



Mr Ross Sneddon of the John Curtin School of Medical Research takes blood samples from water birds during the Australian encephalitis epidemic of 1974.

## Arboviruses ~ hidden hitchhikers

Four years ago an epidemic of Australian (or Murray Valley) encephalitis swept across the continent, covering an unprecedented area. Of the 58 cases reported, 12 died and the remaining 46 became so ill they had to receive treatment in hospital. Consequently, the disease received a great deal of public attention.

Another epidemic struck at the same time, but, since nobody died, we didn't hear much about it. This second epidemic was of a disease known as 'epidemic polyarthritis'.

This malady results in fever, headaches, and pains in the joints. An occasional patient may suffer for up to a year with its after-effects. We don't know how many people were affected in Australia that year, but it seems certain that the total far exceeded the 60 afflicted with Australian encephalitis.

The interesting thing about these two diseases is that they both seem to flare up under similar climatic conditions. Both are caused by what are known as arboviruses, and in the past the two have been confused.

Arboviruses multiply in blood-sucking

insects, or in ticks. They can also infect reptiles, birds, or mammals. Other virus diseases, like myxomatosis, are also spread by biting insects, but in these cases the insect merely acts like a dirty needle. The viruses are transferred from one animal to another, but they don't live and multiply in the insects. These are not arboviruses.

Until recently expert opinion has regarded Australia as remarkably free of

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arboviruses. At one stage only one — dengue — was known. Then in 1951 Mr Eric French, formerly of the Walter and Eliza Hall Institute, Melbourne, and of CSIRO, and Dr J. A. R. Miles of the University of Adelaide showed that the mysterious 'Australian X' disease now known as Australian encephalitis was also caused by a virus that seemed to be carried by mosquitoes. Further studies of later outbreaks confirmed this.

### Forty or more

Today we know of some 40 arboviruses resident in Australia, and the list keeps growing as research continues. Bluetongue virus, which caused such a stir late last year, is but the most spectacular recent discovery. We have a long way to go towards understanding how the Australian encephalitis virus and all these other arboviruses are transmitted.

Of those 40-odd, we know for certain that five can affect human beings. These are dengue virus, Australian encephalitis virus, Ross River virus (the cause of epidemic polyarthritis), and two others called Kunjin and Sindbis. Both of these last two cause fevers. A further four are



known to cause diseases in domestic stock.

Of the remaining 31 arboviruses known, some are suspected of producing fever in Man, but only in exceptional circumstances. Diseases may be attributed to these viruses when we understand them better. All of these viruses except Australian encephalitis have been isolated only during the last 20 years, so there's a great deal that we don't know about them.

It's hoped that dengue outbreaks are a thing of the past; the last one was at Townsville in 1955. Transmission of the disease depends largely on the mosquito *Aedes aegypti*, which nowadays has been driven out of all our larger towns. It still occurs in some smaller country towns in the northern half of Australia and, since dengue still exists in the Pacific region, outbreaks could still occur in such towns should an infected individual come from overseas.

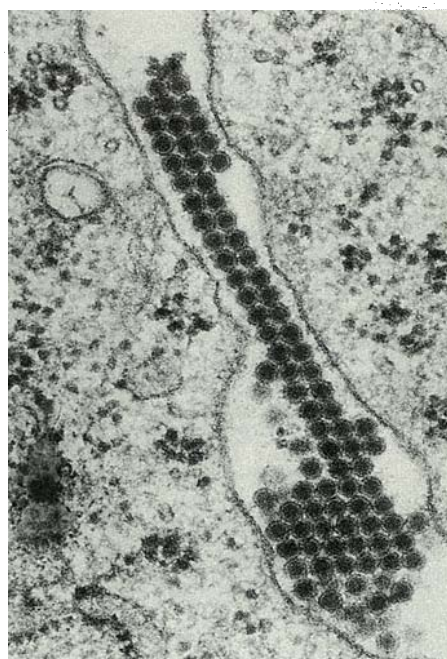
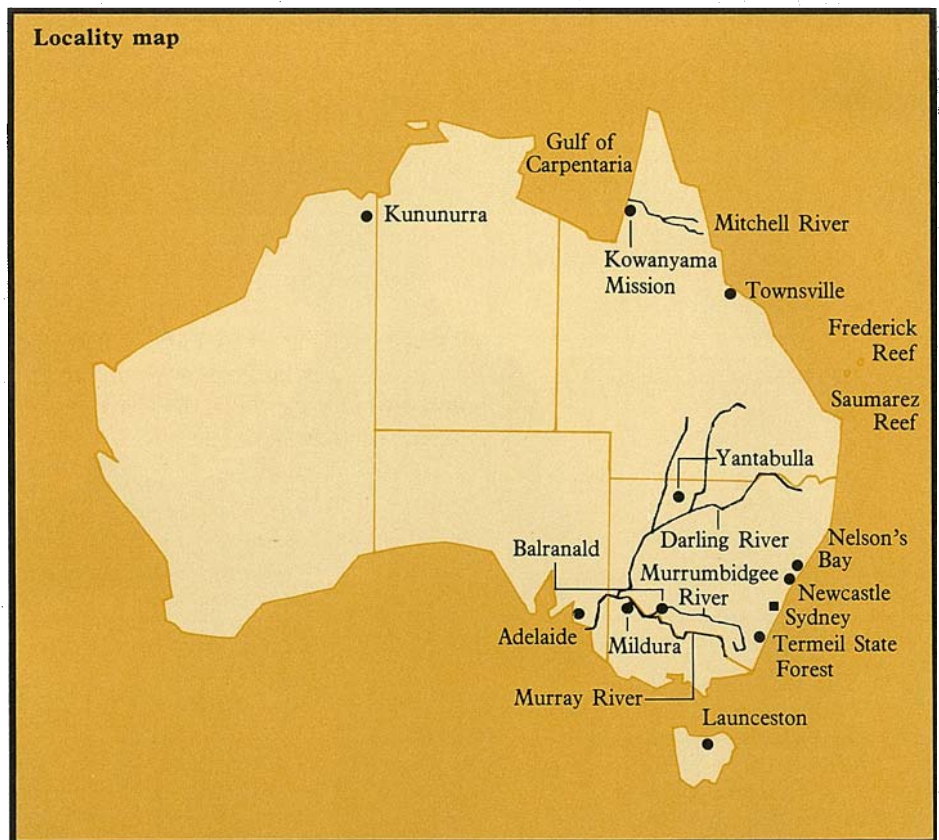
### Killer spurred research

Australian encephalitis has taken the lives of about 150 people during the last 60 years. It was the drive to understand the causes of epidemics of this disease that first led to the discovery of other arboviruses.

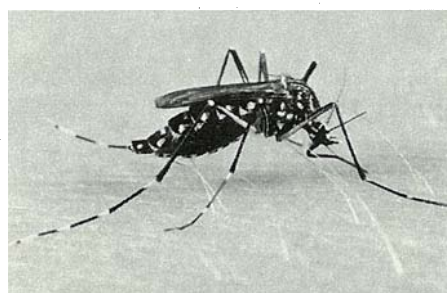
Outbreaks of what was probably Australian encephalitis occurred in 1917, 1918, 1922, 1925, 1951, 1956, 1971, and 1974. Not all reached epidemic proportions.

After studying the Australian encephalitis epidemic of 1951, Dr Miles and Dr S. G. Anderson, formerly of the Walter and Eliza Hall Institute, independently came to the same conclusion: between epidemics the virus survives in northern Australia, or perhaps even in New Guinea. Even though, at that time, no cases of Australian encephalitis ever seemed to occur in these regions, blood samples taken showed that antibodies to the virus seemed common in people there — which implied frequent infection. The virus itself was later isolated in both northern Australia and in New Guinea.

Both Dr Anderson and Dr Miles also showed that epidemics in south-eastern Australia, which always occurred in summer and autumn, happened in years when there had been a wet spring in northern New South Wales, western Queensland, and the Northern Territory. Presumably such seasons allowed the probable mosquito carrier, *Culex annulirostris*, to breed in sufficiently large numbers to enable the virus to spread into the south-



Australian encephalitis virus in mosquito cells — the viruses appear in neat rows.



*Aedes aegypti*, the mosquito that transmits dengue, feeding on a man's arm. The last outbreak of this disease was at Townsville in 1955.

east, where it could cause disease outbreaks among unimmunized people.

In addition, Dr Anderson suggested that water birds may introduce the virus into the epidemic areas when migrating from the north to the south-eastern part of the continent. A wet spring up north would allow birds to breed in large numbers, thus increasing the chances of birds carrying the virus to the south-east.

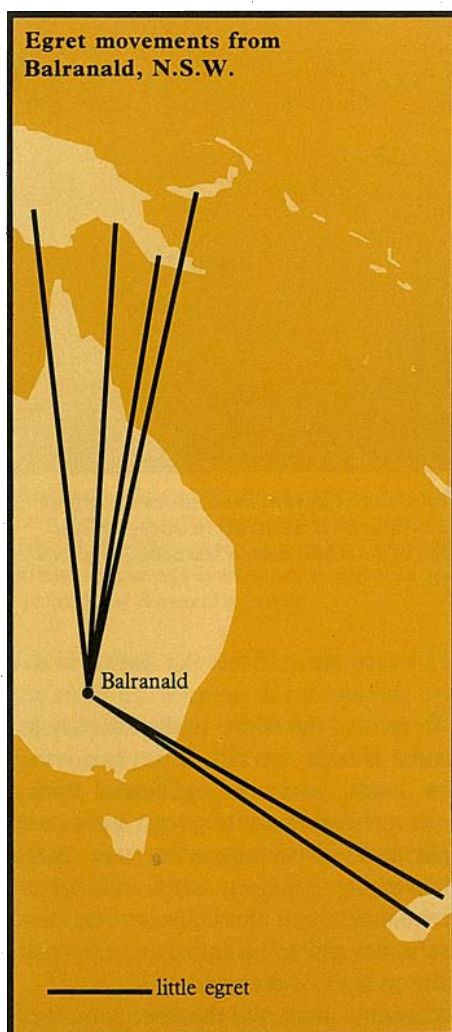
### View unconfirmed

The evidence that has accumulated since these ideas were put forward in 1953 has not completely vindicated them. Partly it confirms this classical view, but it also in part denies it.

Without doubt Australian encephalitis virus is continually present in northern Australia — as the box shows. But studies of the 1974 epidemic of Australian encephalitis have yielded little evidence that it came from the north. In fact, Dr Ian Marshall of the John Curtin School of Medical Research at the Australian National University points out, it's even possible to interpret the evidence as pointing to an epidemic sweeping from south to north.

Unlike most before it, this epidemic didn't only affect the Murray Valley. People suffered from Australian encephalitis also in Queensland, in the Northern Territory, and at Kununurra, W.A. Two people actually contracted the disease in suburban Adelaide.





Water birds frequently move from one part of the Australian continent or New Guinea to another. This diagram shows where birds banded at Balranald, N.S.W., have been recovered.

Dr Marshall notes that the first case of the epidemic was diagnosed at Balranald in southern New South Wales. The disease then flared up in the rest of the region, all the first 20 cases of the epidemic being infected in the Murray Valley. The first case in northern areas didn't occur until the tenth week, by which time the Murray Valley epidemic was two-thirds of the way to being over.

### Do birds carry it?

And what about the suggestion that birds carry the disease down from the north? The Australian Bird-banding Scheme, which is run by the CSIRO Division of Wildlife Research (see *Ecos* 11), has revealed that water birds certainly do move all over the continent, often very rapidly. In addition, early in the 1974 epidemic Dr Marshall and his colleagues found that water birds, particularly the herons, had very high levels of antibodies to Australian encephalitis in their blood. Indeed the researchers actually managed

*Until recently expert opinion has regarded Australia as remarkably free of arboviruses.*

to isolate the virus from a white-faced heron.

High antibody levels implied that the birds had been infected recently, which supported the view that the birds were somehow involved in the cycle by which the disease spreads. But the Canberra workers didn't have information about antibody levels in the birds in the months before the outbreak started, so these high levels didn't prove the point.

Dr Marshall and his colleagues have tried ever year since 1974 to recover Australian encephalitis virus from birds and mosquitoes in the Murray Valley, but all their attempts have failed. This and most of the 12 other arboviruses that were so prevalent in 1974 seem to have disappeared.

But perhaps they haven't. After all, Australian encephalitis is very hard to find in much of northern Australia where we know it's always present. And there's evidence that birds may not even be the major non-insect host in the Murray Valley.

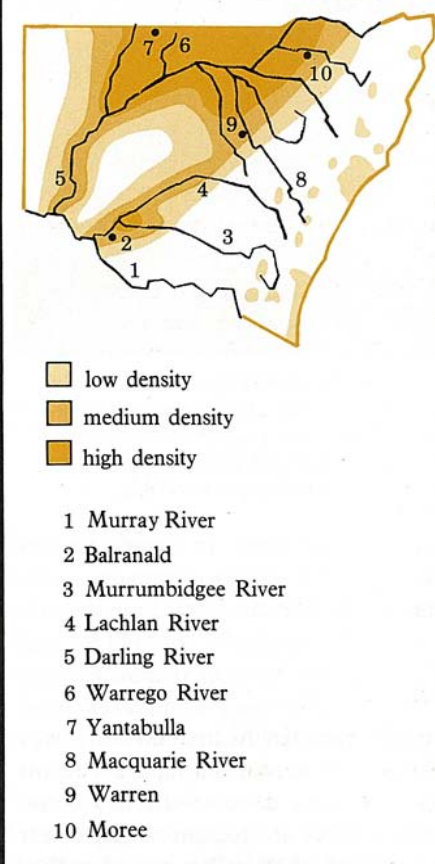
### Or do pigs?

Mr Geoff Gard and his colleagues of the Glenfield Veterinary Research Station, N.S.W., have recently been studying wild pigs in cooperation with Dr Gwendolyn Woodroffe of the John Curtin School of Medical Research. They have shown that



Sentinel herds of cattle are scattered all over Australia and New Guinea. They are routinely bled and tested for arboviruses.

### Wild pig distribution in New South Wales



**Are wild pigs a reservoir for Australian encephalitis virus? Pigs shot near Yantabulla seemed to be infected with the virus in years when no cases of the disease were reported. Note the high pig density near Balranald, where the first case of the 1974 epidemic occurred.**

wild pigs shot at Yantabulla in north-western New South Wales were infected by Australian encephalitis virus during the summers of 1971–72, and 1972–73 — two summers when no cases of Australian encephalitis were reported. (Epidemics occurred early in 1971, and at the beginning of 1974.)

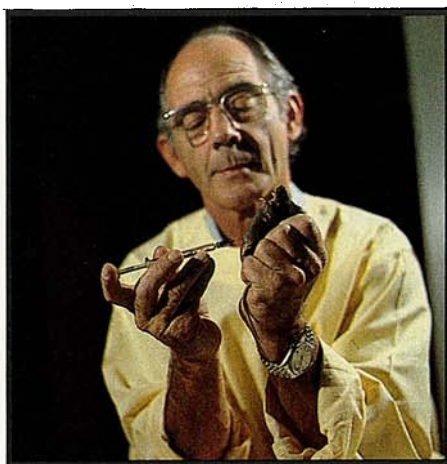
Interestingly, pigs exist in large numbers around Balranald where the first case of the 1974 epidemic was diagnosed. Could it be that the virus survives in the southern part of the continent after all, and flares up when conditions become particularly suitable?

This idea does find some support when we look at what's now known about other Australian arboviruses. During the early 1960s, Dr Ralph Doherty and his colleagues at the Queensland Institute of Medical Research discovered that Ross River virus causes epidemic polyarthritis. (Dr Doherty, the Institute's third Director, became Professor of Social and Preventive Medicine at the University of





**Barmah forest during the 1974 floods in the Murray Valley. After the floods abated, lush grass growth and surface water that remained gave perfect conditions for mosquito breeding.**



**Dr Ian Marshall injects Australian encephalitis into laboratory mice.**



**White-faced heron, sometimes known as the blue crane. Australian encephalitis virus has actually been isolated from this bird.**

Queensland last year.) In some ways this virus appears to behave similarly to the Australian encephalitis one. Like the latter, it is transmitted by mosquitoes and major epidemics flare up at similar times, although isolated cases occur every year throughout eastern Australia.

Recently, Dr Marshall and his fellow researchers have discovered a new strain of the virus at Nelson's Bay near Newcastle, N.S.W. This strain differs slightly from the one found in northern Australia. What's more, the new strain has infected people and remained active but unaltered at Nelson's Bay for at least 7 years in succession.

Dr Marshall is now convinced that the strain at Nelson's Bay is always present. In fact he thinks that it is always around down the whole of the New South Wales northern and southern coastlines. Not long ago his group isolated Ross River virus from mosquitoes as far south as the Termeil State Forest on the South Coast of New South Wales. A case of epidemic polyarthritis had been contracted nearby the year before.

### **Livestock diseases**

A similar situation may also apply with ephemeral fever, a disease of cattle. Major epizootics (the animal equivalent of epidemics) of this virus disease occurred in 1935-36, 1955-56, 1967-68, and 1970-71, and during the period of 1972-74. In addition, sporadic outbreaks occurred in intervening years.

The 1967-68 epizootic swept the whole country from north to south in 4 months. In 1969, the CSIRO Division of Animal Health set up a system of 'sentinel' herds of cattle scattered around the whole of Australia, and in Papua New

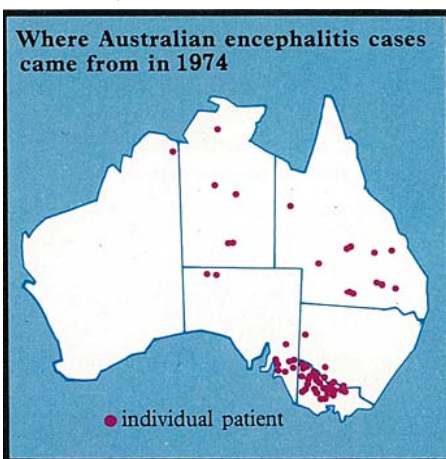
Guinea. These herds, some on private properties and some on government field stations, have been bled routinely and tested for an array of virus infections. The aim has been to get more information about the spread of such diseases as ephemeral fever.

By testing these herds Mr Toby St George, the scientist responsible for setting them up, was able to show that each of the ephemeral fever epidemics after 1970 seemed to start near the Gulf of Carpentaria. Nevertheless, on two occasions separate outbreaks of the disease occurred in the Hunter Valley, N.S.W. It

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**The 1974 Australian encephalitis epidemic covered an unprecedented area. Cases came from a number of localities where the disease had never before been recorded.**

seems very possible that the disease is always present in this area.

In spite of the efforts of the Division of Animal Health, we still do not know the main vector insect for ephemeral fever. However, the disease's spread seems too rapid and too complete to have been achieved by migratory birds. In Africa mosquitoes aren't the culprits. The disease is thought to be transmitted by tiny biting midges of the genus *Culicoides*.

Certainly midges of the same genus are responsible for spreading other diseases in domestic stock in Australia. One — *Culicoides brevitarsis*, which breeds in cattle dung — carries Akabane virus and D'Aguiar virus: the first produces a disease known as curly calf, and the second may cause cattle to abort.

### **World picture**

Most arboviruses found in Australia do not appear to be isolated entities peculiar to this continent. Instead they seem to fit into a world picture of broad groupings. Thus the Australian encephalitis virus is closely related to, but slightly different from, the Japanese encephalitis virus found over much of Asia and Japan. Ross River virus seems to be an Australian variant of another broader Asian group, as does Kunjin.

In the case of stock diseases like ephemeral fever and Akabane, our local strains seem to be all but identical with viruses causing the same diseases elsewhere. But sheep and cattle only arrived in Australia comparatively recently, and so, therefore, did their viruses. Perhaps cattle, sheep, and other domestic stock have been here too short a time for exclusively Australian strains to develop.

In fact Australian arboviruses seem to

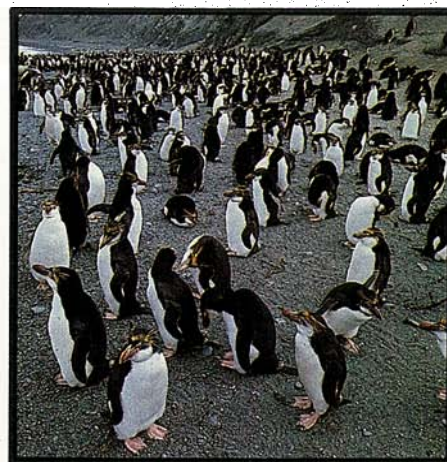




Antarctic skuas and giant petrels feeding on a dead sea-elephant on Macquarie Island. Perhaps these scavengers transfer viruses from one land mass to another around the Antarctic circle.



The radio tower on Saumarez Reef. Illness among radio technicians maintaining this tower first created the interest that led to the isolation of Saumarez Reef virus.



Royal penguins at Nugget's beach, Macquarie Island. 'Nugget' virus came from near here.

fall into three types. The first consists of the stock diseases that seem to infect only imported animals (including those that have gone wild). Those in the second group, which includes Ross River and Australian encephalitis viruses, affect Man, some domestic stock (Australian encephalitis virus probably causes nervous disease in horses), and also wildlife. These viruses have close relatives in Asia.

The third group consists of viruses that have been isolated only from native reptiles and marsupials. There's little evidence to date that these affect Man, or his animals, and most of them seem unrelated to arboviruses in Asia or elsewhere.

It seems reasonable to regard this third group as very ancient viruses that evolved with our wildlife a very long time ago. Very probably most of the second group also had been resident in Australia well before European Man, at least, arrived. Presumably the Australian variants evolved in isolation after being introduced by some means from Asia. So Man may be regarded as walking into an environment where diseases like Australian encephalitis and epidemic polyarthritis already existed. He has merely provided himself as an additional host for these arboviruses.

### Exchange between continents

The existence of groups of arboviruses that vary little within geographical regions such as Asia, Australasia, and Africa suggests that these infectious agents must move around within the regions comparatively rapidly. The fact that arboviruses found in different continents are often related suggests too that some interchange of viruses must occur between continents. Birds have already received

mention as suspected transporters of Australian encephalitis. Probably nothing moves around the world as much as migrating birds, so these have become the obvious suspects.

Migrating birds may transport arboviruses in at least two possible ways: either in their blood, or in parasites — ticks in particular.

In many ways it's easier to imagine how they could carry the viruses over very long distances in ticks. An infected bird will only have virus particles in its blood for a few days before its body defences eliminate them. At this time the bird would perhaps be weakened by sickness — not a propitious moment for successfully completing very long distance flights.

With such thoughts in mind, Mr Durno Murray of the CSIRO Division of Animal Health collected samples of the tick *Ixodes uriae* from a rookery of royal penguins on Macquarie Island and



The brown litter is 26 000 mosquitoes, all of which were caught in a single light trap during one night in Barmah forest during the 1974 Australian encephalitis epidemic.

brought them to Australia. Here he handed them over to the Queensland Institute of Medical Research, which tested them for arboviruses. Two viruses new to science resulted. They received the names 'Nugget' and 'Taggart' — after two horses that had once lived on the island.

### Arctic affinities

Checking these two new viruses against Australian ones revealed no relationship. They were then submitted to the Yale Arbovirus Research Unit, New Haven, U.S.A., which is the World Health Organization's international reference centre for arboviruses. This Unit showed that the two new viruses were related to two groups of viruses, each to a different one, which had only been isolated previously in the Northern Hemisphere. Surprisingly, viruses from the group related to 'Nugget' had previously been isolated only from near the Arctic Circle. What's more, the related viruses had been isolated from the same species of tick — *I. uriae*.

This finding raises intriguing possibilities. This particular tick has a strange distribution. It occurs only near the Arctic Circle and near the Antarctic Circle; now it seems to contain viruses that are related too.

Those ticks collected on Macquarie Island seemed only to be feeding on royal penguins — hardly a likely candidate for transporting the tick and virus from one hemisphere to the other. Perhaps at some time migrating sea birds introduced the ticks and the viruses. The ticks may then have adapted themselves to feed on the royal penguin, and in time a separate virus strain evolved.

Unfortunately this story has its prob-



In 1957, the Queensland Institute of Medical Research began a 4-year survey that would enable it to map the distribution in Queensland of antibodies to a group of arboviruses known as Group B. This group includes Australian encephalitis, dengue, and several other exotic viruses.

The survey showed that every year antibodies to Group B viruses were common in Aboriginal children and adolescents on settlements and missions bordering the Gulf of Carpentaria. This result suggested that these communities were being infected each wet season. Further tests showed that most of these antibodies seemed to be a reaction to infection by Australian encephalitis virus.

In contrast, tests on human serum samples taken from the eastern coast of tropical Queensland revealed antibodies to dengue, but not to Australian encephalitis.

So it appeared that the virus causing Australian encephalitis was active on the eastern side of the Gulf of Carpentaria every year. Could the area act as a reservoir from which the disease spread to the southern part of the continent when conditions were right?

More recent work by the Institute has raised problems for this interpretation. The Queensland researchers turned their attention to trying to isolate the virus itself from mosquitoes — particularly *Culex annulirostris*, the commonest one. They concentrated on Kowanyama (formerly Mitchell River) mission on the Gulf of Carpentaria near the base of Cape York.

A relatively small total (by the standards of the Institute) of 17 000 mosquitoes collected from the mission in 1960–61 yielded no less than eight different viruses. The eight included the Australian encephalitis virus, and this was

the first time that it had actually been isolated from insects. It also included two Asian viruses not previously known to occur in Australia, and five new ones, among which was Kunjin virus.

Like Australian encephalitis virus, several of the others isolated, including Kunjin, are classified in Group B. This meant that many of the earlier blood tests showing reactions to Group B viruses, which had been ascribed to infection by Australian encephalitis, might well have been caused by these others. Almost certainly this was the case. Nevertheless, there seems to be little doubt that the Australian encephalitis virus does occur in the area in most years.

We now know that the virus also permanently remains at Kununurra on the Ord River scheme, W.A. A study group from the University of Western Australia, directed by Professor Neville Stanley, isolated it from *C. annulirostris* mosquitoes during the post-wet-season months of April, May, or June during 1972, 1973, and 1974.

Incidentally, the research group has found that most of the mosquitoes in the area breed in the swamps that surround the diversion dam. Lake Argyle, the lake that recently formed behind the completed Ord River dam, remains almost completely free of mosquitoes. But then, Professor Stanley and his colleagues point out, it is so new that lakeside vegetation suitable for the mosquitoes to breed in has yet to appear. As the lake matures such vegetation may develop, with a consequent increase in the number of mosquitoes. This may bring the risk of tourists becoming infected with Australian encephalitis virus, and contracting the disease.

Such studies leave no doubt that the virus survives in parts of northern

Australia, at least during most years. However, these isolations of the virus from mosquitoes have been from country that has a long dry season. During each dry season the numbers of mosquitoes and other biting insects fall to a low level. How does the virus survive during these periods?

That question remains unanswered. No evidence of virus activity in the dry season has yet been turned up anywhere.

The scientists of the Queensland Institute of Medical Research thought that pockets of rainforest that occur on Cape York seemed likely places where the virus might ride out the dry season. Indeed, such forests have been proved to act as reservoirs for arboviruses on other continents. However, these researchers could find no evidence of virus activity in the rainforest pockets of Cape York.

Professor Doherty and others among his former colleagues at the Institute now suspect that no single area provides a refuge for the virus. Instead it may survive over the whole of northern Australia in isolated pockets whose locations move each season.

Arthropod-borne viruses in Australia and their relation to infection and disease.

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lems. Migrating sea birds living on Macquarie Island come there to breed. They include albatrosses, shearwaters, and petrels. Albatrosses and petrels usually disperse only into the oceans of the Southern Hemisphere. The shearwaters do move into northern latitudes but, as it happens, they don't visit land there. They come to land only to breed — on Macquarie Island.

Once infected ticks have reached

subantarctic latitudes they could conceivably be dispersed from one piece of land to another by scavenging birds like the Antarctic skuas or giant petrels. In fact, one tick of the species *I. uriae* has been removed, still living and full of blood, from the head of a giant petrel caught off Sydney.

It's just possible that arboviruses reach the Southern Hemisphere in stages by passing down the length of South America

via other tick species related to *I. uriae*.

Tests, incidentally, showed that one of the two new viruses isolated from Macquarie Island could multiply when injected into the mosquito *Aedes aegypti*. This may perhaps suggest that viruses found in ticks can be passed by mosquitoes — one of the main vectors of arbovirus epidemics. No evidence could be produced to show that the viruses can infect human beings.



## Antibodies in people at Kununurra

	(% positive to antibody test)		
	Australian encephalitis virus	Sindbis virus	Ross River virus
European adults	53	30	41
European children	24	14	21
Aboriginal adults	96	20	51
Aboriginal children	77	36	68
people resident < 3 years	26	0	23
people resident > 3 years	64	8	41

Many of the 441 people sampled (about one-quarter of the town's population) had been infected with arboviruses at some time. Infection seems to depend on exposure: usually more adults had been infected than children, and more long-term residents had been infected than short-term ones.

## Viruses recovered from mosquitoes in the Murray Valley

virus	number of times isolated
Sindbis	47
Ross River	16
Australian encephalitis	38
Kunjin	110
Edge Hill	2
Koongol	7
Umbre	1
Kowanyama	2
others	17



During February 1974, Dr Marshall and his team recovered some 12 different arboviruses from mosquitoes in the Murray Valley. Two of these were new to science.

## Tropical ticks

The same cannot be said about a virus Mr St George, Professor Doherty, and colleagues isolated from bird ticks on islands of the Great Barrier Reef. The researchers investigated the islands on the Saumarez and Frederick Reefs and nearby coral cays after reports that several technicians who built and serviced the automatic weather-reporting stations on the islands had become ill with a fever.

They reported being bitten by large numbers of ticks.

Studies of the ticks revealed that they contained another new virus, which has since been named Saumarez Reef virus. Blood tests on technicians who had not become ill showed that some had raised antibody levels for this or related viruses. However, none of the men who actually became ill would agree to put themselves on the wrong end of a needle, so the researchers don't know whether these four individuals also had high antibody levels for Saumarez Reef virus.

Russian researchers have isolated a closely related virus from *I. uriae* (the Macquarie Island tick) in the Bering Strait. This virus had caused fever in three biologists trapping gulls and kittiwakes.

## Gulls yield a surprise

The virus samples obtained from the islands on the Barrier Reef came from a type of tick, known as a 'soft' tick, that had been feeding on sooty terns. At much the same time a staff member of a museum in Launceston, Tasmania, collected live ticks from two dead silver gulls on the island and sent them to the Division of Animal Health. These were 'hard' ticks — closely related to, but a different species from, those on Macquarie Island.

They reached Mr St George, who managed to isolate viruses from ticks on each of the birds. As usual, he sent these viruses to the Queensland Institute of Medical Research for identification.

To everybody's surprise, the viruses all

turned out to be identical with those extracted on the Barrier Reef. They too were Saumarez Reef virus, yet here they were coming from a completely different type of tick 2500 km to the south.

Like a few pieces from a jigsaw puzzle, such bits of information tell us little about how the movements of birds are related to the distributions of the ticks and arboviruses.

The pieces we have do nothing more than tantalize. But evidence is accumulating. Dr Harry Hoogstraal and his colleagues of the United States Naval Medical Research Unit in Cairo, for example, have isolated many viruses from sea birds and their ticks in Africa, Australia, and islands in the Indian Ocean and the Coral Sea. Sooner or later a clear picture will emerge.

## More about the topic

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