depending on their phenology and ecology, however the abundance of most functional groups has been found to be most positively related to annual rainfall and riverflow (14, 49).

In the Murray Darling Basin (MDB), large flocks of waterbirds congregate around mosaics of sedgelands, marsh grasslands, lignum shrublands, forests, open lagoons and lakes during seasonal flooding. These mosaics ensure a diversity of habitats to support different waterbird functional groups and keep wetlands primed to support future breeding events (50). Broad-scale flooding outside breeding colony sites is also vital to generate food resources and ensure breeding attempts result in effective recruitment by increasing the likelihood of nestlings surviving until capable of independent dispersal (18). In the MDB these habitats often include agricultural landscapes. In addition to rainfall and riverflow, minimum winter temperature, maximum summer temperature and antecedent rainfall have also been found to be significant influences for the abundance and distribution of dabbling ducks and other shoreline foraging waterbirds (51).

High maximum summer temperatures reduce overall soil moisture content increase evaporation and reduce waterbird habitat extent and quality. This is likely to be especially pronounced in the shallow floodplain wetlands preferred by shoreline foragers and dabbling ducks and in the absence of supplemental habitat availability, can act to condense populations and suppress breeding (52). Milder winters in association with high annual rainfall may promote a longer growth window for wetlands vegetation, increase decomposition and overall wetland productivity throughout all trophic levels.

Australian waterbirds have developed life-histories that are adapted to long periods of privation and episodic periods of, large-scale, intense but temporary productivity. Consequently, when rivers flood, many ecosystems respond rapidly and large numbers of waterbirds may arrive from disparate regions to exploit the pulse in resources and begin to breed (49, 53, 54). This response is perhaps more pronounced in the unregulated riverine ecosystems and floodplain wetlands of the Channel Country of the Lake Eyre Basin (LEB). Unlike the relatively permanent flowing, discrete river channels of the MDB, the three major Channel Country rivers typically comprise vast networks of anastomosing channels and anabranches situated in a broad floodplain, fed mainly by flood waters from rainfall in external, less arid regions to the north. Small to moderate floods occur every few years, major floods in one or a few years per decade. During the wet periods, wetland habitat complexity and availability in the Channel Country is immense with millions of hectares of swamps, channels, lakes, billabongs and waterholes on the distributary floodplains (55). Many of these sites rank among the more important in Australia in terms of abundance and species richness of waterbirds. During flood or 'boom' periods, these provide significant areas of habitat for waterbirds, water persisting in some sub-terminal lakes for several years (56). In dry periods these rivers are usually reduced to a series of waterholes in the channels and isolated lakes. Over 40 colonies of waterbirds were documented in the Channel Country from 2000 to 2009, some holding over 20,000 pairs (Costelloe et al. 2004; Jaensch 2009; Reid et al. 2010).

Colonial waterbirds (both Pelecaniformes and Ciconiiformes) respond to large flows and extensive flooding by breeding in large colonies but usually at relatively few continental locations (50) (Figure 7).

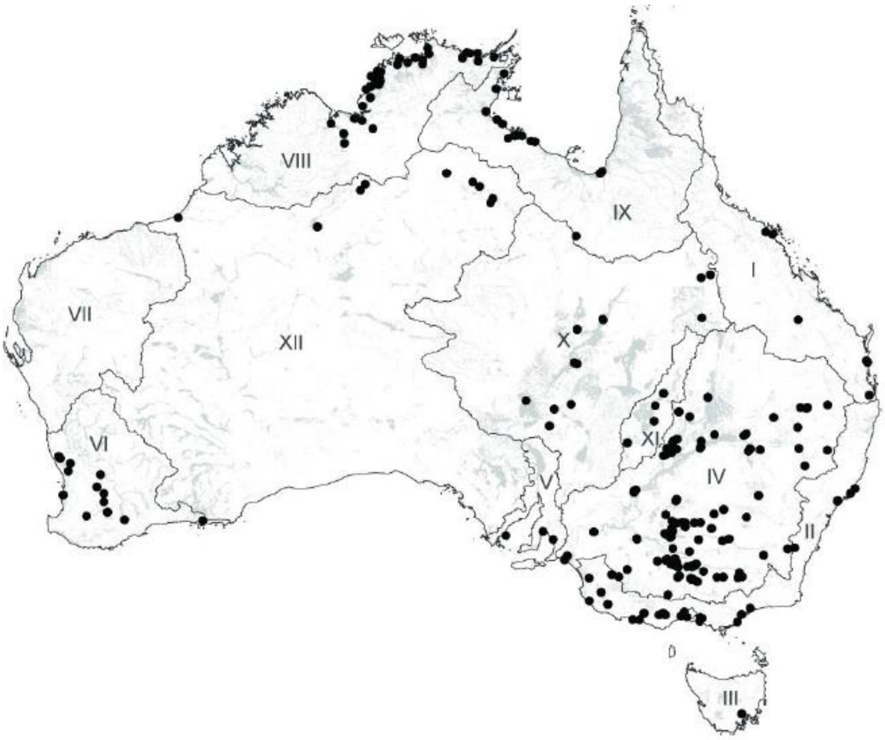


Figure 7. All known colonial waterbird breeding sites (1899-2008) across Australian drainage basins reported in Brandis 2010.

Episodic congregations of colonial breeders can number in their hundreds of thousands and consist of several species. Historically these opportunistic mega-colonies were common throughout the Murray Darling and the Lake Eyre Basins (Figure 7) however intensive hydrological alteration including damming and water diversions to service agriculture have dramatically impacted natural wetting and drying phases in the MDB. With the assistance of targeted environmental watering these colonies are still a feature of the Murray Darling Basin wetland landscape with climate influenced events in iconic sites like the NSW and Victorian Riverina, the Narran Lakes, Macquarie Marshes and Gwydir wetland complexes.

Resource partitioning between species may allow for several colonial breeding species to nest concurrently, utilising variable nesting and foraging habitat in high densities to take advantage of pulses in resources. Wetland mosaics may attract birds from throughout the country increasing potential transmission of viruses and onward transport to new locations.

Nestlings of colonially breeding species of the orders Ciconiiformes and Pelecaniforme may be particularly susceptible to mosquito-borne viruses, as they are altricial or semi-altricial (possessing no or very sparse down (Figure 8)) and nidicolous (confined to the nest) (47, 57). Globally, high arbovirus titres are more commonly found in nestlings than in juveniles and adult. Adults in populations with previous exposure to a virus are more likely to possess antibodies, than viraemias.

Waterbird colonies likely provide a platform to supercharge natural virus cycles and a temporary point of viral dispersal through the daily foraging flights of colony inhabitants. Recent satellite tracking of Australian White Ibis, Straw-necked Ibis and Royal Spoonbill identified that birds target foraging habitat 1-3km from the colony (18). It is important to note that these daily movement routinely incorporated feeding in inundated or damp grass in agricultural landscapes. The breadth of diet displayed by ibis, egrets and herons allows them to exploit these novel habitats. Additionally, recent satellite tracking of Australian Pelican from one colony in Victoria's Gippsland Lakes observed regular daily visits to forage in the local tip, illustrating the generalist behaviour of a species that is widely thought to be an obligate piscivore (D Sullivan pers comm).

Figure 8. An Australian Pelican and 2-week-old chick starting to develop downy plumage in Victoria's Gippsland Lakes. Photo Deb Sullivan.

These satellite tracking studies have reinforced research utilising band resightings which identified distinct variations in movement behaviour both between and within species. Birds displayed varying degrees of resident, nomadic and migratory behaviour post breeding (16, 18, 47, 49, 53, 58, 59). Dispersal distance may be dependent on knowledge of local resources or the age of the individual with some evidence that juvenile birds initially disperse further than adults (16). During post breeding dispersal, birds may fly independently or part of large flocks. Cattle Egret have been recorded in travelling flocks of 700 individuals including members of at least four breeding colonies (16).

The erratic and sometimes gregarious dispersal of waterbirds from sites of potential infection to locations where they are likely to interact with further vectors and hosts of varying origins creates a complex network of transmission. Acknowledging that birds may only remain infection competent for a maximum of seven days post exposure, regular and widespread opportunities for onward transmission must be present for rapid large-scale outbreaks into new regions as recently observed in Australia.

Perhaps the only consistent pattern displayed for ibis and spoonbill dispersing from natal colonies in the Riverina of Victoria and NSW was the general preference for north south movement utilising an apparent flyway down the western base of the Great Dividing Range[6] (18). This pathway, tracking the eastern borders of the Murray Darling Basin, may represents a corridor of more reliable staging areas owing to more extensive, connected floodplain regularly fed by high rainfall– a characteristic that has leant the area to intensive agriculture and livestock production. It is important to note that the findings of this study demonstrate significant overlap between positive detections of JEV (both in pigs and humans) and sites networks and pathways used by ibis and spoonbill. Furthermore several individuals were subsequently observed utilising wetlands of the Channel country and the tropical rivers demonstrating potential pathways from direct point of entry into Australia.

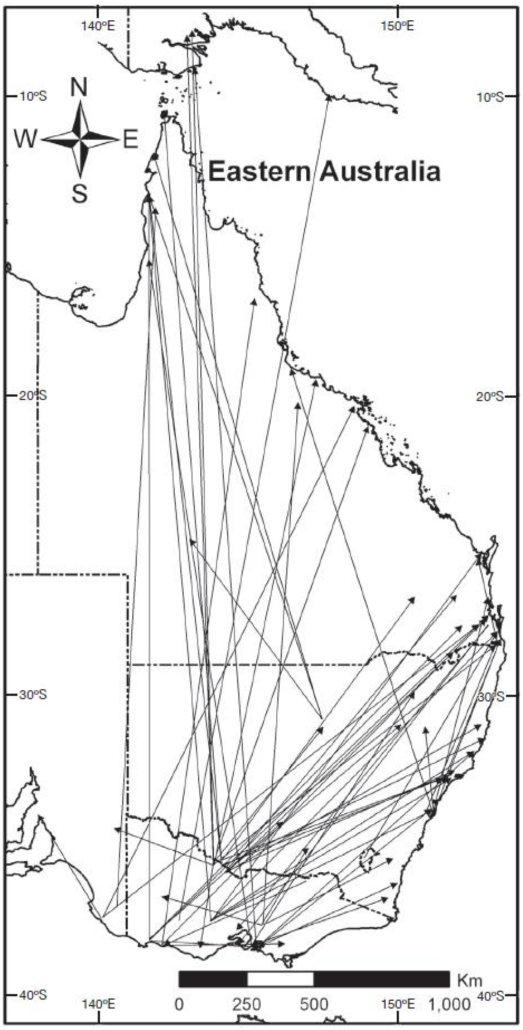


Figure 9. Figure 10. Regional movements (≥ 300 km) of Australian White Ibis (n = 65) along eastern Australia from band recoveries 1955 to 2007.

Waterbirds and pigs

Pigs, feral *Sus scrofa* or domestic *Sus domesticus*, are considered the main amplifying host for JEV, and their infection can correlate with human cases of disease. Intense periods of JEV transmission on Badu Island were facilitated by large community-based pig populations. Investigations into the 1995 outbreak identified that the majority of *Cx. annulirostris* (31%) obtained their blood meals from pigs (7). A contrasting pattern was observed on Cape York Peninsula where pigs accounted for only 10% of blood meals despite the presence of a large, widespread feral pig population, but with few domestic pigs kept in communities (1).

Like waterbirds, feral pig breeding is heavily influenced by the availability and quality of food and habitat. Favourable conditions allow feral pigs to reproduce all year round and at a rapid rate. Sows can breed once they reach about 25 kg or six months of age and can potentially produce two litters of 4–10 piglets in a year. In the Far North, pigs seek out wetlands due to the abundance of food and to moderate body temperature. Part of pig diets throughout their Australian range consist of bird eggs, indeed waterbird colonies may be the target of groups of feral pigs who may decimate birds nesting in sedges, rushes and lignum (18). In 2022 the first cases of Japanese Encephalitis (JEV) were detected in the Northern Territory,

[6] Maps illustrating the paths used by waterbirds can be viewed in *CSIRO & CEWO Waterbird Movement and Breeding research, McGinness et al:* https://research.csiro.au/ewkrwaterbirds/; https://flow-mer.org.au/basin-theme-biodiversity/; https://ewkr.com.au/waterbirds-2019-research-report/).

after a small number of feral pigs in the West Daly region tested positive to the virus. JEV has subsequently been detected in feral pigs in Mapoon on Cape York Peninsula. Even considering an increasing population of feral pigs throughout their Australian range densities of populations may only be sufficient to maintain sporadic outbreak. It is thought that the presence of high-density pig populations greatly increases the likelihood of effective amplification and widescale spread of the virus, particularly into human populations (3, 5). This may prove sufficient as a supplementary mechanism of maintenance amplification and transmission

The high-density piggeries of Australia's southeast were significantly impacted by the 2022 outbreak. The distribution of the effected piggeries, largely down the east and southeast boundary of the Murray Darling Basin, correlates closely with the generalised flyway and breeding season distributions recently identified for Ibis and Spoonbill (60)

Figure 10. In addition to areas which may become inundated, the broad diet of some Ciconiiformes allows them to feed opportunistically on refuse (e.g., feed) and abundant invertebrate populations associated with livestock. Indeed, several species that routinely feed in pasture, marsh and grassland habitats may closely follow larger mammals (cattle, pigs, horses, buffalo) using them as 'beaters' to flush their prey out of hiding. This behaviour is best understood in Cattle Egret (47).

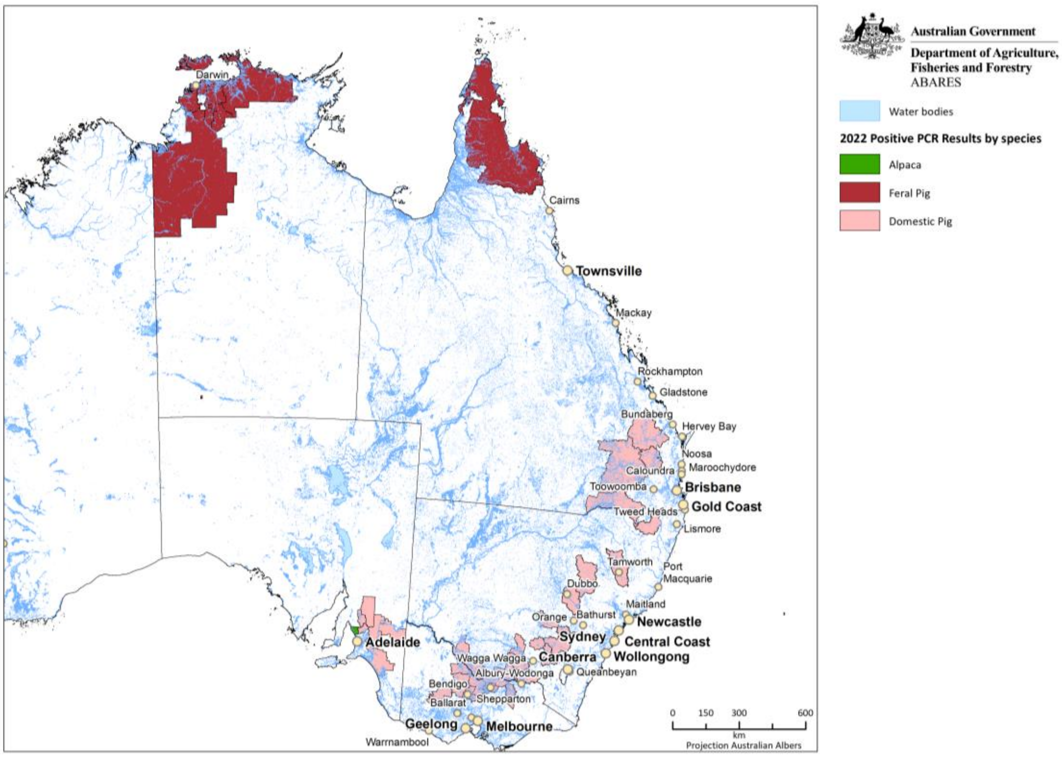


Figure 10. Distribution of confirmed JEV infections in vertebrate animals by local government area, 1 January to 30 June 2022. Source https://sciquest.org.nz/search/results-2/downloadfulltext/171855

The 2022 outbreak

Climate implications of 2015-2022

Here we propose a timeline of events which conspired to create ideal amplification, transmission and spread of JEV from Papua New Guinea into northern Australia, and then more widely into the Murray Darling Basin.

2015-16 El Niño

The strong El Niño event of 2015-16 (Figure 5) caused widespread drought and frost in Papua New Guinea, peaking in late 2015 before breaking in April 2016 (Figure 11). The event was likened to the 1997-98 drought in its scale, duration and intensity and was sufficient to completely draw the Middle Fly River down to 0m AHD and dry the floodplain. Despite the step level change in river height in 1998, these low levels had only ever been recorded during the El Niño of 1987 (61). Similar magnitudes were observed in El Niños of 1993, 1997 and 2004. The system recharged again with the wet of 2016 but the significant drawdown would have provided enormous incentives for wading and grazing waterbirds to exploit the Trans Fly to feed throughout the drying gradient.

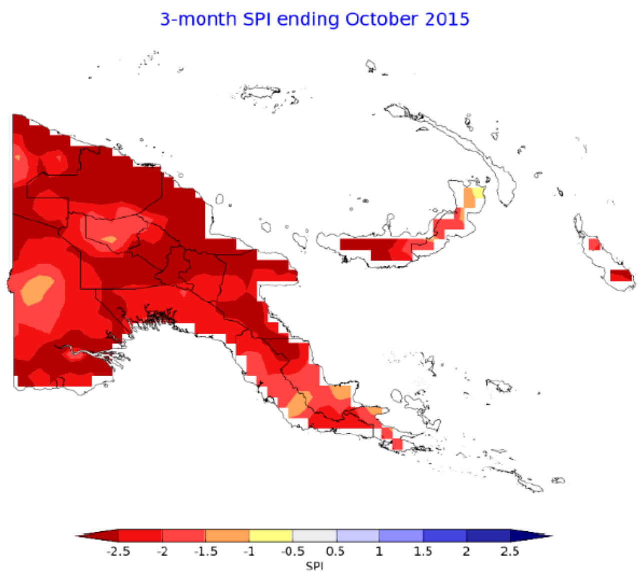


Figure 11. Three month Standard Precipitation Index at the height of the 2015-16 drought in Papua New Guinea. (Z-W Chau et al 2020)

Throughout Australia, the 2015-16 El Niño increased rainfall deficiencies. Pervasive dry conditions and sea level height anomalies associated with the event were implicated in the dieback of about 8,000 hectares of forested tidal wetland in the Gulf of Carpentaria including large areas of the Gulf Plains and western Cape York Peninsula. Mangrove dieback was also observed in Kakadu National Park (62).

The dieback, from which the mangrove and Teatree community still has not recovered, may have had long-term implications for significant breeding colonies of Gulf Plain's heronries. These colonies - several of which numbered >10,000 pairs each in 2011 - contained significant populations of potential host species, namely Nankeen Night-Heron, Intermediate Egret, Royal Spoonbill and Australian White Ibis. In dry years colonies in the middle reaches of these river systems rely on rainfall to generate small floods that inundate some floodplain but some of this water is unlikely to reach the saline coastal zone (40). The severity of the El Niño in 2015-16 is likely to have precluded even small-scale flooding of these floodplain areas and is assumed to have led to the abandonment of breeding attempts across some if not most of the colonies on the Gulf Plains (Figure 12) and western Cape York (39)

The most abundant species nesting in the region and potentially the greatest impacted by loss of floodplain foraging opportunities was Intermediate Egret.

Similar effects of the 2015-16 El Niño were likely experienced at significant heronries across the Far North though perhaps not on the structural level (63).

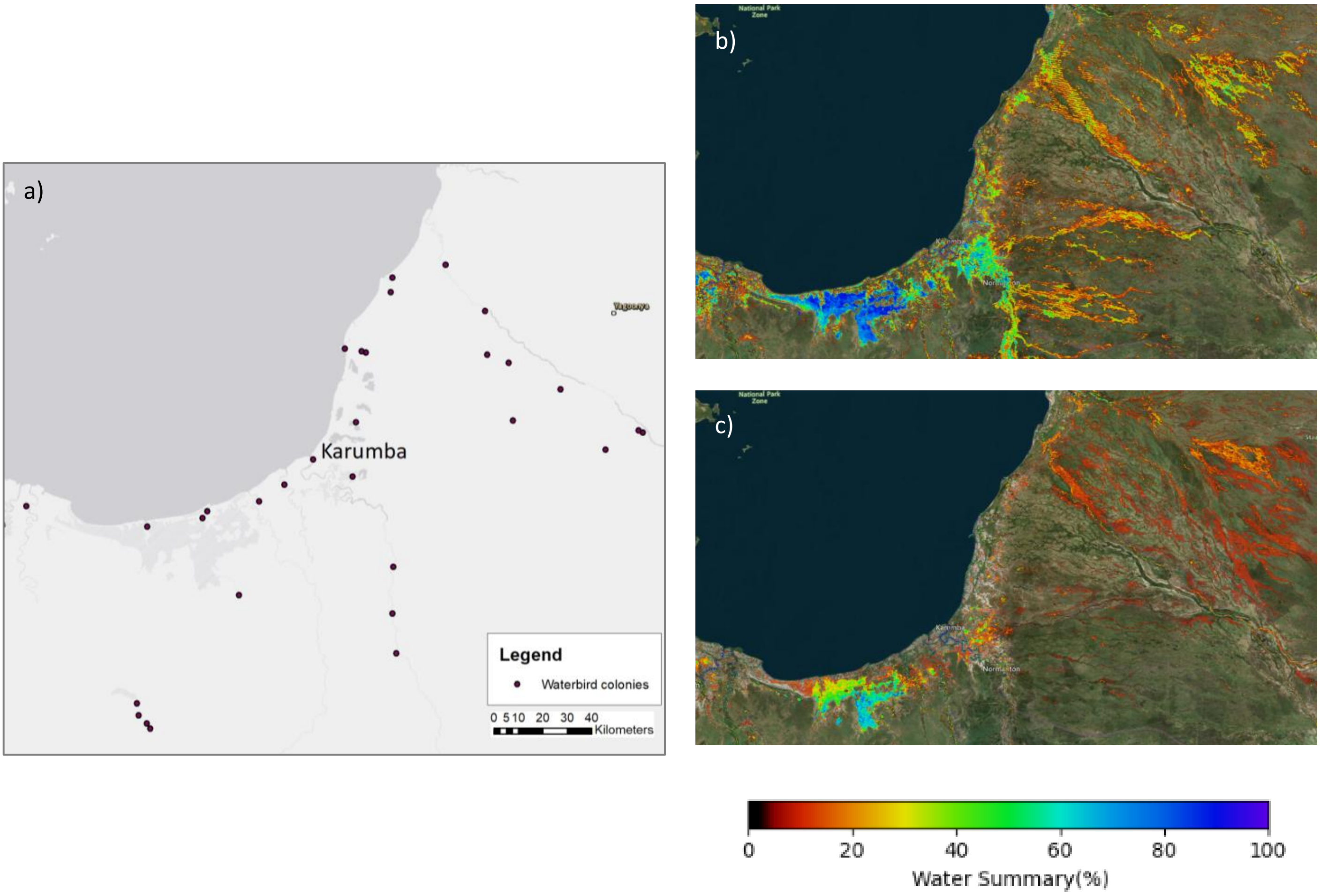


Figure 12. a) Sites of waterbird breeding colonies on Queensland's southern Gulf Plains (40) and Water Summary: what percentage of clear observations were detected as wet (i.e. the ratio of wet to clear as a percentage) for the southern Gulf Plains as derived Geoscience Australia Water Observations, Seasonal Frequency Statistics, November to March (Landsat, Collection 3, 30 m, WO-STATS-NOV-MAR, 3.1.6 for b) 2011/12 and c) 2015/16.

The poor wet season in 2015-16 also resulted in the second poorest breeding participation in Magpie Geese across coastal and sub-coastal floodplains in the Northern Territory survey's 33-year history (64). The East Australian Waterbird Survey reported its lowest ever wetland area index score and the second lowest waterbird abundance on record (65).

Although the timescale may not be relevant to the 2022 outbreak, the 2015-16 event will have likely provided incentives for mass emigration of waterbirds from Australia to the Trans Fly[7] and fostered favourable conditions for maintenance and amplification in populations of transient and resident waterbirds.

[7] Many birds will have also retreated to coastal fringes and other drought refuges in Australia.

February 2017 – February 2020

Conditions across much of the southern Murray Darling Basin were wet throughout late 2016 and early 2017 and waterbirds responded with large scale breeding of ibis, egrets and pelicans in the Macquarie Marshes and Lowbidgee (66). However, on a broader scale, five years of consecutive warm winters from 2015 resulted in overall dry conditions throughout eastern and south-eastern Australia. Three years of positive Indian Ocean Dipole events 2017-20 resulted in exceptional, widespread heat and drought in eastern and north-eastern Australia from February 2017 to February 2020.

Late 2018 to February 2020 El Niño

Two-years of consecutive positive Indian Ocean Dipole events in conjunction with the consecutive Central Pacific El Niño, resulted in cumulative depletion of water in the landscape and culminated in the driest spring on record across Australia (in 2019) and ultimately the 2019-20 megafires. Southern Australia registered its lowest year-to-date rainfall for the same period and delayed or attenuated wet seasons were experienced in the tropical north (Figure 13). Combined with temperatures well above average throughout the year, the Murray Darling Basin experienced its worst three-year drought in over 120 years and total storage levels in the northern Murray–Darling Basin plummeted to below 6% in early 2020 (BOM).

Although COVID-19 restrictions precluded several ongoing monitoring programs from being effectively implemented in the spring/summer of 2019-20, aerial surveys and remote research (satellite tracking (18)) provided some insights into wetland and waterbird responses.

Far North

A third consecutive failed wet season for the Far North resulted in another year of poor breeding participation for Magpie Geese (67) and overall waterbird abundance on the Northern Territory floodplains was low.

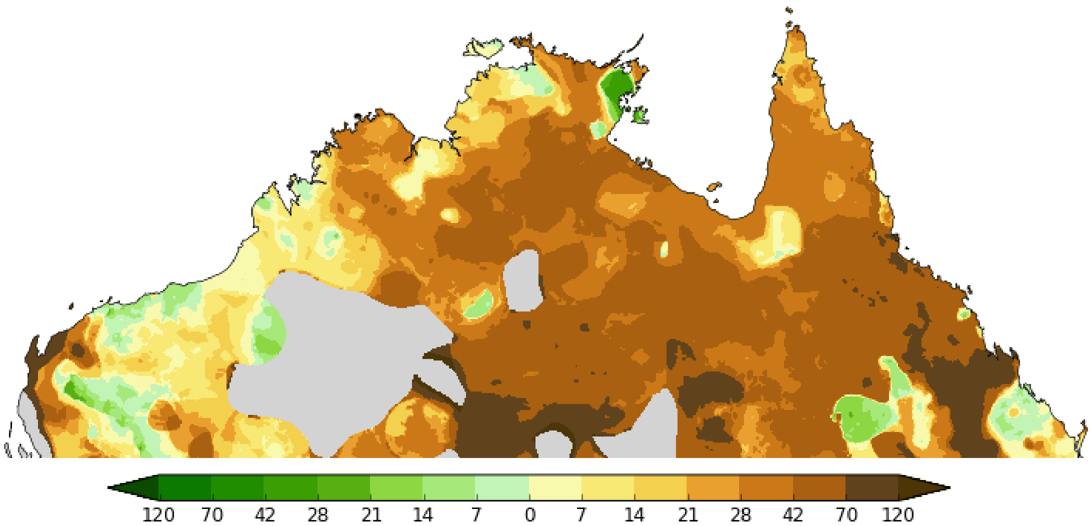


Figure 13. Number of days earlier or later than the long-term average onset of the wet season for summer 2019. (BOM)

Trans Fly region

Similar effects were observed in New Guinea with a late onset of the wet season leaving large areas south of the Star Mountains and Fly River catchment experiencing accumulated low rainfall deficits. These conditions may have been adequately severe for the (now altered) hydrology of the Middle and lower Fly River floodplain to draw down, providing the large expanses of shallow water required for wading and grazing birds to exploit.

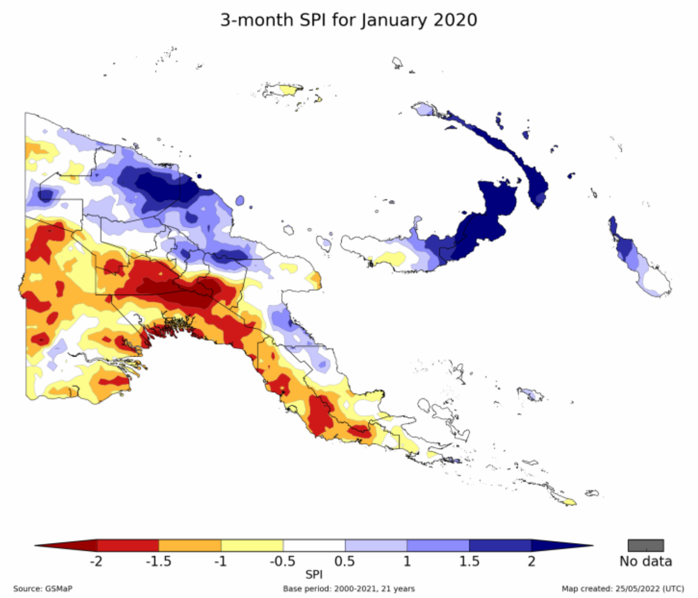


Figure 14. 3 month Standard Precipitation Index for Papua New Guinea at the height of the 2019 El Nino. Generated by Z-W Chau, BOM.

Murray Darling Basin

Exceptionally dry conditions and rainfall deficiencies further reduced inflows throughout the Murray Darling Basin. The Eastern Australian Waterbird Survey's wetland area index was calculated to be the lowest on record surpassing the previous record low 2015-16. For the third year in a row breeding and overall waterbird abundance was well below average. Breeding was recorded at only five very small sites in the entire Murray Darling Basin with only one external (coastal) site in Western Port Bay (Vic) contributing large participation. Straw-necked Ibis and Black Swan made up 97% of the breeding birds recorded in the survey many of which were confined to the Victorian coastline.

As with the 2015-16 summer the prevailing dry and hot conditions accompanied by failed wet season of 2019 may have provided incentive for birds that normally utilise eastern and northern floodplains for summer breeding, to seek alternatives in New Guinea's Trans Fly.

February 2020 – January 2022

Flooding affected much of eastern Australia in February and March 2020 AND BY September is was recognised that a La Niña event was taking place. The 2020-21 event increased the likelihood of rain-bearing weather patterns over eastern Australia and reduced evaporation due to increased cloudiness and reduced temperatures, replenishing soil moisture. A positive Southern Annular Mode (SAM) reinforced the impacts of La Niña and triggered above-average rainfall for most of NSW. Extreme rainfall caused flooding in eastern and central Australia in March 2021 and was followed by continued wet periods on the NSW Southern Slopes to October.

November 2021 was Australia's wettest on record and the coolest since 1999. Total storage levels in the northern Murray–Darling Basin were 90.9% of capacity at the end of the month, compared with just 24.5% at the end of November 2020.

Trans Fly

Wet conditions were persistent in the New Guinean Highlands and Western Provence, recharging the floodplains of the Fly River and significantly reducing habitat suitable for wading and grazing waterbirds (Figure 15).

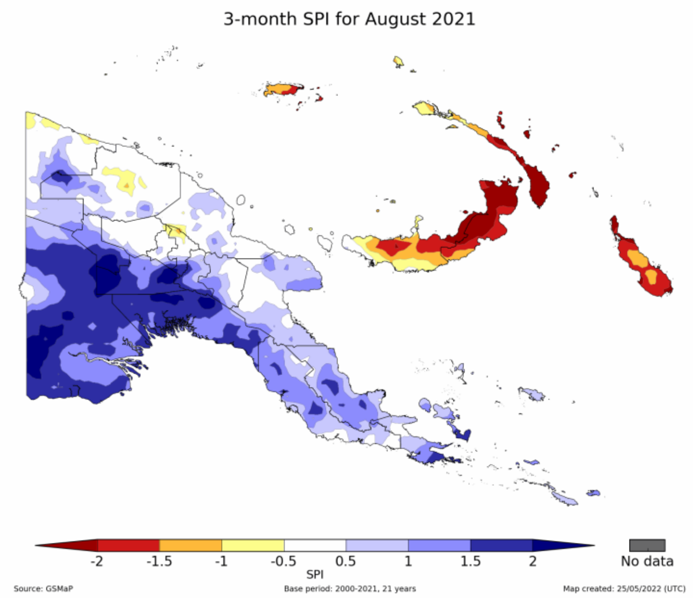


Figure 15. Standard Precipitation Index for Papua New Guinea during the 2021 La Nina event. Generated by Z-W Chau, BOM.

Murray Darling Basin

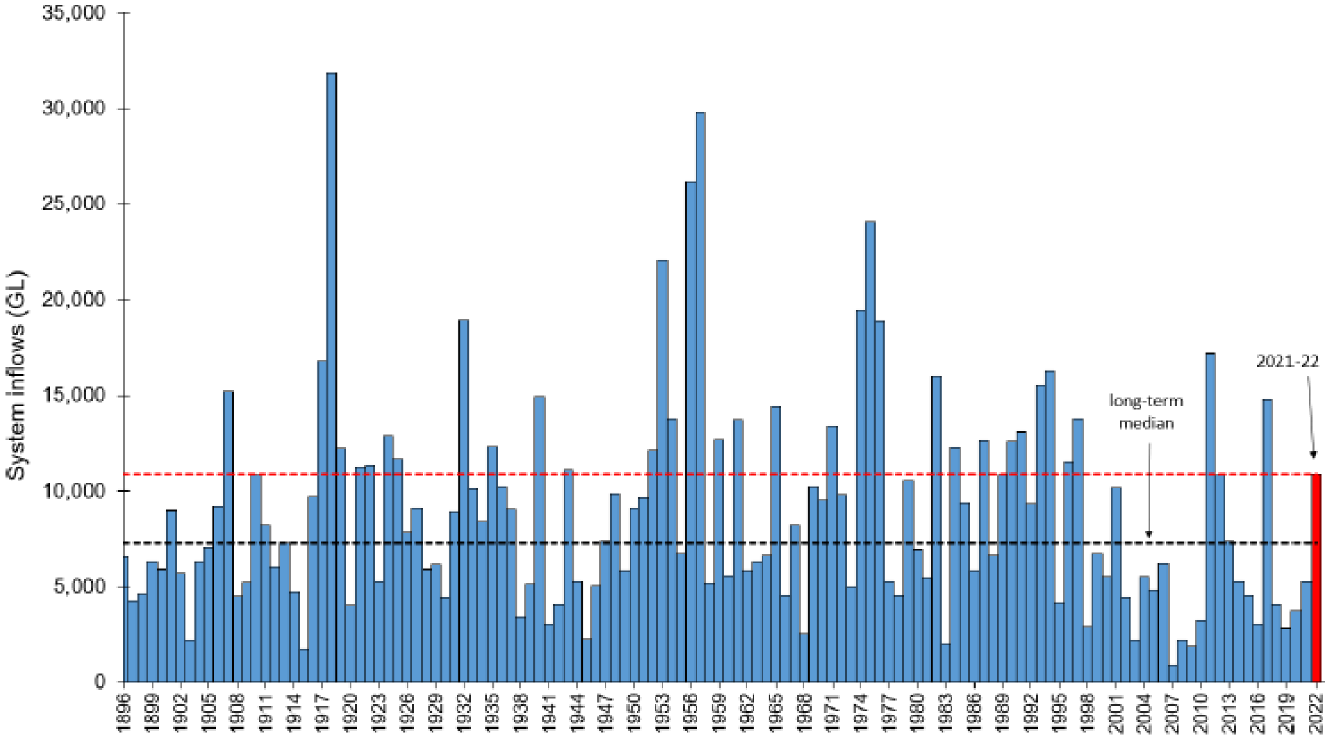
In spring 2020, average rainfall complemented with the targeted use of 200,000 megalitres of NSW and Commonwealth water along the Murrumbidgee River, triggered waterbird breeding. More than 15,000 hectares of wetlands in the Gayini system were inundated by the water release, as well as a similar area in the adjoining Yanga National Park, reaching as far as Yanga Lake in the lower Murrumbidgee floodplain system near Balranald. Over 40,000 breeding waterbirds were recorded in the Lowbidgee Floodplain representing the largest colonial waterbird breeding event in the Basin.

In total, 17 active waterbird colonies in the Lowbidgee, and three active colonies in the mid-Murrumbidgee wetlands were detected from Spring 2020 to late Summer 2021.

The following summer exceeded the 2020-21 summer in wetland extent, waterbird abundance and number and size of waterbird colonies. The River Murray System inflows during the 2021-22 water year (excluding releases from Snowy Hydro, Inter Valley Trade deliveries, managed environmental deliveries from tributaries and inflows to the Menindee Lakes) were approximately 10,870 GL. This inflow volume is around double that recorded for the same period in 2020-21 and around 3,550 GL more than the long-term median inflow volume.

Across the basin the largest colonies were recorded responding to the incredibly wet conditions of November 2021. This included a large colony of 116,500 ibis nests in the Macquarie Marshes and 36,000 nests in the Booligal wetlands. Also, 35,000 ibis nests and 7,000 pelican nests were recorded in the Lowbidgee wetlands and 30,000 pelican nests in Lake Brewster. These colonies also supported high densities of herons, egrets and cormorants, however data summaries were not available at the time of writing this report.

Figure 16. River Murray system inflows—water year totals (to end May) since 1896 (excludes Snowy Hydro inflows, IVT delivery, managed environmental inflows and inflows to Menindee Lakes). Black dashed line shows the long-term median. The red dashed line shows the 2021-22 inflows compared with all years on record since 1896. (BOM)



Several smaller colonies were reported throughout the Murray Darling basin and high-levels of breeding participation were recorded at semi-permanent colonies in coastal areas. Some colonies established in early November were still active in April and May, with some species like Pelican and Black Swan attempting multiple clutches (Sullivan pers com). One multispecies colony in Currawinya National Park's Lake Wyara (Southern Queensland) only established in May and supported up to 15,000 breeding birds.

The profuse flooding and persistent rains which triggered exuberant breeding throughout eastern Australia in 2021/22 provided complex interconnected wetland mosaics which were exploited by high densities of waterbirds. The pulse of resources attracted birds from coastal and near coastal wetlands from South Australia to north Queensland (68). Longitudinal monitoring of coastal lagoon systems north of Townsville reported mass emigration of Pied Heron, Glossy Ibis, Spoonbill and Great, Intermediate and Little Egret which are yet to return (Kennedy unpub). The value and extent of dry season habitat in the south was theoretically also an attractive incentive for dry season migrants or nomads from the Far North and New

Guinea. These movement likely facilitated widescale, frequent interaction between birds carrying virus and those susceptible to infection.

It is significant to note that large scale floods in 1908, 1956, 1975 and 2011 (Figure 16) were suspected to be the triggers for four large outbreaks of Murray Valley Encephalitis across Australia. MEV is an endemic flavivirus that is believed to have originated in Australia's north west and suspected to persist in small pockets of the Far North. Like JEV, Ciconiiformes are thought to play a major role in MEV's natural cycles and to be largely responsible for the wide scale distribution of the virus observed in the major outbreaks mentioned above (8, 57, 69).

Useful waterbird data, gaps, and recommendations

Australia has many large waterbird and wetland datasets which could be useful in modelling climate driven scenarios of JEV spread by waterbirds and several other One Health applications (Appendix 3). However, many of these data exist in siloed databases, and are not dynamically accessible (70). Furthermore, significant temporal, spatial and research gaps preclude our ability to effectively identify waterbird based epidemiological pathways, episodic triggers and ultimately mitigation measures. These gaps are particularly evident for Australia's Far North and Torres Strait Islands and for New Guineas Trans Fly Region[8].

The East Australian Waterbird Survey (EAWS) represents one of Australia's most valuable biodiversity datasets and, in recent years, has been supplemented by a program of research investigating the efficacy of environmental water releases for waterbird recruitment. However, even in the Murray Darling Basin there are gaps in spatial and temporal coverage as surveys do not (and cannot) always occur at the time of year that peak breeding occurs and gaps between flight transects omit significant sites. For example, aerial surveys conducted in spring (including the EAWS and several game duck surveys), can miss breeding that is triggered later in the year and extends into summer. The large- scale breeding of ibis, egrets and pelicans in 2016 to 2017 in the Macquarie Marshes and Lowbidgee was not captured by aerial surveys (71), nor was the large breeding event of ibis at Narran Lakes in 2008 and 2021 (Brandis pers com). Indeed large, episodic colonial waterbird breeding events of the Channel Country and other arid and northern regions typically peak in the months from summer to autumn and are not routinely documented in a systematic fashion.

A recent review of Australian waterbird data demonstrated that significant research (methods and results) are routinely confined by jurisdictional boundaries and preclude effective development of landscape aware wetland and waterbird population management (70, 72). The review, which consolidated over 4 million waterbird records from over 25 databases, demonstrated the utility of broad-scale systematic monitoring but also where gaps in monitoring should be filled in order to track our understanding of how waterbird populations function and can be maintained in Australia.

[8] Capacity o investigate these regions, which are mostly remote from transport infrastructure, is severely limited and access to several regions, especially those outside the Australian mainland, will require complex, culturally sensitive collaborations with traditional owners and governing bodies.

Implementation of the following recommended activities is proposed by BirdLife Australia and waterbird experts who were consulted to address these knowledge gaps:

1. Serological sampling
    1.1. Sampling of potential host species, targeting large waterbird colonies, throughout Australia.

2. Monitoring
    2.1. Regular aerial waterbird colony surveys of the Gulf Plains and western Cape York peninsula (replicating work reported by Jaensch & Richardson 2013 & Taplin 1990).
    2.2. Regular waterbird colony surveys of the Far North of Northern Territory (replicating Chatto 2000).
    2.3. Regular waterbird surveys of the Middle Fly region of PNG (Replicating Halse et al. 1995)
    2.4. Exploratory surveys of floodplain, forested freshwater swamps and intertidal wetlands of southern New Guinea.
    2.5. Continued annual standardised aerial surveys of the Magpie Goose populations across the Northern Territory Range.
    2.6. Continued annual standardised surveys of large colonies in the Murray Darling Basin.
    2.7. Regular surveys of large colonies in the Channel Country, Lake Eyre Basin, when activated.

3. Data management
    3.1. BirdLife Australia to develop its National Waterbird Monitoring Program and associated database which seeks to facilitate wide scale dynamic and referenceable data-sharing and improve context aware coverage of wetlands through standardised monitoring.
    3.2. University of NSW to develop the National Colonial Waterbird Breeding Database.

4. Tracking
    4.1. Satellite tracking of Cattle, Little, Intermediate and Great Egret, Pied Heron, Nankeen Night-Heron and Pelican from colonies in the Murray Darling Basin.
    4.2. Tracking of Cattle, Little, Intermediate and Great Egret, Pied Heron, Nankeen Night-Heron, Royal Spoonbill and Australian White Ibis from heronries in the Far North.
    4.3. Tracking of Wandering Whistling-Duck and Magpie Goose from the western Cape York Peninsula and Torres Strait Islands
    4.4. Tracking of Cattle, Little, Intermediate and Great Egret, Pied Heron and Nankeen Night-Heron from populations in the Middle Fly.

5. Gene flow studies
    5.1. Gene flow studies targeting potential host species across several regions.
    5.2. Further genetic research using Feather Map samples

6. Maintain a waterbird ecology working group associated with Wildlife Health Australia to inform health responses to viruses associated with waterbird reservoirs.

These activities will greatly contribute to knowledge of waterbird movements, population structures and dynamics and natal site fidelity and will ultimately help inform epidemiological models of host-disease dynamics in the waterbird hosts and the mechanisms causing maintenance and amplification of arboviruses in Australia.

In addition to One Health outcomes the activities listed above will contribute significantly to conservation strategies seeking to address ongoing long-term declines in the majority of Australia's wetland dependant birds (70).

Acknowledgements

Many thanks to Stuart Halse, Andrew Storey, Kate Brandis, Jennifer Spencer, David Roshier, Peter Driscol, Tim Clancy, Roger Jaensch, Keith Ward, Taej Mundkur, Simon Kennedy, Deb Sullivan and Heather McGinness for providing additional information on waterbird ecology and research and to Andrew Watkins and Zhi-Weng Chau for generating climate related simulations for Papua New Guinea. Many thanks to Tiggy Grillo, Wildlife Health Australia, for support and advice.

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Appendix 1 – Additional expert considerations in response to the original synthesis

Prepared by Tiggy Grillo (Wildlife Health Australia)

Background:

Noting this report was undertaken as a RAPID SYNTHESIS and presents ***possible*** pathways for introduction and spread within the Australian context, it was recognised that there may be additional plausible avian pathways that have not been captured. Therefore, additional input from experts was sought during a 2nd expert workshop with national and regional waterbird ecologists in October 2022 and is presented below.

During the 2nd expert workshop, feedback sought from waterbird and JEV experts on the following themes:

- Additional avian entry / incursion pathways
- Additional avian dispersal / spread pathways
- Additional avian species worth noting
- Recommendations

Overarching information:

- In 1995 and 1998, five human JEV cases were genotype II from Badu Island in the Torres Strait of northern Queensland
    a. 1995 three human cases, two deaths
    b. 1998 more geographically widespread outbreak, one human case on Badu and one One human case from Mitchell River along the western Cape York peninsula.
- JEV subsequently identified in the Western Province of Papua New Guinea.
- Between 1995 and 2005, a sentinel pig program detected JEV activity every year (except 1999) in the Torres Strait, with genotype 1 viruses from the year 2000 onwards.
- February 2004 - serological detection in sentinel pigs in the Northern Peninsula Area (NPA) on the tip of Cape York Peninsula. Virus detected through subsequent mosquito surveillance in five communities that constitute the NPA.
- November 2020 - Serological evidence in animals (feral pigs, sentinel cattle) in NT via retrospective testing of archived samples. Note: serology suggestive of infection with JEV due to the potential for cross reactive antibodies to endemic flaviviruses.
- April 2021 – genotype IV detection in archived domestic pig from north Qld tested in 2022.
- Between 1999 and 2021 – No endemic human cases in Australia.
- February 2021 - a fatal human case associated with exposure in the Tiwi Islands in the NT sequenced as genotype IV.
- May 2021 - Second human case likely infected just south of Adelaide River town south of Darwin, NT.

- 2022 – Sequences from all pig and human cases in Australia (that were sequenced) are genotype IV.
- Genotype IV has predominantly been reported from Indonesia notably including Bali, one possible isolate from PNG. Timor Leste JEV detections unknown.

Note: multiple avian pathways could possibly occur in parallel.

1. Additional avian entry / incursion pathways

- JEV entry could have been anywhere across northern Australia from Kimberley, WA to Torres/Cape, QLD.
- Possible entry / incursion pathways beyond a Papua New Guinea (PNG) to Torres Straight/Cape York pathway theory presented in the report discussed included entry via the Top End, NT.
    a. Why - Sequencing to date has shown early isolates in NT and all subsequent pig and human JEV detections in Australia to be genotype IV (which is predominantly found in Indonesia, although one possible recent detection of genotype IV in PNG.
    b. How
        i. JEV infected bird, noting
            - Few birds travel the Arafura straight/sea in comparison to the frequent north/south movement of waterbirds across the Torres Strait, which mainly occurs from April and September.
            - No reports of any bird species regularly crossing Arafura straight in the wet season (November to April).
            - Monsoon-related winds are mostly from the north-west. Example: a Javan Pond-Heron was observed in Darwin (from Indonesia) after a north-west blow presents evidence movement across the Arafura straight possible.
            - Geering et al's 1998 suggests some of the potential reservoir hosts, such as egrets, are capable of long distance spread outside of Australia (Geering D.J. et al (1998). Movement patterns of Great, Intermediate and Little Egrets from Australian breeding colonies. Corella 22: 37-46 (absa.asn.au)) Note: doesn't cover Top End bird movements.
        ii. Wind-blown JEV infected mosquitoes
        iii. Other potential entry is by JEV infected exotic mosquitoes escaping from a plane or fishing boat/other vessel.
            - In the NT, the CDC has reported "airport dengue" presumably from a dengue infected *Ae aegypti* flying off a plane at Darwin airport. Airport has a lot of incoming commercial and military planes land from Bali/Indonesia etc.
            - NT medical entomology team have recently found some NT dispersal, presumably out of Darwin, of the exotic to Australia and major JEV mosquito vector in SEA, *Culex tritaeniorhynchus*.

2. Additional avian dispersal / spread pathways

- The rapid synthesis provides comprehensive bird analysis that presents a likely picture of widespread dispersal down the east coast of Australia, explained by the weather (specifically La Nina) and bird survey and movement data presented (noting the major gaps in knowledge flagged in the report).
- Bird movements out of (and into) the NT was further discussed:

a. If entry was via Torres Straight/Cape York pathway, then the cases in the Top End of the NT in 2021 need explanation.
    i. No / limited evidence of substantial movement of waterbirds around the bottom of the Gulf of Carpentaria that would link that Torres Straight flyway to the Tiwi outbreak within an appropriate timeframe. (note additional species information below)
    ii. Possible underlying situation - JEV is endemic in some tropical areas of Australia, persisting in low densities, awaiting conditions conducive to amplification and onward transmission and dispersal, possibly via a waterbird and feral pig cycles, with occasional dead-end asymptomatic spillovers into human populations. (early serological detections provide some evidence here – noting need for careful interpretation)
b. If entry was via the Top End / Tiwi, NT, then movement of JEV from NT to Murray Darling Basin (MDB) needs explanation.
    i. Genotyping of JEV isolates and waterbird movements support this (see below)

3. Additional avian species worth noting

- Bird movement.
    a. Straw-necked Ibis - Adult and immature Straw-necked Ibis regularly visit the Top End, including the Tiwi Islands, during the dry season, but migrate back to the MDB to breed.
    b. Some studies have also shown Straw-necked Ibis and Glossy Ibis breed (at times) in the Barkly wetlands (Barkly Tablelands, NT; up to 5500 pairs of Straw-necks in 1995, breeding generally under-recorded due to vastness of the wetlands (for example see Important Wetlands directory – Tarrabool Lake, NT) and more frequently in the Channel Country, Qld/SA (many colonies from 10s – 100s of pairs with some > 15-20K pairs). During some of the severe drought years of the past two decades, the Channel Country colonies may have been the only source of recruitment. (Jaensch 2009: Map 3, Table 2).
    c. From the GPS satellite-track and sample waterbirds from MDB breeding sites, a handful have been tracked flying from the MDB to the Top End and a few have been tracked *returning* from the Top End (note: network coverage, transmitter or harness issues may have limited numbers tracked; McGinness pers comms)
    d. White ibis are another possibility, but there are many resident birds.
    e. Sarus Cranes - evidence from feathers that the same individual bird has been sampled at both ends of the migration (Brandis et al 2020; Nevard et al 2020).
    f. See Feather Map below from Brandis et al 2020 for other species.

4. Additional recommendations
    a. Analysis to assist with the JEV entry question (outside scope of this report):
        i. Whole genome sequencing of JEV (if) recovered from feral pigs from across the top of Australia (e.g. WA, NT, QLD) and comparison with the 2022 genomes from southern States may provide some resolution on timelines and origins.
        ii. Genetic relatedness of the mosquitos in northern Australia and our neighbouring countries.
    b. Analysis to assist both JEV entry and dispersal (within scope of this report):
        i. bird tracking studies or genetic relatedness studies of the ardeid (and other possible reservoir hosts)
        ii. analysis of intersecting the movements and abundance of waterbirds with JEV outbreaks

iii. Disease surveillance in a range of wild bird species at breeding sites.

Figure from Brandis KJD et al 2020.

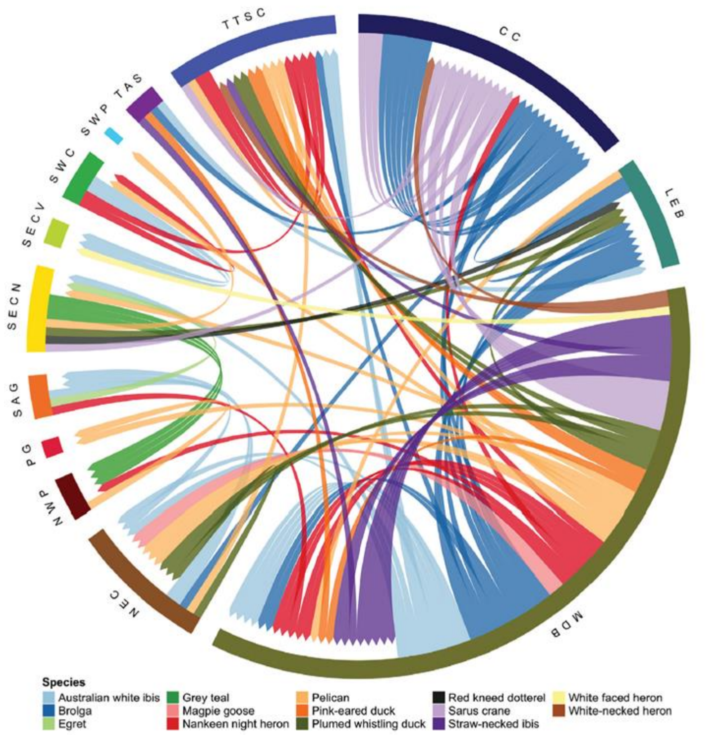

FIGURE 2 Chord diagram highlighting modeled movement of nomadic waterbird species among river basins in Australia. Origins of feathers predicted from elemental signatures are displayed on the outer ring and location where feathers were found are represented by arrows. Each arrow represents a unique feather (see Table S4). River basins are as follows: CC, Carpentaria Coast; LEB, Lake Eyre Basin; MDB, Murray-Darling Basin; NEC, North East Coast; NWP, North Western Plateau; PG, Pilbara-Gascoyne; SAG, South Australian Gulf; SECN, South East Coast – New South Wales; SECV, South East Coast – Victoria; SWC, South West Coast; SWP, South Western Plateau; TAS, Tasmania; TTSC, Tanami-Timor Sea Coast

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Appendix 2 -Distributions of potential host species

Solid areas comprise known species distributions. Hashed areas comprise potential distribution. Maps generated using composite data from BirdLife International, BirdLife Australia and Wetlands International. *Distributions represent current estimated ranges at 2020 and may not identify areas of low reporting rates or recent records.*

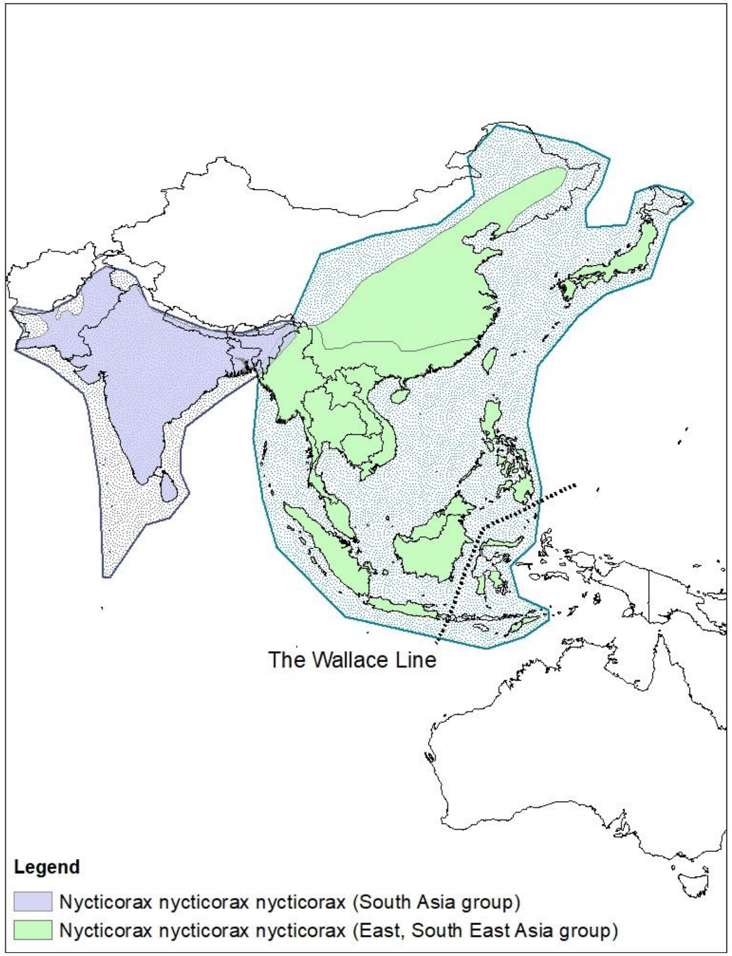

Figure 17. Species distribution of Black-capped Night-Heron .

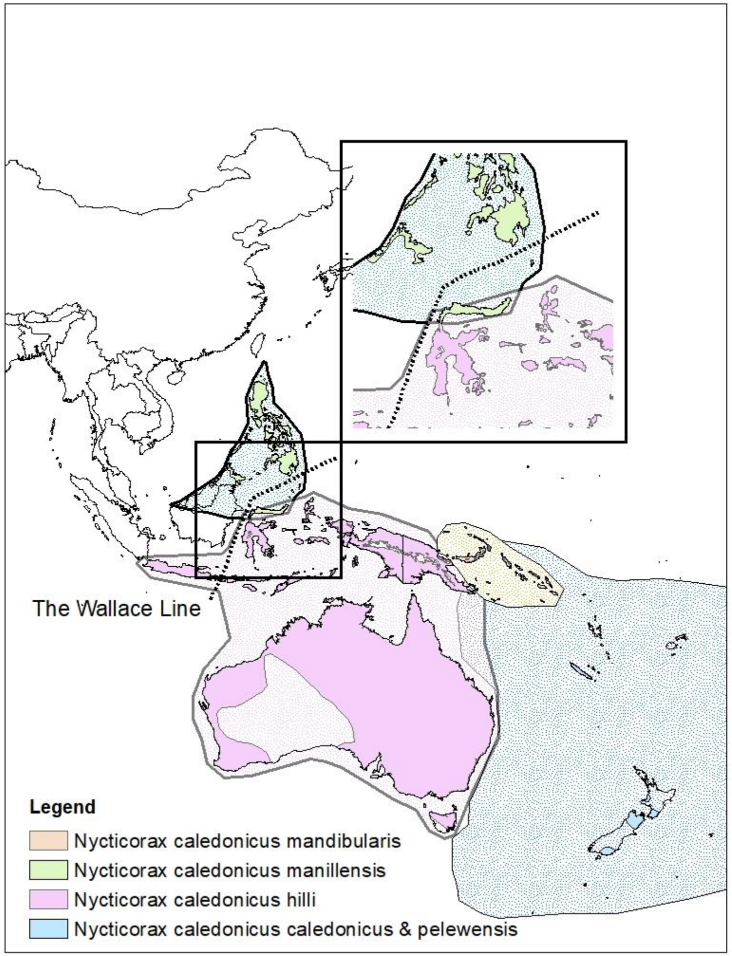

Figure 18. Species distribution of Nankeen Night-Heron.

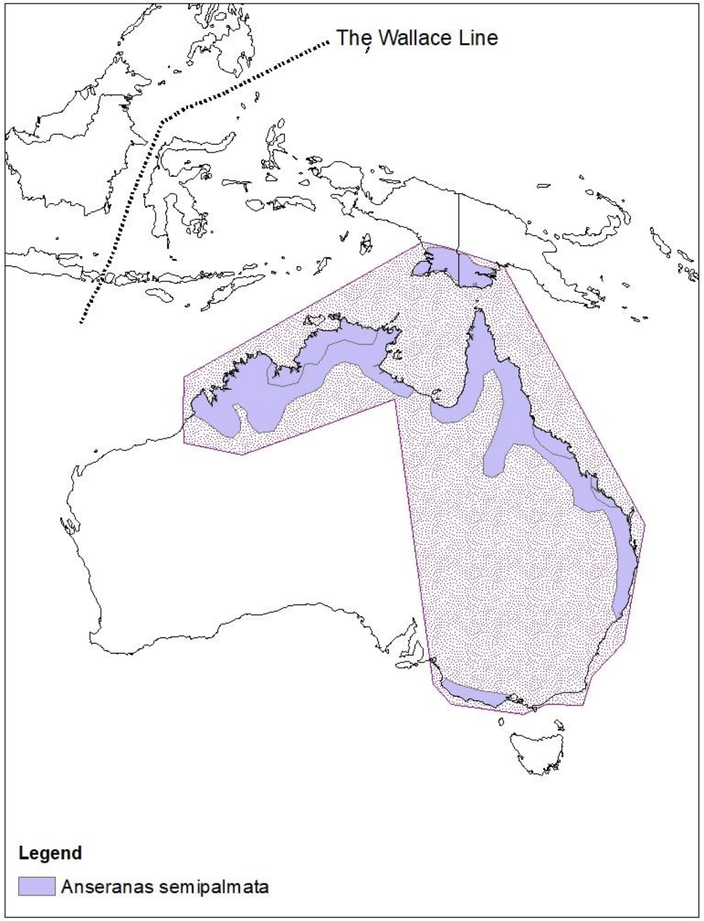

Figure 19. Species distribution of Magpie Goose.

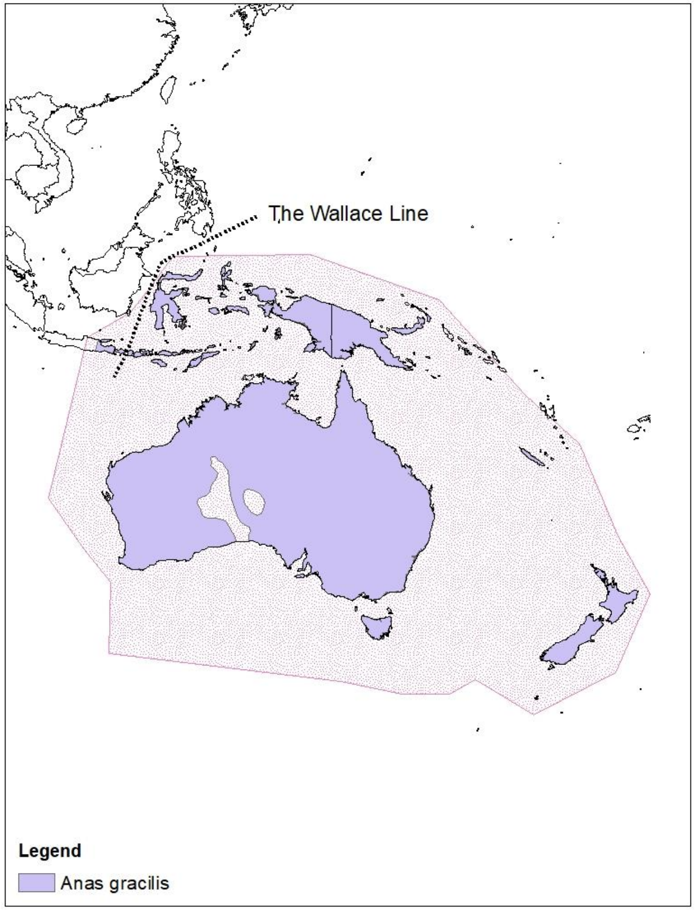

Figure 20. Species distribution of Grey Teal

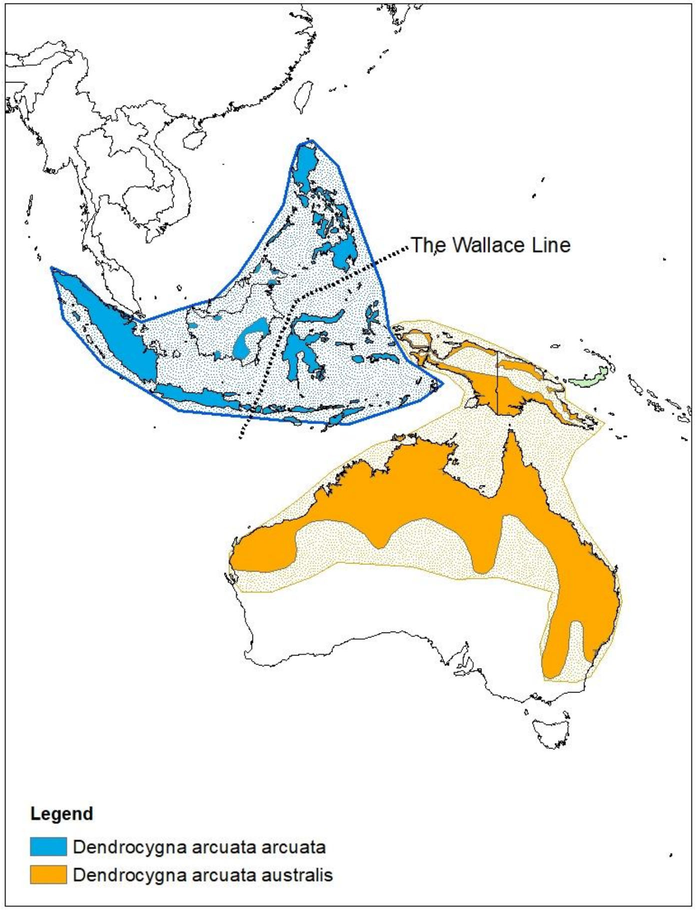

Figure 21. Species distribution of Wandering Whistling-duck.

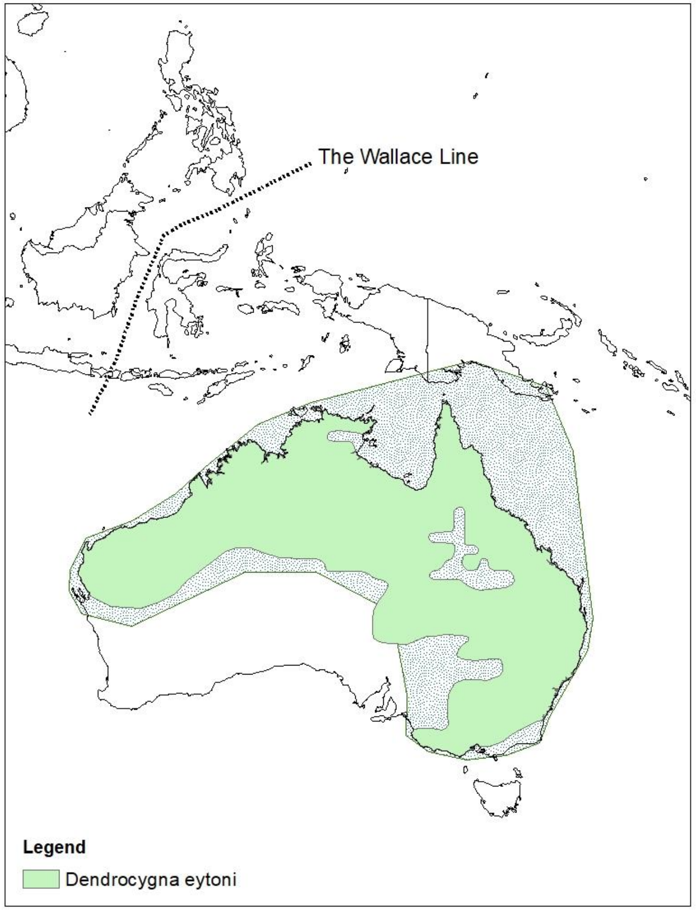

Figure 22. Species distribution of Plumed Whistling-duck

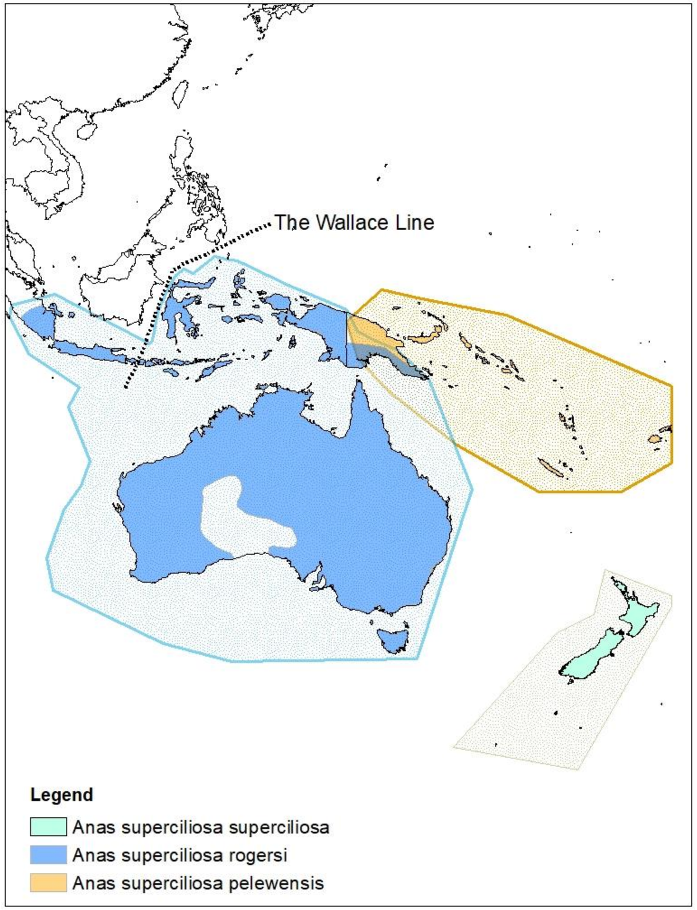

Figure 23. Species distribution of Pacific Black duck.

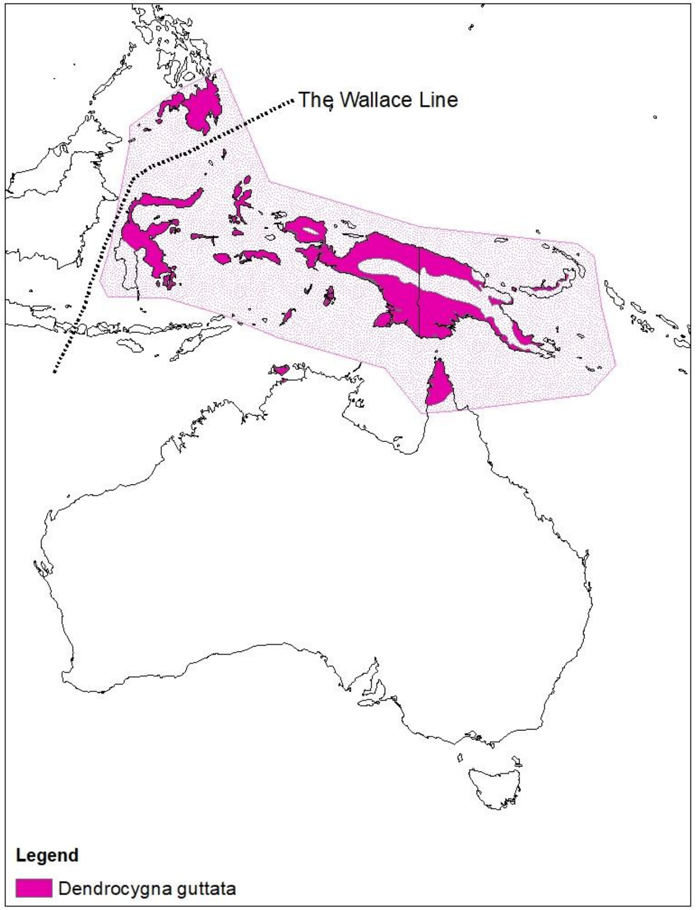

Figure 24. Species distribution of Spotted Whistling-duck.

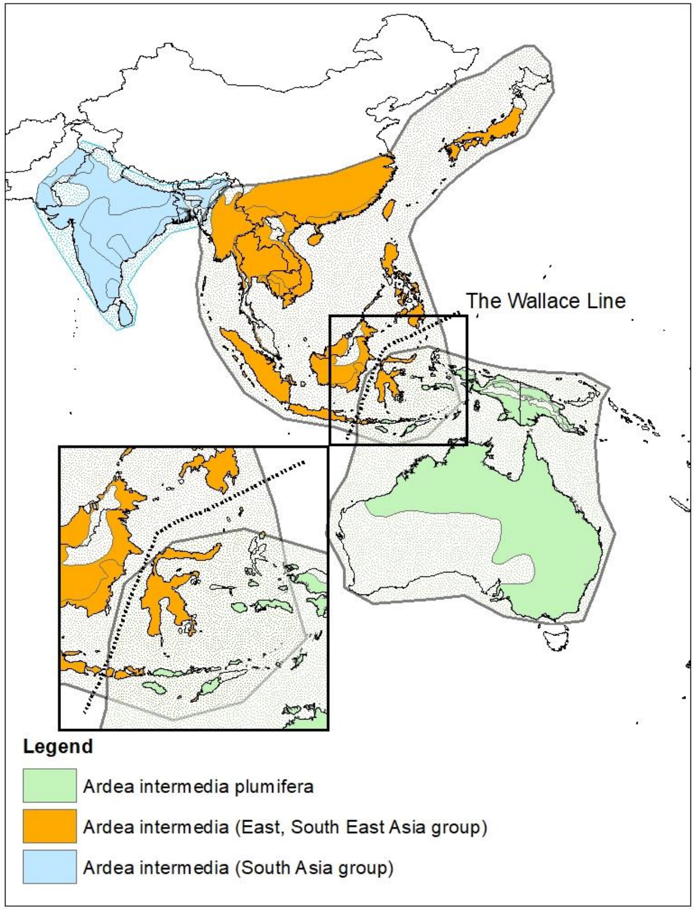

Figure 25. Species distribution of Intermediate Egret.

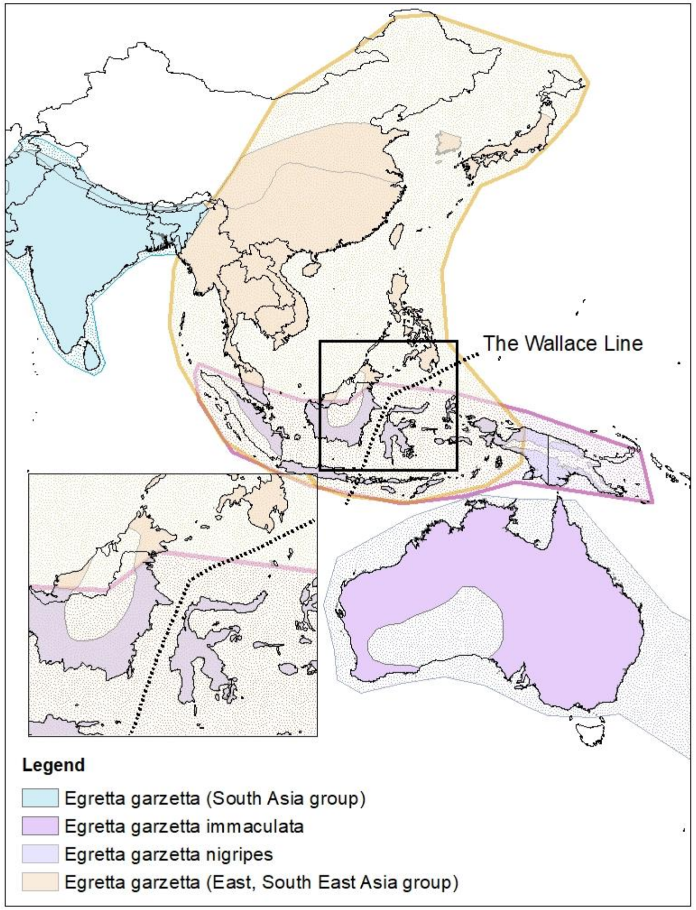

Figure 26. Species distribution of Little Egret.

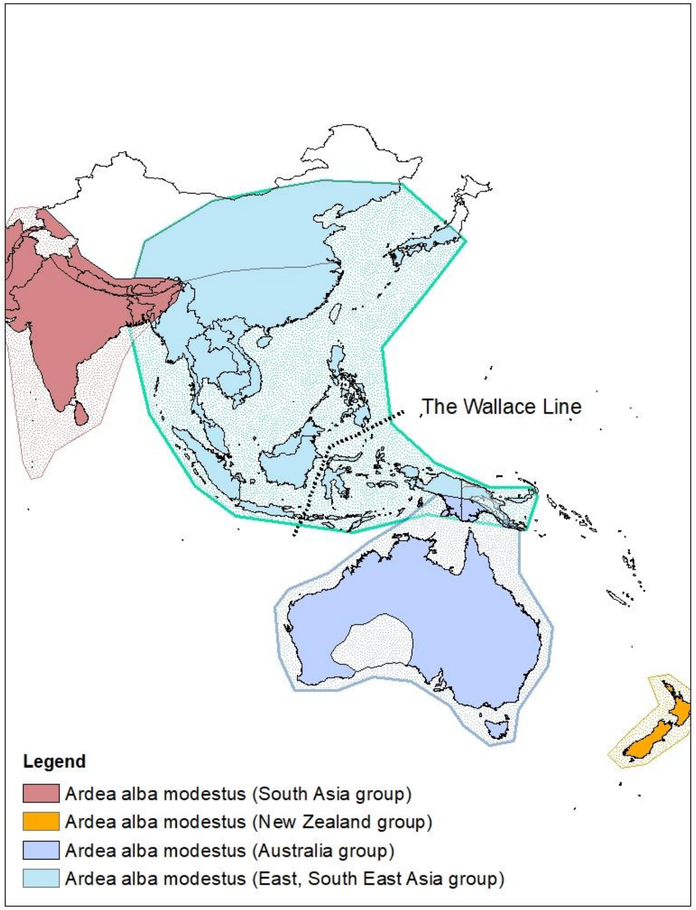

Figure 27. Species distribution of Great Egret.

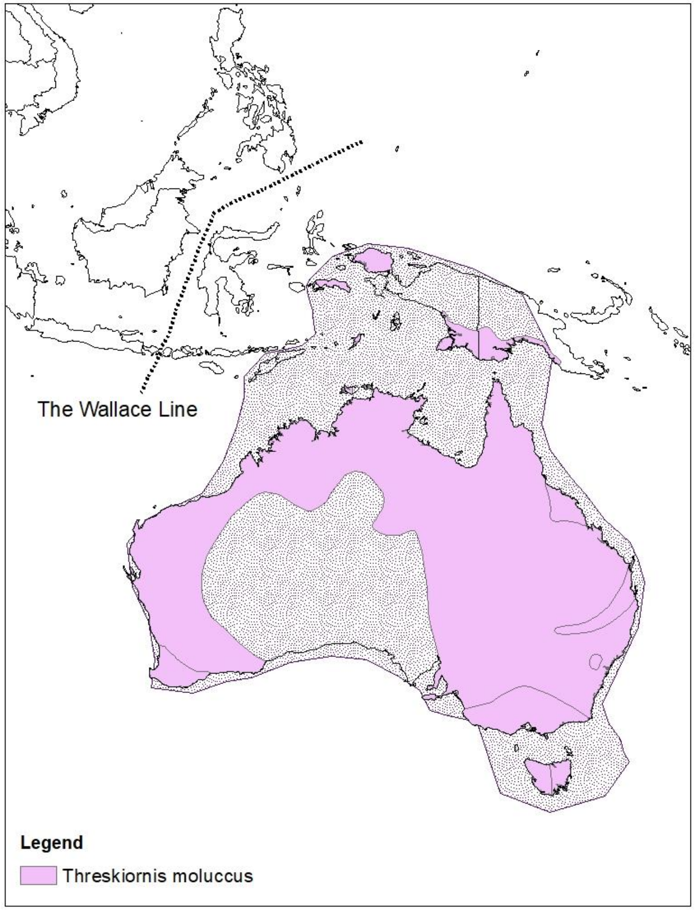

Figure 28. Species distribution of Australian White Ibis.

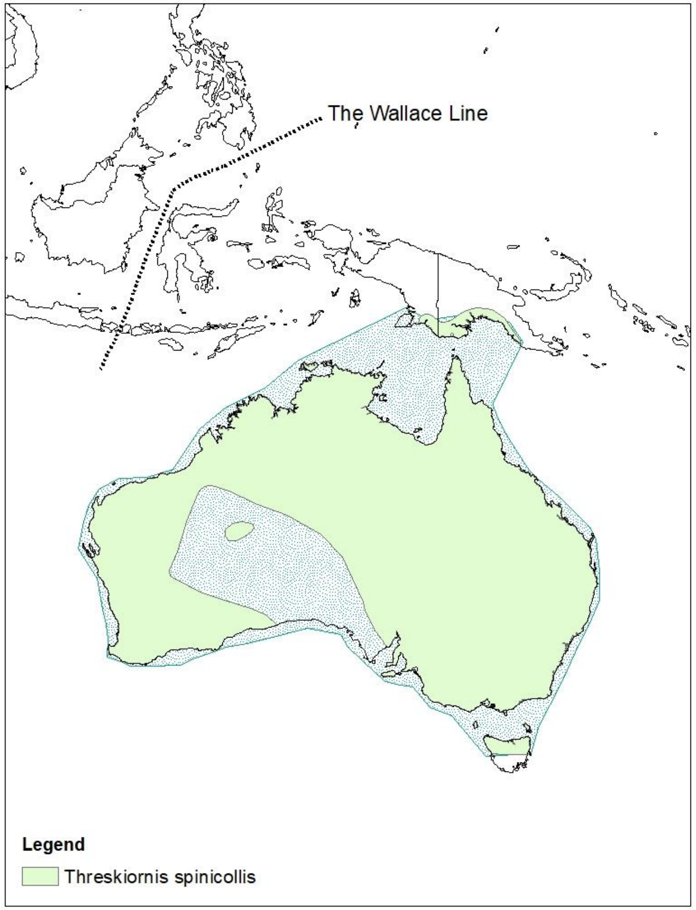

Figure 29. Species distribution of Straw-necked Ibis.

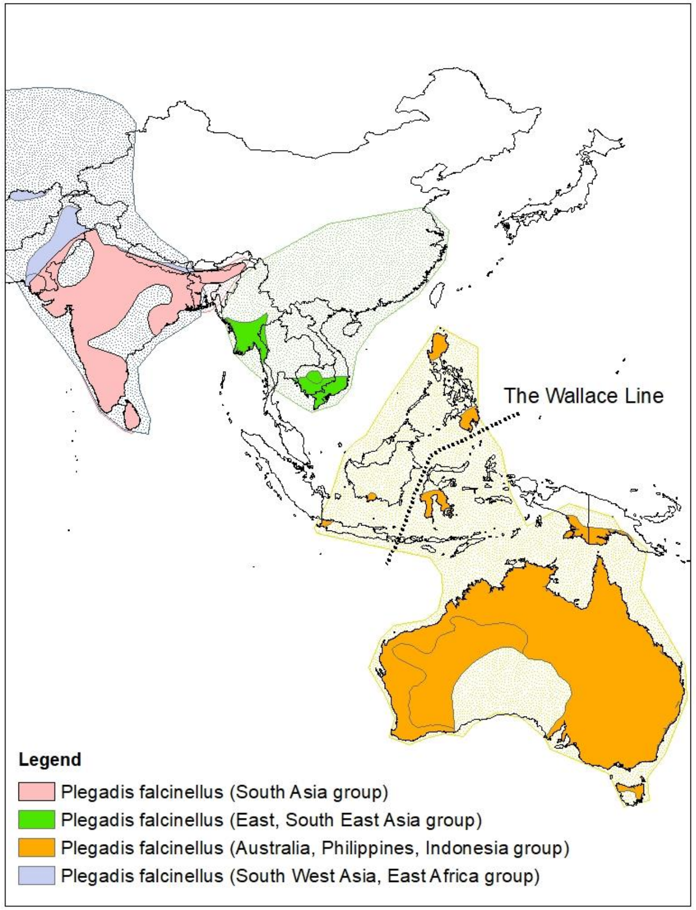

Figure 30.Species distribution of Glossy Ibis.

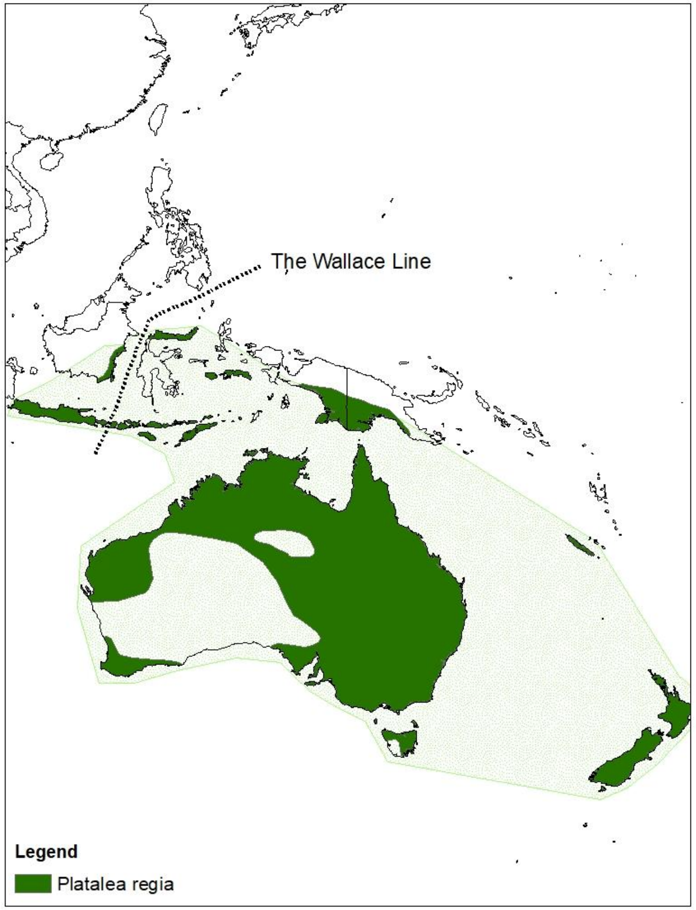

Figure 31. Species distribution of Royal Spoonbill.

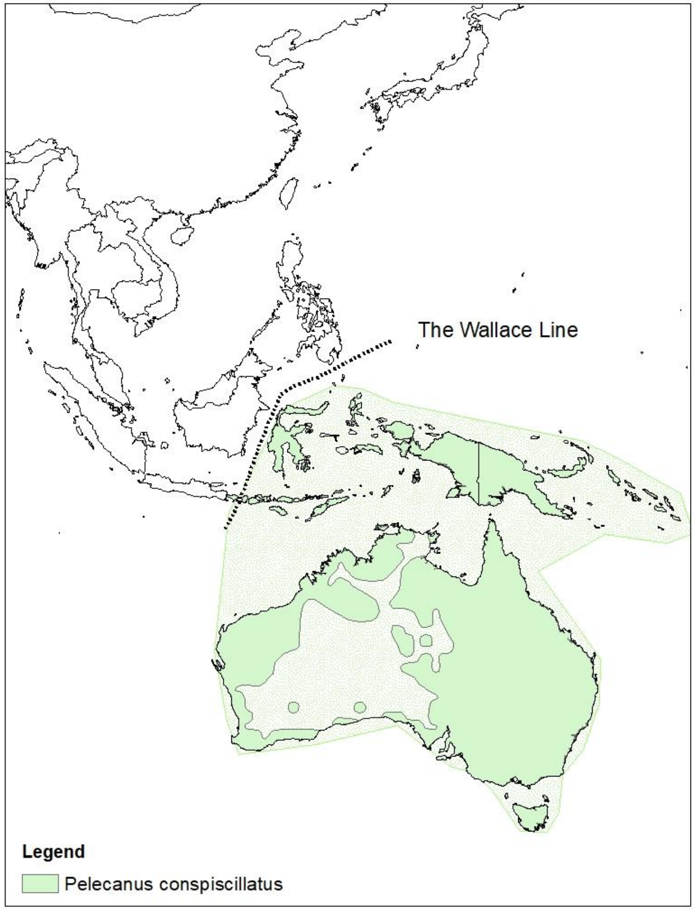

Figure 32. Species distribution of Australian Pelican.

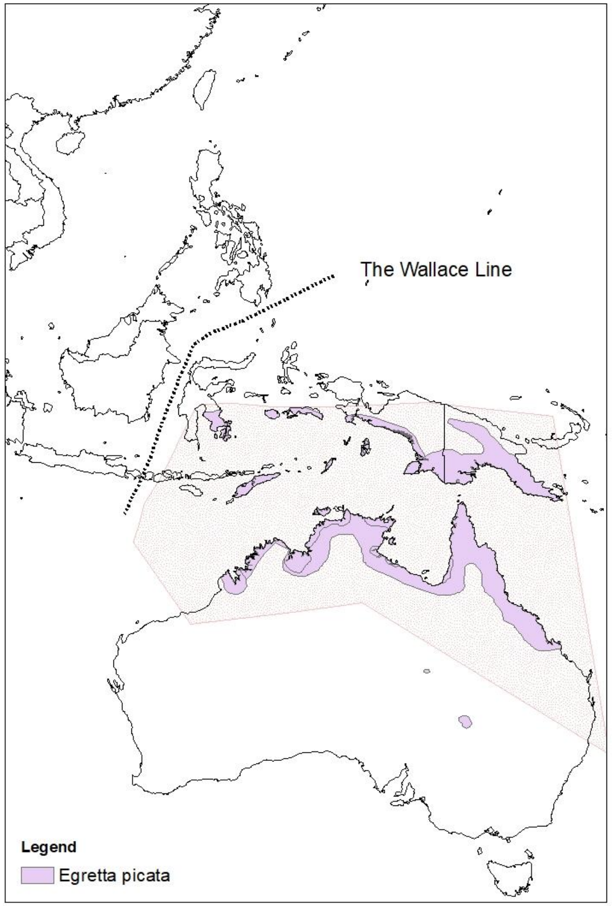

Figure 33. Species distribution of Pied Heron.

Appendix 3 – Waterbird data

Table 1. Overview of known Australian waterbird datasets. Note: some significant, regional waterbird datasets are contained within other datasets and not discreetly identified in the table below (e.g. Gippsland Lakes Ramsar compliance counts – contained within Birdata,

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
Department of Environment, Land, Water and Planning (DELWP) / Arthur Rylah Institute (ARI)	Western Port Philip Bay waterbird counts.	Wetland counts	1981-ongoing	2-6 times / year	All shorebirds	Two wetland complexes in Vic	Ground counts	Yes	Yes	Yes	Counts
	Victorian Summer waterbird counts	Duck counts	2007-ongoing	Annual (November)	All species	Wetlands throughout VIC	Ground counts	Yes	Yes	Yes	Counts
	WetMAP	Wetland counts	2015-2019	monthly	All species	13 watered sites and additional counterfactuals	Ground counts	yes	yes	yes	Counts
Game Management Authority (GMA) /ARI	Aerial Duck Count	Duck counts	2020-ongoing	Annual (November)	8 duck species	Wetlands throughout VIC	Aerial counts, Ground counts	Yes	yes	yes	Counts
DPIPWE - Department of Natural Resources and Environment (TAS)		Duck counts	1987-ongoing	Annual	Duck species only	Wetlands throughout north and east Tasmania	Ground counts	Yes	Yes	Yes	Counts
DEW – Department for Environment and Water (SA)	Spring Waterfowl and wetland surveys.	Wetland counts	2003-present	Annual	Duck species only	85 wetlands throughout se SA	Aerial counts, Ground counts	Yes	Yes	Yes	Counts

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
Birds South Australia	SA Water waterbird monitoring	Wetland counts	2015 - present	variable				yes	no	no	counts
David Paton	Coorong waterbird surveys	Wetland counts	2000-present	Annual	All species	Coorong	Ground Counts	Yes	Yes	Yes	Counts
DPIE – The Department of Planning and Environment (NSW)		Wetland counts	2008-present	1-2 times / year	All species	4 major wetland complexes in inland NSW	Ground counts, boat-based counts, drone counts, and remote sensing	Yes	Yes	Yes	Counts
	Annual Waterfowl Quota	Duck Counts Duck Tracking		Annual	Waterfowl	NSW Riverina	Aerial counts, Ground counts	Yes	Yes		Counts
DEPWS - Department of Environment, Parks and Water Security (NT)	Waterbird colony counts	Colony counts	1980-2015	Largely one-off	Colonial waterbirds	Coastal floodplains throughout NT	Aerial	Yes	No	Yes	Counts
	Magpie goose census	Species-specific counts	1983 – 2000- 2011 - ongoing	Annual	Magpie Goose and Brolga	Top End Floodplains	Aerial	Yes	No	Yes	Counts and breeding status
Austecology for Project Seafarm		Wetland surveys and colony counts	2015 -2019	annual	All waterbirds	Legune floodplain wetlands and adjacent intertidal wetlands, NT.	Ground counts and aerial surveys	no	no	To some extent	Counts and breeding status

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
CLCAC – Carpentaria Land Council Aboriginal Corporation (QLD)		Waterbird breeding colonies.	2009-2014	annual	Colonial waterbirds	Gulf Plains bioregion, QLD	Ground counts and aerial surveys	no	no	To some extent	Counts of nests or breeding pairs.
		Waterbirds.	2009-2019	Opportunistic, northern wet season and dry season.	All waterbirds.	Gulf Plains bioregion, QLD	Ground counts	no	no	No	Counts
		Migratory shorebirds	2014-2022	Opportunistic generally.	Migratory shorebirds	Gulf Plains bioregion and islands, QLD	Ground counts and aerial surveys	no	no	some	counts
CQU – Central Queensland University		Waterbirds (data additional to survey work on Capricorn Yellow Chat).	2004 to present	Annual	All waterbirds	Marine plain wetlands of Capricornia (Broad Sound to Gladstone), QLD.	Ground and boat-based counts	no	no	no	counts
Dr Julian Reid (ANU) and Roger Jaensch (independent consultant)	Arid Flow	Wetland surveys and colony counts	2000 - 2009	Annual in wet years	All waterbirds	Channel Country bioregion, QLD and SA.	Ground counts and aerial surveys	no	no	To some extent	Counts and breeding status

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
CSIRO – Commonwealth Scientific and Industrial Research Organisation is an Australian Government	Waterbird	Colonial-nesting waterbirds					Ground counts, boat-based counts, drone counts, and remote sensing				
BirdLife Australia / Australasian Wader Study Group	Birdata (formerly atlas of Australian Birds)	Atlas	1998-present (includes data dating back to 1901)	Variable	All species	Nationwide	Various	Yes	Variable	To some extent	Counts / Occupancy
	First Atlas of Australian Birds	Atlas	1977–1981	Variable	All species	Nationwide	Various	yes	yes	yes	counts
	Nest Record Scheme	Breeding records	1963 - present	Variable	All species	Nationwide	Various	Yes	Variable	no	Breeding behaviour
	Australian Painted Snipe Project	Species-specific counts	1998-present	Variable	Australian Painted-snipe	Across distribution range	Ground counts and remote sensing.	Variable	Variable	Variable	Counts
	Bittern Project	Species-specific counts	1998-present	Variable	Australasian Bittern and Little Bittern	Across distribution range	Ground counts, drone counts, and remote sensing	Variable	Variable	Variable	Counts

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
	Beach-nesting Birds Program	Species-specific counts	1980 - present	Biennial	Resident shorebirds and terns	400-500 coastal sites across mainland SE Australia	Ground counts	Yes	Yes	Yes	Counts
		Wetland Counts (Murray Darling Basin Database)	1994-1997	Annual to tri-annual	All species	Wetlands across inland NSW and Northern Vic	Ground counts	Yes	Variable	Yes	Counts
		Wetland counts (Victorian Waterbird Database)	1987-1992	Annual to monthly	All species	100s of wetlands across VIC	Ground counts	Yes	Variable	Yes	Counts
	National Shorebird Monitoring Project	Wetland counts	1973-present	Annual, bi-annual to monthly	All species	1000s wetlands across Australia	Ground aerial and boat-based counts.	Yes	Variable	Yes	Counts
DBCA – Department of Biodiversity, Conservation and Attractions		Wetland counts (salinity study)	1997-2013	Annual	All species	Selected wetlands in SW WA	Ground counts	Yes	Yes	Yes	Counts
DBCA –		Wetland counts (SW WA)	1981-1987	Annual to monthly	All species	100s of wetlands across SW WA	Ground counts	Yes	Variable	Yes	Counts

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
Department of Biodiversity, Conservation and Attractions		Wetland counts	1987-1992	Annual	All species	100s of wetlands across SW WA	Ground counts	Yes	Variable	Yes	Counts
		Wetland counts	2006-2015	Bi-annual	All species	Wetland complex in Esperance region	Ground counts	Yes	Variable	Yes	Counts
eBIrd Australia - Cornel Lab (USA)		Range of datasets	NA	Variable	All species	Nationwide	various	Variable	Variable	Variable	Variable
VBA – Victorian Biodiversity Atlas		Range of datasets	NA	Variable	All species	Vic	various	Variable	Variable	Variable	Variable
ALA – Atlas of Living Australia		Range of datasets	NA	Variable	All species	Nationwide	various	Variable	Variable	Variable	Variable
BDBSA – Biological Databases of South Australia		Range of datasets	2003-present	Variable	All species	SA	various	Variable	Yes	Yes	Counts
NVA WA - Natural Values Atlas		Range of datasets	NA	Variable	All species	WA	various	Incidental	Variable	Variable	Incidental
NVA Tas – Natural Values Atlas		Range of datasets	NA	Variable	All species	Tas	various	Incidental	Variable	Variable	Incidental
WildNet		Range of datasets	NA	Variable	All species	QLD	various	Incidental	Variable	Variable	Incidental

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
Bionet		Range of datasets	NA	Variable	All species	NSW	various	Incidental	Variable	Variable	Incidental
UNSW – University of New South Wales	East Australian Waterbird Surveys	Wetland surveys and colony counts	1983-present	Annual	All species	Eastern 1/3 of continent	Aerial surveys, ground counts.	Yes	Yes	Yes	Counts
UNSW – University of New South Wales	MDBA Murray Icon counts	Wetland counts	2007-present	Annual	All species	MDB wetlands	Ground counts, boat-based counts, drone counts, and remote sensing.	Yes	Yes	Yes	Counts
	Flow-MER	Wetland counts	2010-present	Annual	All species	MDB wetlands	Ground counts, boat-based counts.	Yes	Yes	Yes	Counts
		Colonial Waterbirds	2008-present	Annual (1-4) surveys	Colonial nesting species	MDB wetlands	Ground counts, boat-based counts, drone counts, and remote sensing	Yes	Yes	To some extent	Counts & Breeding status
	Feather Map	Genetic sampling	2018 - present	NA	waterbirds	national	Remote analysis of feather samples	NA	NA	NA	NA
	National Colonial Waterbird Database	Atlas	1899 – 2008 (currently being updated)								Counts & Breeding status
AWC – Australian Wildlife Conservancy		No data	NA	NA	NA	NA		NA	NA	NA	NA

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
Melbourne Water	Melbourne Water regional Bird Monitoring project	Wetland counts	2012-present	Annual to monthly	All species	Over 200 Wetlands in Greater Melbourne	Ground counts and boat-based counts.	Yes	Yes	Yes	Counts
North Central CMA	Ramsar compliance surveys.	Wetland surveys and colony counts	2009-present	waterbirds	waterbirds	25 wetlands in northern VIC	Ground counts	Yes	Yes	Yes	Counts & Breeding status
Goulburn Broken CMA	Ramsar compliance surveys.	Wetland surveys and colony counts	2009-present	Up to 6 times / year	waterbirds	Barmah Millewa Ramsar site	Ground counts	Yes	Yes	Yes	Counts & Breeding status
		Colonial waterbird database	1905-present	variable	waterbirds	Hattah Lakes Ramsar site	Ground counts	Yes	Yes	Yes	Counts & Breeding status
Mallee CMA	Ramsar compliance surveys.	Wetland surveys and colony counts	2009-present	Up to 6 times / year	All species	Barmah Millewa Ramsar site	Ground counts	Yes	Yes	Yes	Counts & Breeding status
QWSG – Queensland Wader Study Group	Shorebird monitoring	Wetland counts	1993-present	Annual to monthly	All species	100s of QLD wetlands	Ground/boat counts	Yes	Yes	Yes	Counts
COG – Canberra Ornithological Group		Wetland counts	1987-present	At least annually	All species	Wetlands in ACT / NSW	Ground counts	Yes	Yes	Yes	Counts
MYSMA –		Coastal counts	2004-present	Tri-annual	Shorebirds	80 Mile Beach, Roebuck Bay	Ground counts	Yes	Yes	Yes	Counts

Data source	Program name	Monitoring type	Time Period	Survey frequency	Species	Range	Method	Standardised method?	Standardised effort?	Fixed sites used?	Unit
Monitoring of Yellow Sea Migrants Australia											