

2011-2012 Annual Report



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EXECUTIVE OVERVIEW

- **For the 2011-2012 season**, the NSW Arbovirus Surveillance Program: (i) monitored mosquito vector populations and undertook surveillance of arbovirus activity through virus isolation in the NSW inland, coastal regions and metropolitan Sydney, (ii) monitored flavivirus transmission through the testing of sentinel chickens across inland NSW. Most sites operated between November and April.
- **The climatic conditions** leading up to the 2011-2012 season for the inland were of above to well above average rainfall for the last quarter of 2011 and first quarter of 2012, and there was widespread flooding across inland regions. The Forbes hypothesis was suggestive of probable MVEV activity for the 2011-2012 season, while the Nicholls was not. For the coast, conditions were mostly similar with almost continual rainfall over the summer months and into early autumn.
- **For the inland**, the excessive precipitation and flooding meant that mosquito numbers were again extremely high, with over 170,000 mosquitoes trapped. While down upon 2010-2011, this season's collection was five times larger than that of 2009-2010. One trap from Griffith yielded over 18,000 mosquitoes - a record for the program. There was considerable arboviral activity, with 67 isolates (18 RRV, 18 SINV, 1 EHV, 15 KOKV, 1 KUNV, and 14 unknowns), and 15 seroconversions in the sentinel chickens (14 MVEV and 1 KUNV). The MVEV seroconversions were limited to early-mid December 2011, while the KUNV isolate and seroconversion were from later in the season (mid-March for the isolate and late April for the seropositive).
- **Human notifications from the inland** of RRV and BFV totalled 288, which was slightly below the long term average of 322, and included 250 RRV and 38 BFV. There was one human case of MVEV disease from northern NSW.
- **As of August 2012**, both the Forbes and modified Nicholls hypotheses are indicating a possible MVEV epidemic for the season of 2012-2013.
- **For the coast**, mosquito numbers were again well below previous years, largely due to the lowest collections of *Aedes vigilax* to date. With reduced abundance of the major coastal vector, arboviral activity was relatively minimal. There was a total of ten isolates, including 5 RRV, 1 EHV, and 4 unknowns. Most of the isolates were from freshwater breeding mosquitoes, notably *Coquillettidia linealis* and *Culex annulirostris*.
- **Coastal disease notifications** of RRV and BFV totalled 493 cases, including 237 RRV and 256 BFV, and this was well below the average of 737. The statistical local area that produced the highest case load was Tweed, with 52 notifications (32 RRV & 20 BFV). There was also one human case of KUNV disease from the south coast of NSW; this is the first human case from the coast.
- **For the Sydney locations**, five sites operated and mosquito numbers were slightly lower than the last season. Again, this was due to the very small collections of *Aedes vigilax*. There was one isolate of RRV from Lambert Park in Ryde. Human notifications were well below the average of 81, with a total of 39 cases including 32 RRV and 7 BFV.
- **The NSW Arbovirus Surveillance Web Site** <http://www.arbovirus.health.nsw.gov.au/> continued to expand and now has over 281MB of information with 2,150+ pages.

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NSW ARBOVIRUS SURVEILLANCE AND MOSQUITO MONITORING PROGRAM 2011-2012

INTRODUCTION

The aim of the Program is to provide an early warning of the presence of Murray Valley encephalitis virus (MVEV) and Kunjin (KUNV) virus in the state, in an effort to reduce the potential for human disease. In addition, the Program compiles and analyses mosquito and alphavirus, especially Ross River (RRV) and Barmah Forest (BFV), data collected over a number of successive years. This will provide a solid base to determine the underlying causes of the seasonal fluctuations in arbovirus activity and the relative abundance of the mosquito vector species, with the potential to affect the well-being of human communities. This information can then be used as a basis for modifying existing local and regional vector control programs, and creation of new ones.

METHODS

Background

Arbovirus activity within NSW has been defined by the geography of the state, and three broad virogeographical zones are evident: the inland, the tablelands and the coastal strip (Doggett 2004, Doggett and Russell 2005). Within these zones, there are different environmental influences (e.g. irrigation provides a major source of water for mosquito breeding inland, while tidally influenced saltmarshes along the coast are highly productive), different mosquito vectors, different viral reservoir hosts and different mosquito borne viruses (e.g. MVEV and KUNV occur only in the inland, while BFV is active mainly on the coast, and RRV is active in both inland and coastal areas). As a consequence, arboviral disease epidemiology often can be vastly different between regions and thus the surveillance program is tailored around these variables.

Arbovirus surveillance can be divided into two categories: those methods that attempt to predict activity and those that demonstrate viral transmission. Predictive methods include the monitoring of weather patterns, the long-term recording of mosquito abundance, and the isolation of virus from vectors. Monitoring of rainfall patterns, be it short term with rainfall or longer term with the Southern Oscillation, is critical as rainfall is one of the major environmental factors that influences mosquito abundance; in general, with more rain come higher mosquito numbers. The long-term recording of mosquito abundance can establish baseline mosquito levels for a location (i.e. determine what are 'normal' populations), and this allows the rapid recognition of unusual mosquito activity. The isolation of virus from mosquito vectors can provide the first indication of which arboviruses are circulating in an area. This may lead to the early recognition of potential outbreaks and be a sign of the disease risks for the community. Virus isolation can also identify new viral incursions, lead to the recognition of new virus genotypes and identify new vectors. Information from vector monitoring can also reinforce and strengthen health warnings of potential arbovirus activity.

Methods that demonstrate arboviral transmission include the monitoring of suitable sentinel animals (such as chickens) for the presence of antibodies to particular viruses (e.g. MVEV and KUNV within NSW), and the recording of human disease notifications. Sentinel animals can be placed into potential ‘hotspots’ of virus activity and, as they are continuously exposed to mosquito bites, can indicate activity in a region before human cases are reported. Seroconversions in sentinel flocks provide evidence that the level of virus in mosquito populations is high enough for transmission to occur.

The monitoring of human cases of arboviral infection usually has little direct value for surveillance, as by the time the virus activity is detected in the human population, often not much can be done to control the viral transmission. Via the other methodologies, the aim of the surveillance program is to recognise both potential and actual virus activity before it impacts greatly on the human population, so that appropriate preventive measures can be implemented. The recording of human infections does, however, provide important epidemiological data and can indicate locations where surveillance should occur.

These methods of surveillance are listed in order; generally, with more rainfall comes more mosquito production; the higher the mosquito production, the greater the probability of enzootic virus activity in the mosquito/host population; the higher the proportion of virus infected hosts and mosquitoes, the greater the probability of transmission and thus the higher the risk to the human population. The NSW Arbovirus Surveillance and Mosquito Monitoring Program undertakes the first four methods of arbovirus surveillance and the results for the 2011-2012 season follow.

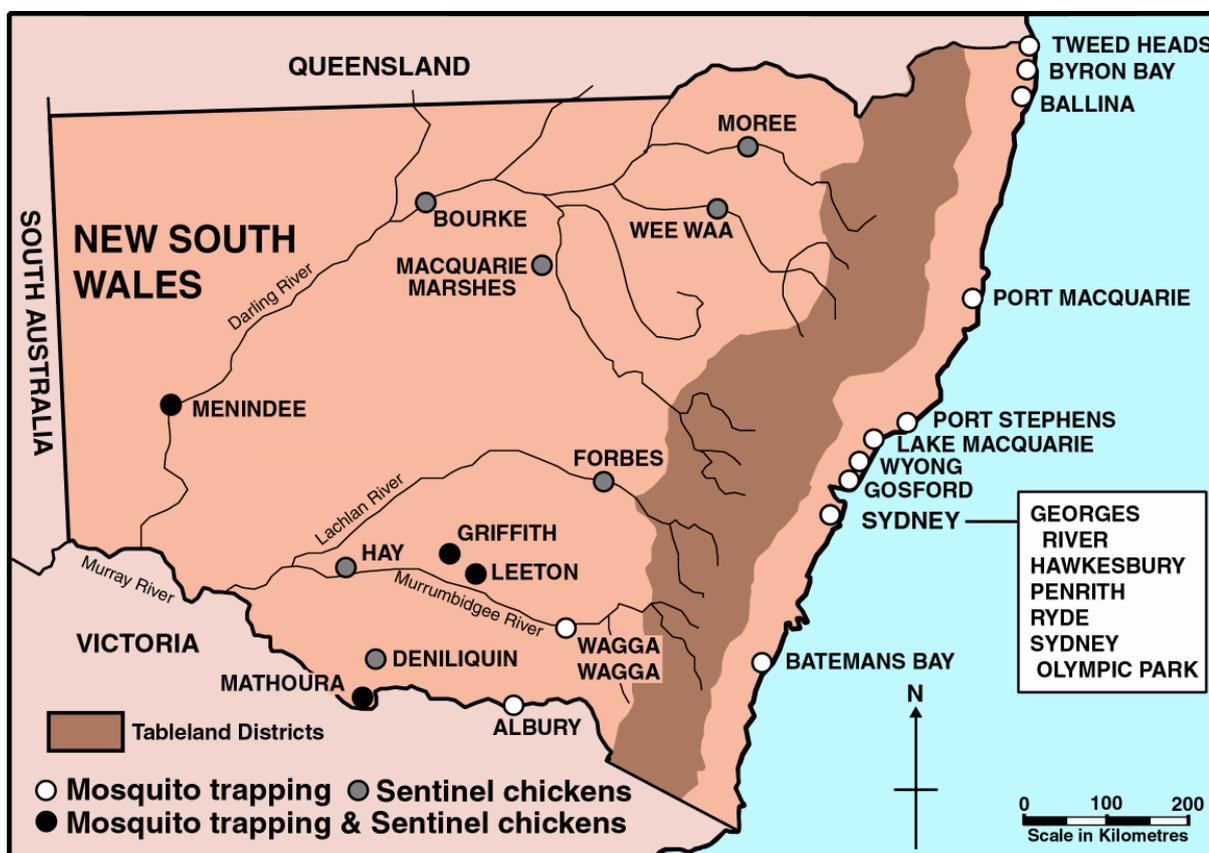


Fig 1. Mosquito trapping locations and Sentinel Chicken sites, 2011-2012.

MONITORING LOCATIONS

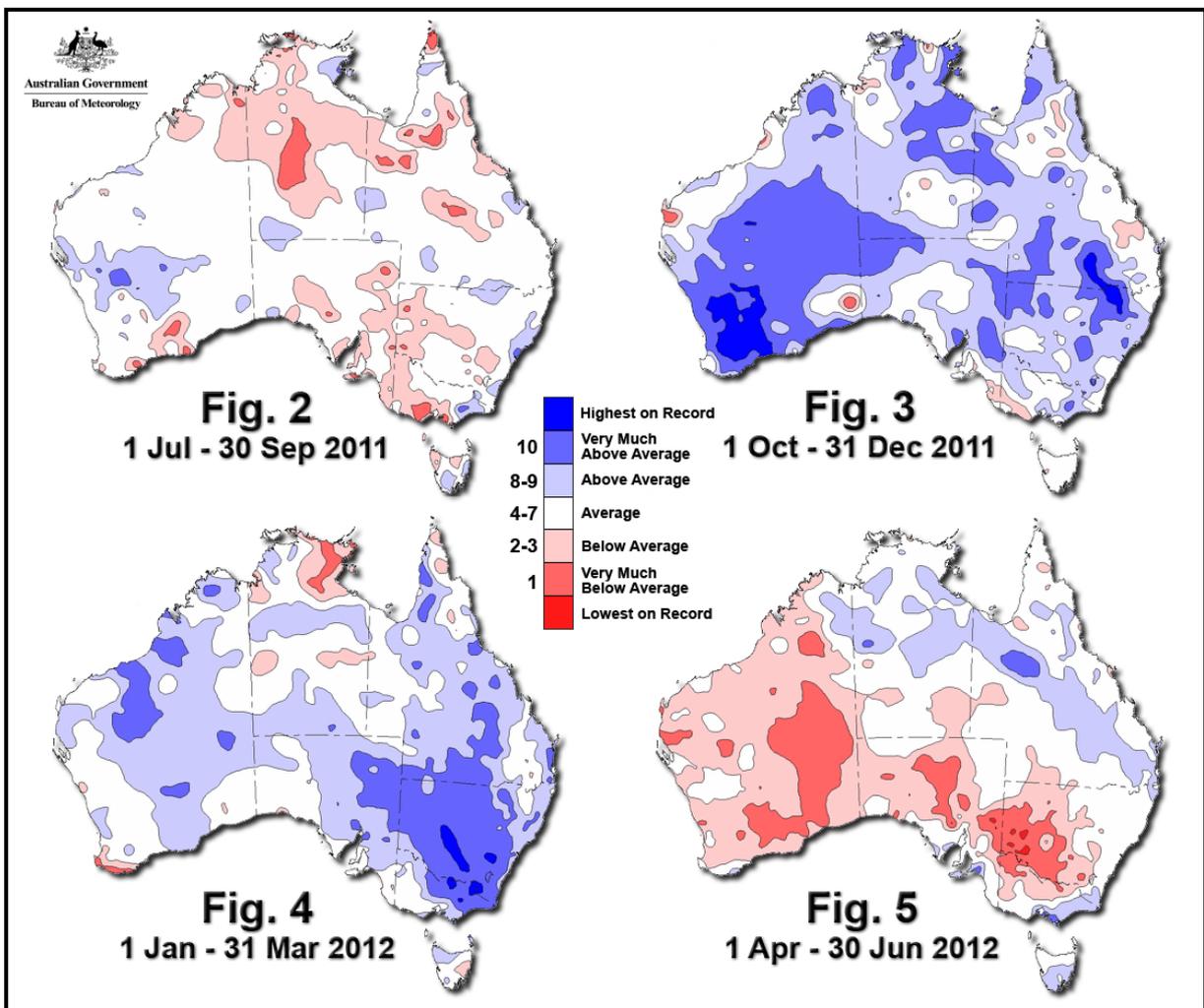
<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/location/locations.htm>

For 2011-2012, mosquito-trapping sites were operated at 6 inland, 9 coastal and 5 Sydney locations (Fig 1). Chicken sentinel flocks were located at 11 locations.

WEATHER DATA

<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/climate/climate.htm>

Mosquito abundance is dictated principally by rainfall patterns and irrigation practices in inland regions, while in coastal regions tidal inundation along with rainfall is important. Temperature and/or day-length are often critical in determining the initiation and duration of mosquito activity for species in temperate zones. Hence, the monitoring of environmental parameters, especially rainfall, is a crucial component of the Program.



Figures 2-5. Australian Rainfall deciles for the three month periods, Jul-Sep 2011, Oct-Dec 2011 Jan-Mar 2012 & Apr-Jun 2012. The stronger the red, the drier the conditions. Conversely, the stronger the blue, the wetter the conditions. *Modified from the Australian Bureau of Meteorology, 2012.*

The first quarter of 2011 saw rainfall that was above to very much above average across southeastern and central Australia, and record rainfall levels were experienced along the Murray. For the three months of April to June 2011, conditions were quite dry and precipitation levels were below average for much of NSW. Similar conditions prevailed during the third quarter of 2011 with most of the inland of NSW having below normal rainfall, while the north coast had above average precipitation (Figure 2). The last quarter of 2011 saw a return to much wetter conditions, with most of the state, and inland regions of the country, experiencing rainfall that was above, to very much above, average (Figure 3). The first quarter of 2012 was extremely wet for NSW, with most of the inland having very much above average precipitation and for some areas it was even the highest on record. Much of this rain fell during March and there was associated widespread flooding across the inland. The entire coastal strip also was very wet during these three months (Figure 4). The second quarter of 2012 experienced drier rainfall patterns and the inland had mostly below, to very much below, average precipitation (Figure 5).

Temperatures for the last half of 2011 were above average until December and thereafter below average for the remainder of the mosquito season due to the increased cloud cover with the greater rainfall.

MVEV Predictive Models

Two main models have been developed for the prediction of MVEV epidemic activity in southeastern Australia: the Forbes (1978) and Nicholls (1986) hypotheses.

Forbes associated rainfall patterns with the 1974 and previous MVEV epidemics, and discussed rainfall in terms of 'decile' values. A decile is a ranking based on historical values. The lowest 10% of all rainfall values constitute decile 1, the next 10% make up decile 2, and so on to the highest 10% of rainfall constituting decile 10. The higher the decile, the greater the rainfall.

The Forbes hypothesis refers to rainfall levels in the catchment basins of the main river systems of eastern Australia. These include:

- The Darling River system,
- The Lachlan, Murrumbidgee & Murray River systems,
- The Northern Rivers (that lead to the Gulf of Carpentaria), and
- The North Lake Eyre system.

The hypothesis states that if rainfall levels in these four catchment basins are equal to or greater than decile 7 for either the last quarter of the previous year (e.g. October-December 2010) or the first quarter of the current year (January-March 2011) and the last quarter of the current year (October-December 2011), then a MVEV outbreak is probable. By comparing the relevant quarterly rainfall amounts with historical decile 7 years, it is possible to obtain a ratio; a figure of 1 or greater indicates that rainfall was above the historical decile 7 average (Table 1). Rainfall was above decile 7 for all of the catchment basins for the last quarter of 2010, and also for first and last quarters of 2011, thus the Forbes hypothesis was fulfilled for the season of 2011-2012 (Table 1). At the time of writing this report in July 2012, all the catchment basins have received rainfall greater than decile 7 for the last quarter of 2011 and almost the first quarter of 2012, thus the model is suggesting a possible

risk of an MVEV epidemic for the upcoming 2012-2013 season.

Table 1. Rainfall indices for the main catchment basins of eastern Australia as per Forbes hypothesis, relevant to the 2011-2012 and 2012-2013 seasons.

Catchment Basin	Oct-Dec 2010	Jan-Mar 2011	Oct-Dec 2011	Jan-Mar 2012
Darling River	1.72	1.01	1.30	1.29
Lachlan/Murrumbidgee/Murray Rivers	1.99	2.02	1.08	3.10
Northern Rivers	1.80	1.38	1.19	0.95
North Lake Eyre system	2.28	1.39	1.37	1.01

The Nicholls hypothesis uses the Southern Oscillation (SO) as a tool to indicate a possible MVEV epidemic. Typically atmospheric pressures across the Pacific Ocean tend to be low on one side of the ocean and high on the other. This pattern then oscillates from year to year. Nicholls noted a correlation between past outbreaks of MVEV and the SO (as measured by atmospheric pressures at Darwin) for the autumn, winter and spring period prior to a disease outbreak. For the autumn, winter and spring periods of 2011, the SO values were respectively: 1009.07mm, 1013.43mm and 1010.50mm (indicated on Figure 6 by the yellow arrows). The graph on the right has been modified (i.e. updated) to include those MVEV active years between 2000 and 2012 (added to the MVEV tallied black columns), and includes the values for the years 2000-2001, 2007-2008, 2010-2011 and 2011-2012. The SO values leading up to the 2003-2004 season were not included as there was only one detection of MVEV, which may have resulted from over-wintering mosquitoes. As of August 2012, the autumn Nicholls' value for 2012 is 1009.43mm and within the range of values during past MVEV outbreak years, while the incomplete winter value of 1013.30 is also inside the range.

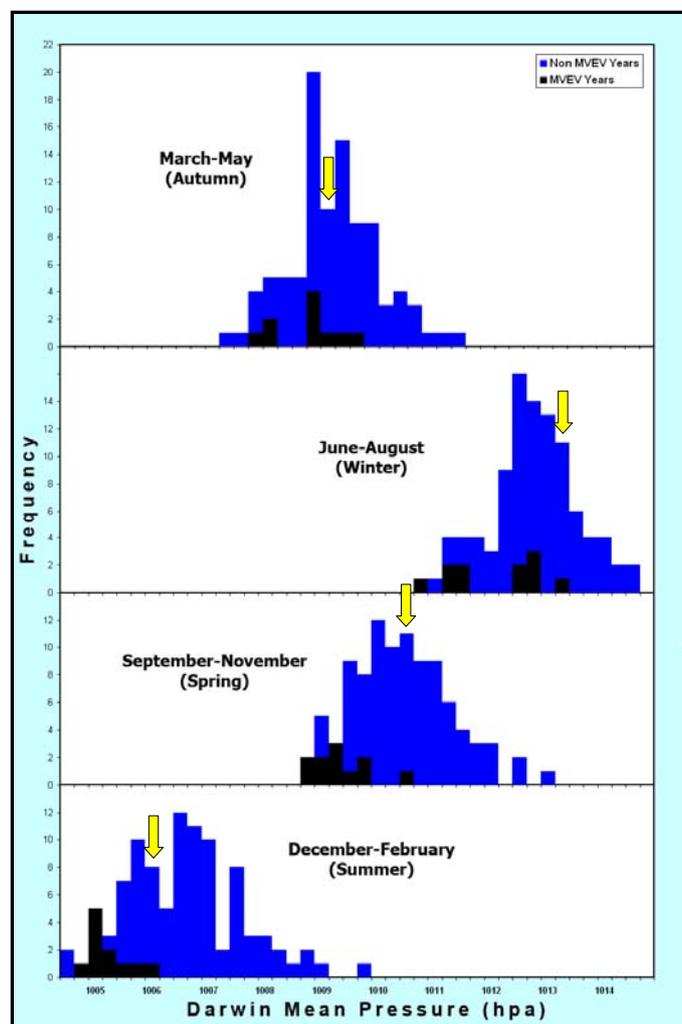


Figure 6. The SO by seasons prior to MVEV active years, according to Nicholls (1986), updated up to Autumn 2012. The black bars represent the pre-MVEV active seasons. The yellow arrows indicate the respective SO values relevant to the 2011-2012 season.

It is important to note that the Forbes hypothesis was calculated on environmental conditions experienced during major MVEV epidemic seasons and the models do not propose to predict low to moderate level activity. Thus, negative MVEV models do not necessarily indicate an absence of MVEV activity. Also, these climatic based models do not take into account unusual environmental conditions such those experienced during the summer of 2008, whereby a low pressure cell that began in northern Australia moved through to the south and possibly facilitated the movement of MVEV into NSW (Finlaison *et al.*, 2008). A similar phenomenon may have occurred during the 2010-11 season, whereby a low pressure cell that formed from Tropical Cyclone Yasi and moved into Victoria bringing intense rainfall, coincided with major MVEV and KUNV activity (Doggett *et al.* 2011). Nor do these models take into account virus existing in cryptic foci in southeastern Australia.

MOSQUITO MONITORING

Methods

Mosquitoes were collected overnight in dry-ice baited Encephalitis Vector Surveillance type traps. They were then sent live in cool, humid Eskies via overnight couriers to the Department of Medical Entomology, Institute of Clinical Pathology and Medical Research (ICPMR), Westmead, for identification and processing for arbovirus isolation. The mosquitoes were identified via taxonomic keys and illustrations according to Russell (1993, 1996), Dobrotworsky (1965) and Lee *et al.* (1980 – 1989). A brief description of the main mosquito species for NSW appears in Appendix 2.

Mosquito abundances are best described in relative terms, and in keeping with the terminology from previous reports, mosquito numbers are depicted as:

- 'low' (<50 per trap),
- 'moderate' (50-100 per trap),
- 'high' (101-1,000 per trap),
- 'very high' (>1,000 per trap), and
- 'extreme' (>10,000 per trap).

All mosquito monitoring results (with comments on the collections) were placed on the NSW Arbovirus Surveillance Web site, and generally were available within 1-2 days of sample receipt into the laboratory. Access to each location's result is from:

<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/results/results.htm>.

Results

Overall, 241,563 mosquitoes representing 62 species were collected in NSW during the 2011-2012 season, which was slightly down upon the previous season but well over twice that of the total mosquito collection in 2009-2010. *Culex annulirostris* was the most abundant and most important of the inland mosquito species during the summer months, whereas *Coquillettidia linealis*, *Aedes notoscriptus*, *Culex annulirostris*, *Aedes vigilax*, *Culex sitiens*, and *Verrallina funerea* were the most numerous species on the coast. A full summary of the results on a location-by-location basis is included in Appendix 1 and the complete mosquito monitoring results are available on the NSW Arbovirus Surveillance web site.

Inland

The total of 170,005 was slightly below the previous season total of 207,992 trapped in 2010-2011, but was five times larger than that of 2009-2010 season's collection, and almost 15 times that of the 2008-2009 season total. *Culex annulirostris* was the dominant species yielded at most sites and comprised 68.0% of the total inland collections. *Anopheles annulipes* (17.8%) was the next most common species, with *Aedes theobaldi* (6.7%) and *Aedes vittiger* (4.1%).

Coastal

In total, 56,148 mosquitoes comprising 54 species were collected from coastal NSW and this was slightly above the previous season's total collection but well below the 2009-2010 collection of approximately 83,000. The most common species collected were *Coquillettidia linealis* (26.7% of the total coastal mosquitoes trapped), *Aedes notoscriptus* (11.5%), *Culex annulirostris* (10.9%), *Verrallina funerea* (8.3%), *Culex orbostiensis* (6.6%), *Coquillettidia xanthogaster* (6.2%), *Aedes vigilax* (5.7%), *Culex sitiens* (5.3%), *Aedes multiplex* (5.6%), *Mansonia uniformis* (3.0%), and *Aedes procax* (2.2%). For most years, *Aedes vigilax* is usually by far the most predominant species and generally comprises 50-60% of the coastal collections.

Metropolitan Sydney

A total of 15,410 mosquitoes, comprising 28 species, was collected from metropolitan Sydney and this was slightly down upon the previous season's total collection. *Culex sitiens* (21.8% of the total Sydney mosquitoes trapped) was the most common species, followed by *Aedes notoscriptus* (27.1%), *Culex annulirostris* (17.0%), *Aedes vigilax* (7.0%), *Coquillettidia linealis* (5.0%), *Anopheles annulipes* (4.5%) and *Culex quinquefasciatus* (4.1%). Similar to the coast, for most years *Aedes vigilax* tends to dominate the Sydney collections.

ARBOVIRUS ISOLATIONS FROM MOSQUITOES

<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/about/methods.htm>

Methods

Viral isolation methods were as per earlier annual reports (Doggett *et al.*, 1999, 2001). ELISA assays were used to identify any suspected viral isolate and can identify the alphaviruses - BFV, RRV and Sindbis (SINV), and the flaviviruses - MVEV, KUNV, Alfuy (ALFV), Edge Hill (EHV), Kokobera (KOKV) and Stratford (STRV). Any isolate that was not identified by the assays was labelled as 'unknown'. A short description of the various viruses and their clinical significance is detailed in Appendix 3. Positive results were sent to Dr Jeremy McNulty, Director, Communicable Diseases Branch, NSW Health, to the relevant Public Health Unit, and posted on the NSW Arbovirus Surveillance Web Site (under 'Mosquito/Chicken Results', and under each location's surveillance results).

Results

<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/results/virusisolates.htm>

From the mosquitoes processed, there were 77 viral isolates; 67 from the inland and ten from coastal locations. These are listed in Tables 2 and 3.

Table 2. Arbovirus isolates from Inland NSW, 2011-2012.

LOCATION	Date Trapped	Mosquito Species	Virus						
			RRV	SINV	EHV	KOKV	KUNV	Virus?	Tot
GRIFFITH	4-Jan-12	<i>Culex annulirostris</i>	1						1
LEETON	18-Jan-12	<i>Culex annulirostris</i>	1						1
GRIFFITH	31-Jan-12	<i>Anopheles annulipes</i>						1	1
MENINDEE	20-Feb-12	<i>Culex annulirostris</i>				1			1
BOURKE	21-Feb-12	<i>Culex annulirostris</i>				1			1
LEETON	21-Feb-12	<i>Culex annulirostris</i>				1			1
GRIFFITH	22-Feb-12	<i>Anopheles annulipes</i>	1						1
GRIFFITH	22-Feb-12	<i>Culex annulirostris</i>	3			2			5
MENINDEE	27-Feb-12	<i>Culex annulirostris</i>		5				2	7
BOURKE	12-Mar-12	<i>Culex annulirostris</i>				1			1
LEETON	13-Mar-12	<i>Culex annulirostris</i>	1			3			4
MATHOURA	13-Mar-12	<i>Aedes vittiger</i>						1	1
MATHOURA	19-Mar-12	<i>Aedes theobaldi</i>	1						1
MATHOURA	19-Mar-12	<i>Culex annulirostris</i>	4	2		2	1	1	10
MURRAY	20-Mar-12	<i>Aedes vittiger</i>						2	2
MURRAY	20-Mar-12	<i>Culex annulirostris</i>						1	1
LEETON	21-Mar-12	<i>Culex annulirostris</i>	2	6		3		1	12
GRIFFITH	26-Mar-12	<i>Aedes theobaldi</i>		1					1
GRIFFITH	26-Mar-12	<i>Aedes vittiger</i>				1			1
GRIFFITH	26-Mar-12	<i>Culex annulirostris</i>		3					3
WAGGA	26-Mar-12	<i>Culex annulirostris</i>		1					1
GRIFFITH	2-Apr-12	<i>Aedes theobaldi</i>	1					1	2
GRIFFITH	2-Apr-12	<i>Culex annulirostris</i>						1	1
LEETON	3-Apr-12	<i>Aedes theobaldi</i>	2						2
GRIFFITH	16-Apr-12	<i>Aedes theobaldi</i>						1	1
LEETON	17-Apr-12	<i>Aedes theobaldi</i>	1						1
LEETON	17-Apr-12	<i>Anopheles annulipes</i>						1	1
GRIFFITH	1-May-12	<i>Aedes theobaldi</i>			1				1
GRIFFITH	1-May-12	<i>Aedes vittiger</i>						1	1
TOTAL			18	18	1	15	1	14	67

Table 3. Arbovirus isolates from Coastal NSW, 2011-2012.

LOCATION	Date Trapped	Mosquito Species	Virus			
			RRV	EHV	Virus?	Tot
PORT STEPHENS	7-Feb-12	<i>Coquillettidia linealis</i>	1			1
WYONG	21-Feb-12	<i>Culex annulirostris</i>	1			1
RYDE	27-Feb-12	<i>Coquillettidia linealis</i>	1			1
PORT STEPHENS	28-Feb-12	<i>Coquillettidia linealis</i>		1		1
BALLINA	29-Feb-12	<i>Coquillettidia linealis</i>			1	1
PORT STEPHENS	6-Mar-12	<i>Coquillettidia linealis</i>	1			1
WYONG	19-Mar-12	<i>Culex annulirostris</i>			1	1
HAWKESBURY	21-Mar-12	<i>Aedes procax</i>			1	1
BYRON BAY	10-Apr-12	<i>Aedes vigilax</i>			1	1
BYRON BAY	24-Apr-12	<i>Aedes notoscriptus</i>	1			1
TOTAL			5	1	4	10

RRV = Ross River virus, SINV = Sindbis virus, EHV = Edge Hill virus, KOKV = Kokobera virus, KUNV = Kunjin virus, Virus? = unknown (not BFV, RRV, SINV, EHV, KOKV, KUNV, MVEV, STRV).

SENTINEL CHICKEN PROGRAM

<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/about/chickenmethods.htm>

Location of flocks

The 2011-2012 season began on 27th October 2011 with the first bleed and ended on 30th April 2012 with the last. A total of eleven flocks each containing up to 15 Isa Brown pullets was deployed, with one flock each at Bourke, Deniliquin, Forbes, Griffith, Hay, Leeton, Macquarie Marshes, Menindee, Moama (near Mathoura), Moree and Wee Waa (Figure 1).

Methods

The NSW Chicken Sentinel Program was approved by the Western Sydney Local Health Network Animal Ethics committee. This approval requires that the chicken handlers undergo training to ensure the chickens are cared for appropriately and that blood sampling is conducted in a manner that minimises trauma to the chickens. The chickens are cared for and bled by local council staff and members of the public. Laboratory staff are responsible for training the chicken handlers. A veterinarian (usually the Director of Animal Care at Westmead) must inspect all new flock locations prior to deployment to ensure animal housing is adequate. Existing flocks are inspected approximately every two years. The health of each flock is reported weekly, and is independently monitored by the Animal Ethics Committee via the Director of Animal Care.

Full details of the bleeding method and laboratory testing regimen were detailed in the 2003-2004 NSW Arbovirus Surveillance Program Annual Report (Doggett *et al.* 2004).

Results are disseminated via email to the relevant government groups as determined by NSW Health and are placed on the NSW Arbovirus Surveillance website. Confirmed positives are notified by telephone to NSW Health and Communicable Diseases Network, Australia.

Results

The season began with 164 pullets and six deaths were recorded during the program. A total of 2,660 samples was received from the eleven flocks in NSW over the six-month period in 2011-2012. This represented 5,320 ELISA tests (excluding controls and quality assurance samples), with each specimen being tested for MVEV and KUNV antibodies.

There were a number of seroconversions to MVEV and KUNV; these are listed in Table 4 below.

Table 4. Seroconversions to MVEV and KUNV in the sentinel chicken flocks, 2011-2012.

LOCATION	Date Bled	MVEV	KUNV	Total
Forbes	30-Apr-12		1	1
Hay	4-Dec-11	1		1
	11-Dec-11	1		1
Leeton	12-Dec-11	8		8
Macquarie Marshes	13-Dec-11	3		3
Moama	7-Dec-11	1		1
Total		14	1	15

HUMAN NOTIFICATIONS

<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/human/human.htm>

The notification of human arboviral infections is based on laboratory notifications, which define cases as being 'confirmed', 'presumptive', 'inconclusive' or 'negative' (Mackenzie *et al.* 1993). A 'confirmed' infection is where there is at least a fourfold rise or fall in antibodies between paired sera, with the first blood sample begin taken early in the disease phase (the 'acute' sample) and the second sample taken during convalescence of the illness (the 'convalescent' sample). The detection of the virus by isolation or through molecular techniques also constitutes a 'confirmed' infection. A 'presumptive' infection is where there is IgM antibody in the acute sera, or moderate or high antibody (such as IgG) with IgM antibodies. An 'inconclusive' infection has little to no IgM antibody in the acute sample or stable antibody levels in two convalescent samples without IgM antibodies. A 'negative' infection has no specific arbovirus antibody.

Table 5. Arbovirus notifications according to former Area Health Service, July 2011 - June 2012*.

Month	CS	NS	WS	WE	SW	CC	HU	IL	SE	NR	MN	NE	MA	MW	FW	GM	SA	Total
RRV	6	7	4	10	5	14	86	7	0	85	33	67	29	32	33	89	12	519
BFV	0	3	0	0	2	9	56	12	2	133	38	8	8	3	7	12	8	301
Total	6	10	4	10	7	23	142	19	2	218	71	75	37	35	40	101	20	820

CS = Central Sydney, NS = Northern Sydney, WS = Western Sydney, WE = Wentworth, SW = South Western Sydney, CC = Central Coast, HU = Hunter, IL = Illawarra, SE = South Eastern Sydney, NR = Northern Rivers, MN = Mid North Coast, NE = New England, MA = Macquarie, MW = Mid Western, FW = Far Western, GM = Greater Murray, SA = Southern Area. *Data from 'GODSEND'.

Table 5 contains the number of laboratory notifications of human RRV and BFV infection by former Area Health Service (AHS) for NSW. The former AHSs data were used, rather than the current, to allow for a comparison of notification trends over time. Also note that the majority of notifications are 'presumptive' infections. As a result there are likely to be significant errors in the data given the high false positive rate of commercial kits (20% false positives, L. Hueston, *pers. comm.*), the degree of

cross-reactivity of closely related arboviruses, the persistence of IgM for long periods (18 to 48 months) in genuine infections, and the fact that antibody is produced regardless of clinical disease (L. Hueston, *pers. comm.*). In an investigation of serologically diagnosed BFV cases from the mid-north coast of NSW, it was found that there was a significant amount of over-diagnosis (Cashman *et al.* 2008), which appears to be continuing.

The total number of RRV and BFV notifications for the period July 2011 to June 2012 was 820 and included 301 BFV and 519 RRV. This season had one of lowest totals of notifications, the fourth lowest since that of 1994/95, and well below the long term average of 1,140. The coastal region accounted for 493 (60.1% of the state total) of the BFV and RRV notifications, which was well below the seasonal average of 737. The 288 notifications (35.1% of the state total) from the inland were slightly below the average of 322. Within the Sydney region there were 39 cases reported, less than half that of the seasonal average (81 notifications).

From the coast, the Northern Rivers and Hunter Health Services received the highest notifications (218 and 142 respectively) with the New England having 75 and the Mid-North Coast having 125. Combined, these four areas accounted for 61.7% of all the arbovirus notifications for the state. From the inland, the Greater Murray AHS had the highest number of notifications (101), with the Far West having 40.

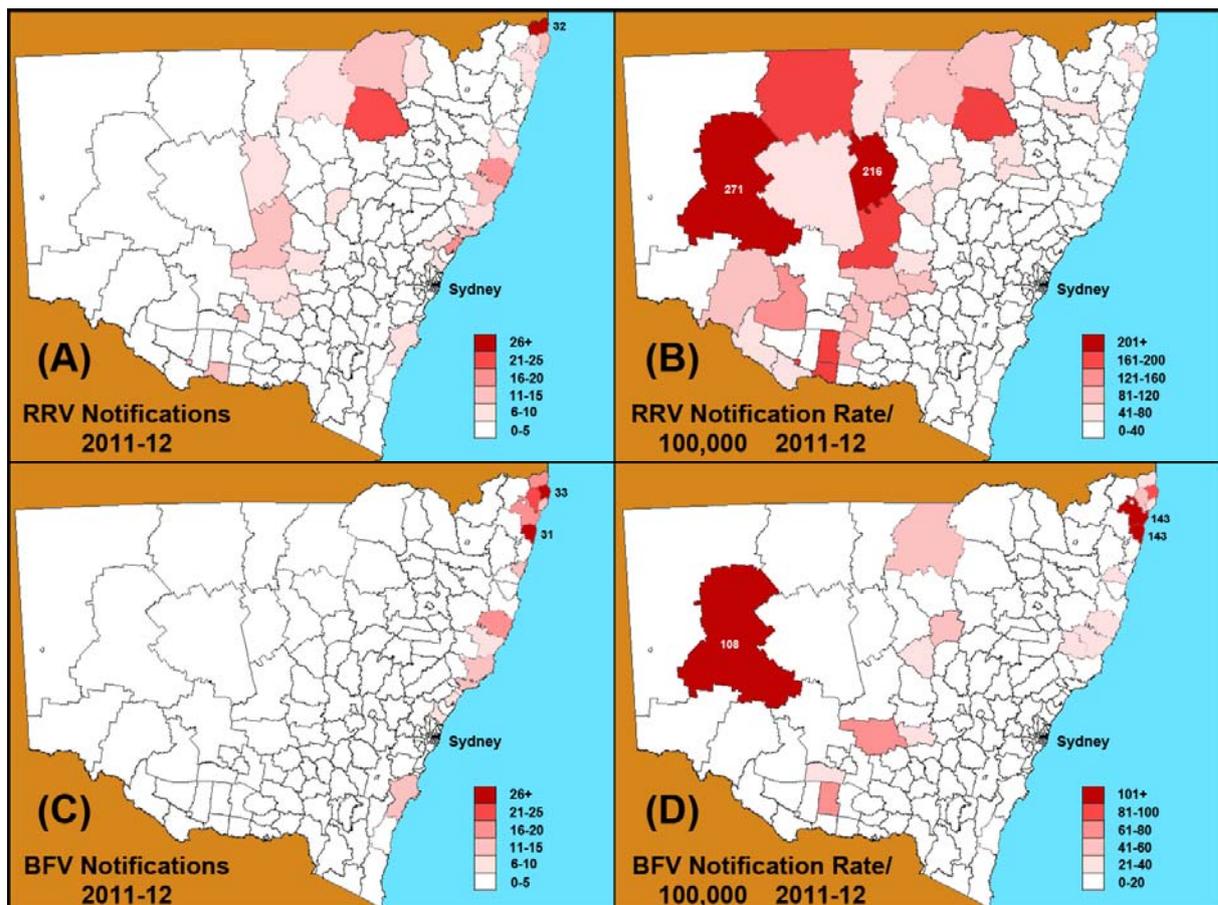


Figure 7. Notifications and notification rates of RRV and BFV by Statistical Local Areas for NSW for Jul 2011 to Jun 2012. (A) RRV notifications. (B) RRV notification rate/100,000 population. (C) BFV notifications. (D) BFV notification rate/100,000 population. Note that different scales are used on each graph. Data from 'GODSEND'.

Figure 7 depicts the notifications and notification rates of RRV and BFV by Statistical Local Area (SLA) for NSW during the 2011-2012 mosquito season.

There was one MVEV notification from northern inland NSW and one KUNV notification from the NSW south coast.

DISCUSSION

The Inland. The extremely heavy rainfall throughout inland NSW during the second half of 2011 and first quarter of 2012 resulted in a massive explosion in mosquito populations, and considerable arbovirus activity. With the heavy rainfall and the MVEV activity of the previous season there was an increased level of surveillance, particularly following the early MVEV detections in the sentinel chickens. Over 170,000 mosquitoes were trapped during the season, which was slightly down upon the 2010-2011 total when over 200,000 were yielded; however, the current seasonal total was five times larger than the collection made in 2009-2010 and almost 15 times that in the season of 2008-2009. At some sites there was a rapid explosion in mosquito populations; numbers at Menindee jumped from 12 to over 8,000 in one week during mid-February, while at Moama, mosquito collections went from 37 to over 4,000, also in one week, albeit in mid-March. Around this time, Councils and Public Health Units in the affected areas received numerous complaints about mosquitoes. There were 67 isolates from the mosquitoes, including 18 RRV, 18 SINV, one EHV, 15 KOKV, one KUNV and 14 unidentified. There were 14 seroconversions in the sentinel chicken flocks including 14 MVEV and one KUNV.

The MVEV seroconversions occurred over a limited period in early December, between 4-13/Dec/11, albeit in the quite disparate areas of Hay, Leeton, Macquarie Marshes and Moama. All of these sites had MVEV seroconversions in the previous season except Hay, which operated no flocks at the time. There was also one human case from northern inland, which was contracted around the same period. Interestingly, despite widespread MVEV activity along the Murray in 2010-2011 with 46 seroconversions, the Victorian arbovirus program did not detect any activity this season (Lynch *et al.*, 2012). The KUNV activity was much later in the season; the isolate from Griffith occurred from mosquitoes trapped on 19/Mar/12, while the seroconversion from Forbes was detected from the bleed taken on 30/Apr/12, the bleed from the 26/Apr being negative. The KUNV at Forbes coincided with a horse case of KUNV that was reported on 2/May/12 (NAMAC update, 23/May/12). The Victorian arbovirus surveillance program also had two KUNV seroconversions during April in their sentinel chickens, one each at Barmah and Nyah West (Lynch *et al.*, 2012). No human cases were reported from inland NSW. The other flavivirus that was particularly active was KOKV, with 18 isolates occurring over a widespread region including; Bourke, Menindee, Griffith and Leeton. While no human cases were reported from NSW, there was a single case in Queensland (no details available, NAMAC Update, 23/May/12).

As per the previous season, there was a high number and proportion of floodwater *Aedes* collected, notably *Aedes theobaldi* and *Aedes vittiger*. Viruses such as MVEV are known to survive in the eggs of floodwater *Aedes* species, which is a mechanism by which the virus can be maintained from one mosquito season to the next (Broom

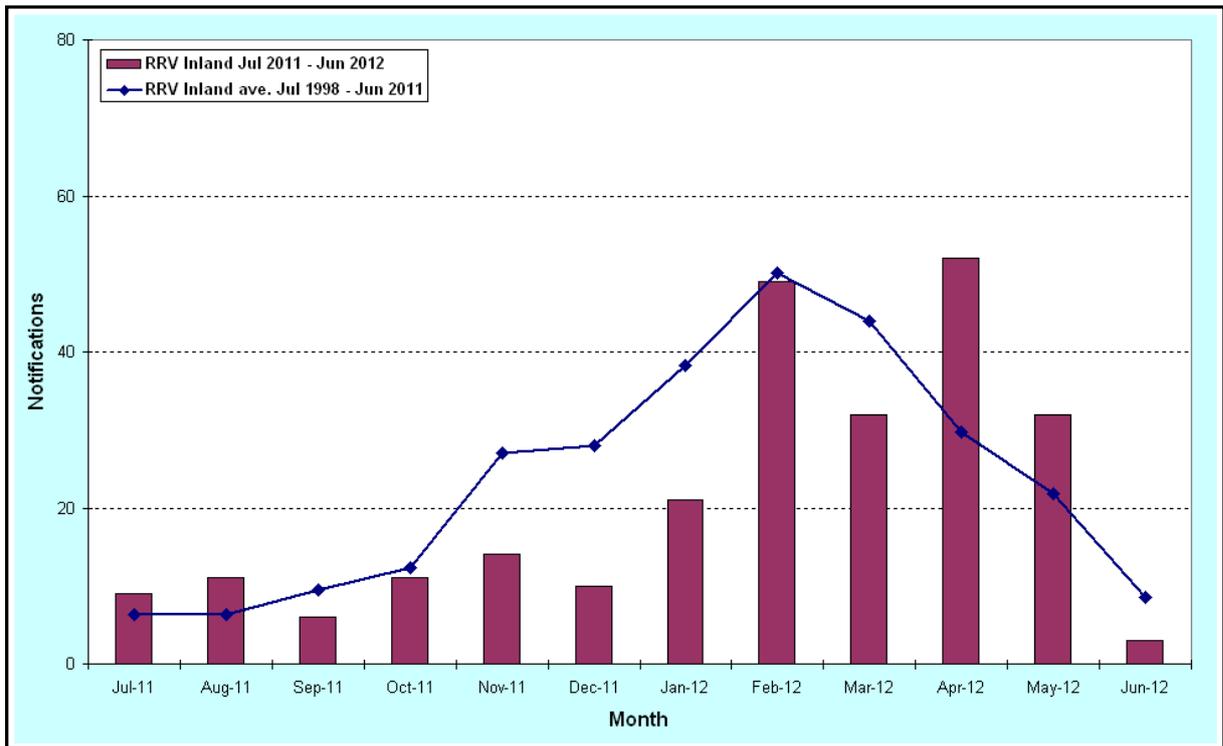


Figure 8. Notifications of RRV per month from inland NSW. The bars are for 2011-2012 season and the line represents the long term average. Data from 'GODSEND'.

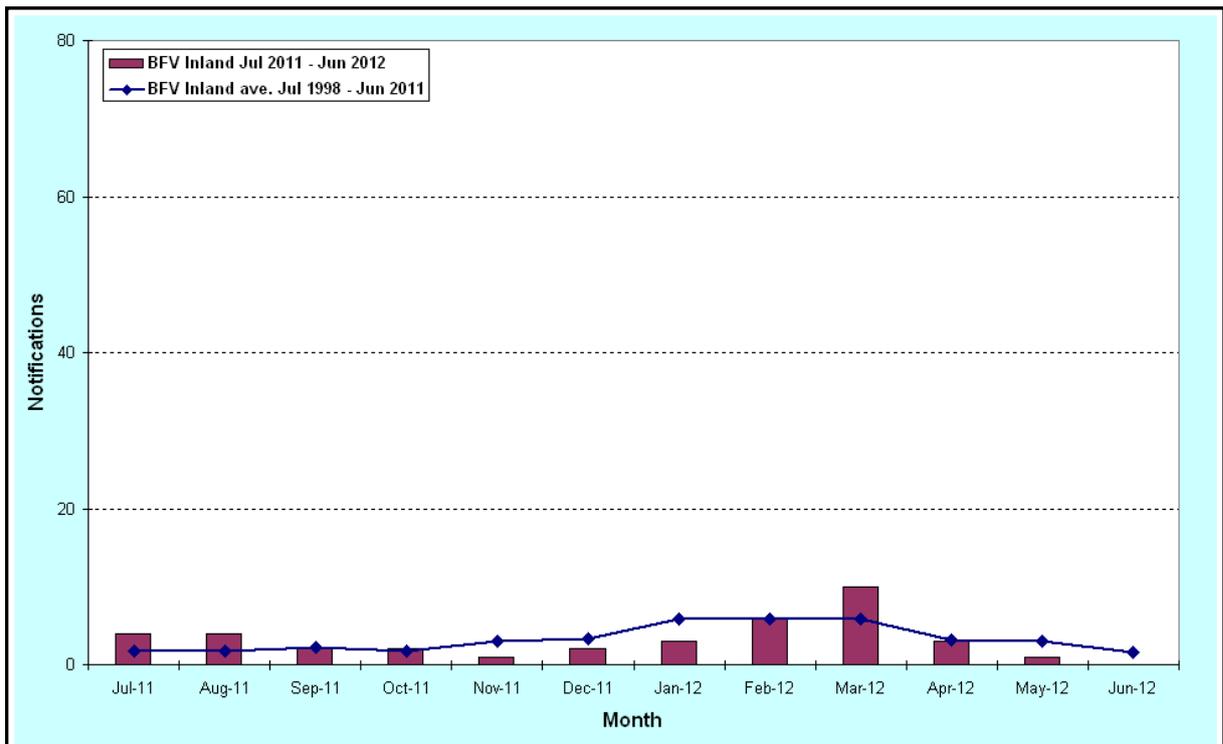


Figure 9. Notifications of BFV per month from inland NSW. The bars are for 2011-2012 season and the line represents the long term average. Data from 'GODSEND'.

et al. 1995). This may have happened in late 2003 when there was one seroconversion in the sentinel chicken flock to MVEV at Menindee despite two concurrent seasons of dry weather (Doggett *et al.* 2004). As all the seroconversions this season were in flocks that demonstrated activity last season, perhaps the virus again overwintered in *Aedes* eggs, and may not have been reintroduced to the

region. As there was again high numbers of *Aedes* in this season's collection, the risk of MVEV recurring in 2012-2013 must be considered as a possibility.

For the MVEV climatic based models, the Forbes hypothesis is indicating probable activity for the 2012-2013 season. Additionally for the modified Nicholls theory, both the Autumn values and the incomplete winter value are in line with past pre-MVEV active years. In light of the activity over the last two seasons, the possibility of overwintering virus must be considered. Recently, however (as of 31/July/12), the Bureau of Meteorology announced that current climatic indicators are suggesting a possible El Niño event (BOM 2012b). If this was to occur (and the indicators are close to threshold values), then a drier summer is likely, which should lead to reduced vector abundance across the inland.

Eighteen RRV isolates were made from the inland during the course of the season, and were temporally spread, occurring over 4/Jan to 17/Apr/12. All came from either Griffith or Leeton, most (12) from *Culex annulirostris*, five from *Aedes theobaldi* and one from *Anopheles annulipes*. Normally 18 RRV isolates is a high number and would be associated with elevated human notifications; however, the number of RRV reported cases was slightly lower than average this season. The two prior seasons had case loads considerably above average, and the lower notifications this season may just represent 'herd immunity' effects, namely when there is recent activity of an infectious agent within a community a number of individuals will become immune; this can reduce the potential and size of subsequent outbreaks. The SLAs that produced most RRV cases (Figure 7a) included Narrabri (25 notifications, this was also the highest for the 2010-2011 season), Moree Plains (15), Berrigan (15), Deniliquin (13), Broken Hill (12), Lachlan (12), Leeton (11), Dubbo (9) and Walgett (8). The highest rates (Figure 7b) were from the SLAs of Central Darling (271 cases/100,000 population), Bogan (216), Jerilderie (191), Narrabri (190), Lachlan (179), Berrigan (178), Deniliquin (167), and Bourke (162). Interestingly the top 19 highest notification rates this season for RRV were again all from the inland, indicating the greater risk of arboviral disease for communities from inland regions of NSW.

The season of 2010-2011 saw the largest outbreak of BFV for the inland region of NSW to date. For 2011-2012, no isolates were made and there were few human notifications, only 38 (Figure 7c&d, Figure 9, Table 6). As mentioned under the Coastal discussion below, it is probable that there is considerable misdiagnosis of this condition. There were also 18 SINV isolates and no human cases were reported.

The Coast. Arbovirus activity along the coast was again relatively low for the season of 2011-2012. There were only six identified isolates: five RRV, one EHV, plus four unknowns. Human notifications were also well down; the 493 reported cases (299 BFV and 264 RRV) was over 30% lower than the long term average of 737 (Table 6). Notifications of RRV were down by around 20% (256 RRV notifications this season compared with the average of 309, Figure 10), while BFV reports were almost half (237 versus the average of 428, Figure 11), with much fewer cases of both diseases reported during normal peak incidence.

The graph of BFV notifications over this season (Figure 11) suggests that newly acquired cases of human BFV disease were consistent throughout the year. However, this data makes no epidemiological sense. During the cooler months of the

year, mosquito vectors are largely inactive and so the risk of viral transmission is minimal. In a recent study reviewing notified human cases of BFV from mid north coast NSW, it was found that false positive rates were very high (19%), while only 4/37 (10.8%) of the patents had confirmed serology compatible with recent infection (Cashman *et al.* 2008). It is not known if the errors relate to poorly performing serological tests, delays in notification reporting, patients being tested without good clinical suspicion of disease (thus inapparent infections are being detected), or a combination of these or other factors. With a potentially large error rate, epidemiological interpretation of the notification data is virtually impossible.

Table 6. Notifications of BFV & RRV infection* per virogeographic regions of NSW, per season from 1994-1995 to 2011-2012 (*after Doggett 2004, Doggett & Russell 2005*).

Season	BFV				RRV			
	Coastal Cases ¹	Inland Cases ²	Sydney ³	Total	Coastal Cases ¹	Inland Cases ²	Sydney ³	Total
94/95	233	8	7	248	163	45	14	222
95/96	141	9	3	153	399	511	32	942
96/97	155	19	16	190	731	566	250	1,547
97/98	103	14	2	119	162	129	41	332
98/99	208	26	8	242	575	522	117	1,214
99/00	158	22	6	186	359	341	43	743
00/01	367	18	3	388	432	218	115	765
01/02	371	14	11	396	135	73	6	214
02/03	407	21	6	434	395	57	10	462
03/04	303	26	6	335	417	176	41	634
04/05	394	33	9	436	327	87	23	437
05/06	536	58	20	614	730	419	119	1,268
06/07	504	47	38	589	428	196	52	676
07/08	471	49	17	537	638	453	105	1,196
08/09	355	38	10	403	614	275	63	952
09/10	246	41	6	293	511	493	119	1,123
10/11	299	112	38	424	264	349	25	638
11/12	256	38	7	301	237	250	32	519
Total	5,507	593	213	6,313	7,517	5,160	1,207	13,884
Ave⁴	309	33	12	354	428	289	69	786

¹Represents the former Area Health Services of CC, HUN, ILL, MNC, NR and SA. ²Represents the former Area Health Services of FW, GM, MAC, MW and NE. ³Represents the former Area Health Services of CS, NS, SES, SWS, WEN and WS. ⁴This is the fourteen season average of 1994/95 to 2010/11. *Data from 'GODSEND'.

Despite the lower viral activity, rainfall on the coast was well above average for the 2011-2012 season, with almost continual rainfall through the early part of 2012. For many regions, more rainfall results in greater mosquito number, which in turn leads to greater arbovirus activity and more human cases. In contrast, mosquito numbers were well down, especially that of *Aedes vigilax*, the saltmarsh mosquito. This season, *Aedes vigilax*, which is the main arboviral vector, only comprised 5.7% of all mosquitoes trapped from the coast when normally this figure is around 50-60%. Continual rainfall can prevent the saltmarsh habitat from undergoing the usual tidal inundation and drying cycle which is so important for egg maturation in the species.

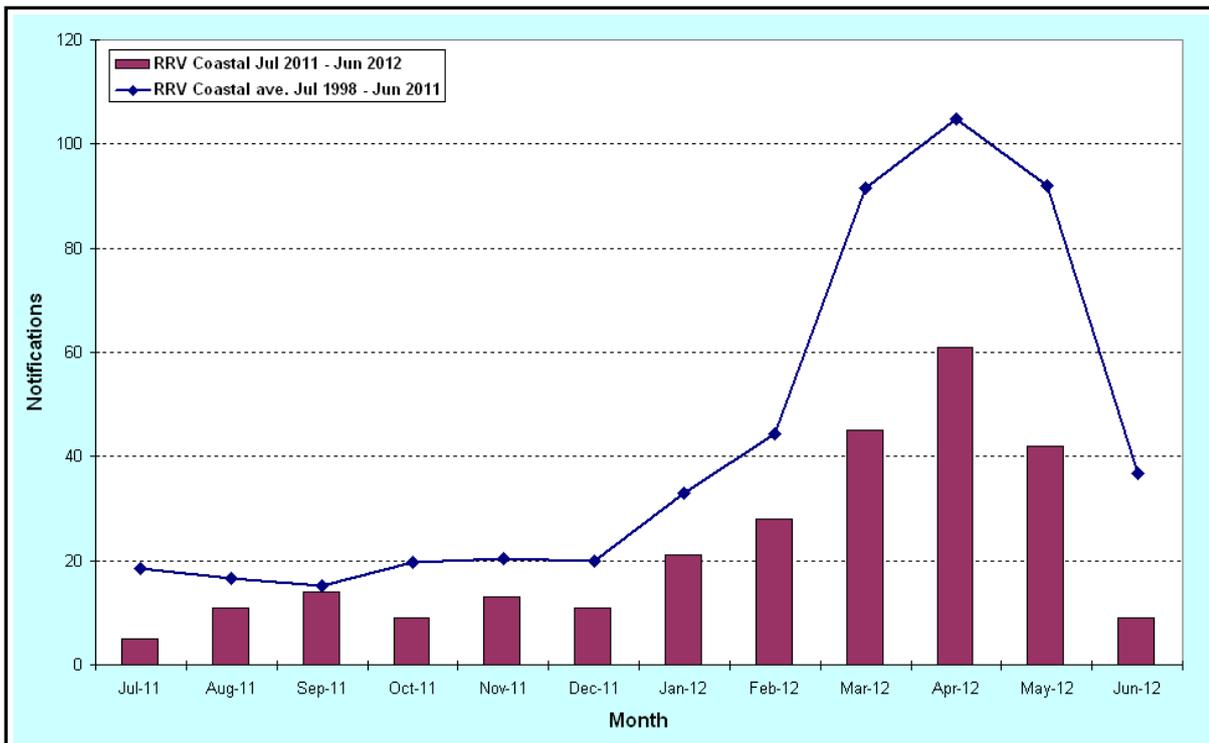


Figure 10. Notifications of RRV per month from coastal NSW. The bars are for 2011-2012 season and the line represents the long term average. Data from ‘GODSEND’.

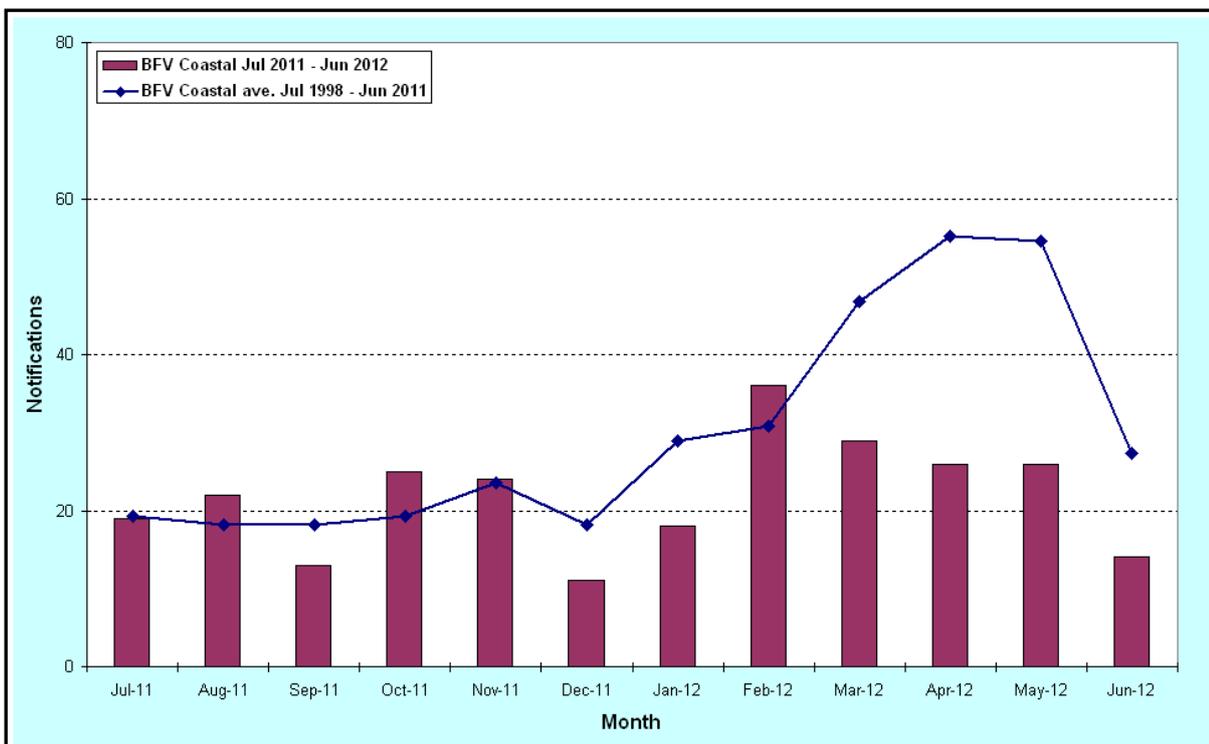


Figure 11. Notifications of BFV per month from coastal NSW. The bars are for 2011-2012 season and the line represents the long term average. Data from ‘GODSEND’.

The continual wetting of the habitat may have allowed predators to access the marshes thereby preventing the populations of the species from building up as per normal years. As *Aedes vigilax* is the major coastal vector, the lower numbers are presumably one of the main reasons for the reduction in human notifications this season. With the increased precipitation, it was the freshwater mosquitoes that

predominated including *Coquillettidia linealis*, *Aedes notoscriptus* and *Culex annulirostris*.

In terms of overall notifications for SLAs along the coast, Tweed had the greatest number with 52 (20 BFV & 32 RRV), followed by Byron (33 BFV & 13 RRV) and Maclean (31 BFV & 7 RRV, Figures 7a&c). In relation to notification rates (Figures 7b&d), Richmond River was the highest for the coast with 210/100,000, followed by Maclean (175), and Byron (139).

In early 2011 there was a major outbreak of KUNV in horses along the coast (and inland) that resulted in numerous infections and a series of deaths. This season there was no widespread activity (one case was reported in a horse which is discussed under the Sydney summary); however, there was one human case from the south coast of NSW. This is the first ever case of KUNV disease anywhere from the coastal strip and it is unknown if the virus will continue to persist on the coast and present an ongoing human health risk.

The KUNV activity over the last two years follows the isolation of KOKV for the first time from coastal NSW in 2009-2010. The mechanism of virus transferral to the coast is presently unknown, although in most years there was extensive activity of each virus across the inland. In light of this, the possibility of MVEV occurring along the coast in the future can not be readily dismissed, which highlights the need for ongoing virus surveillance in regions considered otherwise non-endemic. Recently, it was found that species of waterbirds thought to be reservoirs of MVEV can cross the Great Dividing Range (GDR) into southern Victoria (Guay *et al.* 2012) and this is a means by which the virus could be transferred to populated regions. For NSW, the GDR is much higher and broader than central Victoria and probably represents a greater barrier to migrating birds; presently it is not known the extent to which birds cross to the NSW coast.

If the predicted El Niño event does occur (BOM 2012b), then the concomitant reduced rainfall is likely to be more favourable for *Aedes vigilax* production. In light of the relatively low activity over the last two seasons, increased RRV and/or BFV activity may occur.

Sydney. For the Sydney region, five trapping sites operated over 2011-2012. Total mosquitoes trapped were slightly down upon the previous season; however, there were six sites undertaking monitoring in 2010-2012. Like the remainder of the coast, *Aedes vigilax* numbers were a record all time low, comprising only 7% of all the species trapped. Normally, this mosquito dominates the collections and tends to comprise around 50% of the yield.

With the major coastal vector being in low densities, it is not surprising that human notifications were well down upon normal; the 39 cases (32 RRV & 7 BFV) being less than half of the long term average of 81 (69 RRV & 12 BFV). How many of these were locally acquired is unknown and it is likely that many of the patients became infected elsewhere in the state in the more hyperendemic regions.

There was one isolate of RRV from *Coquillettidia linealis* collected at the Lambert Park site within Ryde. As this isolate was from a heavily populated region of Sydney,

in the subsequent weeks all the mosquitoes were processed for virus isolation and no further isolates were made.

As noted above, there was a major outbreak last season of KUNV and numerous cases in horses were reported from Western and South Western Sydney. There was also an isolate made from the Baulkham Hills region of Lower Portland (Doggett *et al.* 2011). No isolates were detected from the Sydney region this season; however, there was one case of KUNV in a horse from Richmond in Western Sydney which was reported on 2/May/2012 (NAMAC update, 23/May/2012).

THE NEW SOUTH WALES ARBOVIRUS SURVEILLANCE WEB SITE

<http://www.arbovirus.health.nsw.gov.au/>

The NSW Arbovirus Surveillance web site was established in early 1999 to facilitate the rapid dissemination of surveillance results (Doggett *et al.*, 1999b). An additional important function is to provide information on mosquitoes and the arboviruses they transmit. Over the last year, the site has continued to grow to the current size of 281MB, and has 2,150+ pages of information.



Added to the site since the last annual report includes:

- Archived data for the 2011-2012 season,
- Weekly rainfall summaries,
- Monthly rainfall summaries, with long-term averages,
- Monthly rainfall and temperatures maps,
- Daily high tides,
- Monthly SOI updates.

Appendix 1. LOCATION-BY-LOCATION SUMMARY

<http://www.arbovirus.health.nsw.gov.au/areas/arbovirus/results/results.htm>

Inland Locations

Albury: mosquito numbers were mostly 'low' to 'medium', with only two 'high' catches, both from Kremer St during mid-summer. There were no arboviral isolates from the trapped mosquitoes. Sentinel chicken flocks did not operate at Albury.

Bourke: mosquito collections were undertaken from early January to late March, with numbers mostly varying between 'low' and 'medium', with one 'high' collection in mid-February. There were two KOKV isolates, one each in mid-February and mid-March, and both from *Culex annulirostris*. There were no seroconversions to MVEV or KUNV in the sentinel chickens.

Deniliquin: no mosquito collections were undertaken this season, and there were no seroconversions to MVEV or KUNV in the sentinel chickens.

Forbes: no mosquito collections were undertaken this season. There was one seroconversion in the sentinel flocks to KUNV from the bleed taken 30/Apr/2012.

Griffith: the overall collection of mosquitoes for this season was one of the greatest to date for any one site, with just over 91,000 trapped for the season. This was, however, slightly down upon the 2010-2011 season. Collections at Hanwood were consistently 'very high' from mid November until early April, with two notable peaks. There was a trap of just over 9,000 mosquitoes during the first week of January and a second peak of almost 19,000 mosquitoes in mid-March. The latter collection was a record for the history of the NSW Arbovirus Surveillance Program. Barren Box had lower mosquito numbers, although numbers were mostly 'very high' from mid-December to mid-February. Due to major flooding, the trap could not be set at Barren Box after early March and was relocated to Yenda, which also produced some 'very high' collections. There were a total of 29 isolates; these are listed in Table 8 below. There were no seroconversions to MVEV or KUNV in the sentinel chickens.

Hay: sentinel chicken were operated this season and there were two MVEV seroconversions; one each from the weeks of 4/Dec and 11/Dec/11. No mosquito trapping was undertaken.

Leeton: mosquito numbers from Farm 347 were well above average for the entire season and mostly 'very high' from January onwards, one collection in mid-March yielding over 8,000 mosquitoes. Almond Rd also had 'very high' numbers for most of the season after mid-January. There were 22 arboviral isolates, which are listed in Table 9 below. There were eight MVEV seroconversions, all from the bleed taken on 12/Dec/11.

Macquarie Marshes: no mosquito trapping was undertaken this season. There were three MVEV seroconversions from the bleed taken on 13/Dec/2011.

Moree: sentinel chickens were operated between 15/Feb and 27/Apr/2012, and there were no seroconversions to either MVEV or KUNV. No mosquito trapping was

undertaken.

Table 8. Arbovirus isolates from Griffith, 2011-2012.

Site	Date Trapped	Mosquito Species	Virus						
			RRV	SINV	EHV	KOKV	KUNV	Virus?	Tot
Barren Box	4-Jan-12	<i>Culex annulirostris</i>	1						1
Hanwood	31-Jan-12	<i>Anopheles annulipes</i>						1	1
Barren Box	22-Feb-12	<i>Culex annulirostris</i>				1			1
Hanwood	22-Feb-12	<i>Anopheles annulipes</i>	1						1
Hanwood	22-Feb-12	<i>Culex annulirostris</i>	3			1			4
Hanwood	19-Mar-12	<i>Aedes theobaldi</i>	1						1
Hanwood	19-Mar-12	<i>Culex annulirostris</i>	4	2		2	1	1	10
Yenda	26-Mar-12	<i>Aedes theobaldi</i>		1					1
Yenda	26-Mar-12	<i>Aedes vittiger</i>				1			1
Yenda	26-Mar-12	<i>Culex annulirostris</i>		3					3
Hanwood	2-Apr-12	<i>Culex annulirostris</i>						1	1
Yenda	2-Apr-12	<i>Aedes theobaldi</i>						1	1
Yenda	16-Apr-12	<i>Aedes theobaldi</i>						1	1
Yenda	1-May-12	<i>Aedes theobaldi</i>			1				1
Yenda	1-May-12	<i>Aedes vittiger</i>						1	1
TOTAL			10	6	1	5	1	6	29

Table 9. Arbovirus isolates from Leeton, 2011-2012.

Site	Date Trapped	Mosquito Species	Virus				
			RRV	SINV	KOKV	Virus?	Tot
Almond Rd	18-Jan-12	<i>Culex annulirostris</i>	1				1
Farm 347	21-Feb-12	<i>Culex annulirostris</i>			1		1
Almond Rd	13-Mar-12	<i>Culex annulirostris</i>	1		3		4
Almond Rd	21-Mar-12	<i>Culex annulirostris</i>	2	2		1	5
Farm 347	21-Mar-12	<i>Culex annulirostris</i>		4	3		7
Farm 347	3-Apr-12	<i>Aedes theobaldi</i>	2				2
Farm 347	17-Apr-12	<i>Aedes theobaldi</i>	1				1
Farm 347	17-Apr-12	<i>Anopheles annulipes</i>				1	1
TOTAL			7	6	7	2	22

RRV = Ross River virus, SINV = Sindbis virus, EHV = Edge Hill virus, KOKV = Kokobera virus, KUNV = Kunjin virus, Virus? = unknown (not BFV, RRV, SINV, EHV, KOKV, KUNV, MVEV, STRV).

Mathoura: trapping was again undertaken at Picnic Point and at Moama from where the sentinel chicken flock was located. Mosquito collections at Picnic Point were mostly 'low' to 'medium', with the occasional 'high' yield. Similarly, Moama produced mostly 'low' to 'medium' numbers up until mid March, where collections rose from a total of 37 (first week of March) to over 4,000 in the next week, and this was followed by a yield of almost 7,000 mosquitoes for the week thereafter. The first of the two large collections were dominated by the floodwater species, *Aedes vittiger*, while *Culex annulirostris*. There were four unknown isolates three from *Aedes vittiger* and one from *Culex annulirostris*, all were from Moama, one from mosquito collections made on 13/Mar and the rest from 20/Mar/12. There was one MVEV seroconversion in the sentinel chicken flock from the bleed taken on 7/Dec/11.

Menindee: Mosquito collections were undertaken at one site which producing variable collections throughout the season. During mid-February, collections were 'low', while the following week produced a 'very high' number of almost 8,000 mosquitoes. There were six arboviral isolates; one KOKV from *Culex annulirostris* trapped on 20/Feb/12 and 5SINV from *Culex annulirostris* trapped on 27/Feb/12. There were no seroconversions to MVEV or KUNV in the sentinel chickens.

Wagga Wagga: trapping was undertaken at two sites on nine occasions. Collections were mostly 'high' in number early in the season becoming 'medium' to 'low' during January; however, 'high' numbers were trapped in late March. There was one isolate of SINV from *Culex annulirostris* trapped on 26/Mar/12. No sentinel chicken flocks operated from Wagga Wagga.

Wee Waa: sentinel chickens were operated between 15/Mar and 25/Apr/2012, and there were no seroconversions to either MVEV or KUNV. No mosquito trapping was undertaken.

Coastal Locations

Ballina: trapping continued at the two sites of North Creek Road and Pacific Pines. North Creek Road consistently produced greater mosquito numbers and was 'high' or greater for the entire season, with two 'very high' yields, one each in mid-November and early March. The dominant mosquito species at this site varied through the year and was mainly *Coquillettidia xanthogaster*, *Aedes notoscriptus* and *Culex sitiens*. Pacific Pines also mostly had 'high' mosquito production, with *Aedes multiplex*, *Coquillettidia xanthogaster* and *Coquillettidia linealis* being the main species captured. There was one unknown isolate not BFV, RRV, SINV, MVEV, EHV, KOKV, KUNV, or STRV) from *Coquillettidia linealis* trapped at Lennox Heads on 29/Feb/12.

Batemans Bay: only two collections were made in the early part of the season, both from the Council Depot site. Mosquito numbers were 'medium'. No viruses were isolated from the mosquitoes.

Byron Bay: traps operated at Luan Court and Wirree Drive. Luan Court tended to produce 'low' mosquito numbers for most of the season. Wirree Drive yielded several 'high' collections and *Aedes notoscriptus* was the main species collected. There was one RRV isolate from *Aedes notoscriptus* trapped at Laun Court on 24/Apr and one unknown from *Aedes vigilax* also trapped at Laun Court, but on 10/Apr/12.

Gosford: two sites at Gosford were again monitored this year: Empire Bay and Killcare Heights. Mosquito numbers tended to be 'low' for the entire season at Killcare Heights, while Empire Bay produced several 'high' collections this year. Although the start of the season yielded a number of 'high' collections, these tended to be dominated by *Aedes notoscriptus*. *Aedes vigilax* was rarely the main species captured. No viruses were isolated from the mosquitoes.

Lake Macquarie: collections were undertaken from three sites: Belmont Lagoon, Dora Creek and Teralba, and mostly 'low' numbers were trapped. Some 'high' collections were yielded from Dora Creek during mid-February to mid-March, and

were dominated by *Aedes multiplex*. No viruses were isolated from the mosquitoes.

Port Macquarie: Trapping was again undertaken at the two sites of Lord St and Partridge Creek. Collections tended to be greater at the latter site and were consistently 'high' from February onwards. The mosquito traps were dominated by freshwater breeding species including *Culex annulirostris*, *Coquillettidia linealis* and *Mansonia uniformis*. No arboviral isolates were yielded.

Port Stephens: monitoring of mosquitoes was reduced to three sites this season; Gan Gan, Karuah and Heatherbrae. The collections, as per the norm, varied substantially in mosquito abundance and species composition between the sites, which reflects the diverse mosquito breeding habitats within the region. Some trapping sites for example, are near freshwater habitats, while others are near saltmarsh environments. For the 2011-2012 season, mosquito numbers tended to be down upon previous years largely due to the smaller than usual collections of *Aedes vigilax*. Gan Gan yielded mostly 'high' collections, with *Coquillettidia linealis* being the main species trapped. Karuah produced similar sized collections, although *Aedes vigilax* was more predominant. Thus mosquito numbers were 'high' throughout the season. As per usual, Heatherbrae yielded the most mosquitoes for any site along coastal NSW. There were five weeks of 'very high' collections over February to March, with one collection of almost 7,000 mosquitoes in late February. These largest collections tended to be dominated by *Coquillettidia linealis*, *Culex orbostiensis* and *Culex annulirostris*. There were three arboviral isolates all being from *Coquillettidia linealis* trapped at Heatherbrae. There were two RRV, one each from collection made on 7/Feb and 6/Mar, and one EHV from 28/Feb/12.

Tweed Heads: mosquito collections were quite variable through the season and there were a number of 'high' collections, particularly at Beltana Drive, where greater yields were made. The larger collections were all dominated by *Culex sitiens*. In contrast, *Aedes vigilax* collections were well down; the most caught in one week was only 9. No virus isolation was undertaken.

Wyong: trapping was undertaken at two sites: Ourimbah and Halekalani. Collections were consistently 'low' to 'medium' and dominated by *Aedes notoscriptus*. Occasional larger collections were dominated by *Culex annulirostris*. There was one isolate of RRV from *Culex annulirostris* trapped on 21/Feb/12, and one unknown from *Culex annulirostris* trapped on 19/Mar/12.

Sydney Locations

Georges River: trapping was again undertaken at the same three sites of Alford's Point, Lugarno and Illawong, on four occasions only. Most of the collections were 'high' in number, but usually not dominated by *Aedes vigilax* as per past seasons. No arboviral isolates were yielded.

Hawkesbury: trapping was undertaken four sites on various weeks, including at Wheeny Creek, Yarramundi, Sackville and McGraths Hill. The collections were generally 'low' to 'medium' for much of the season with some 'high' mosquito numbers early to mid-March. The larger collections were dominated by *Culex annulirostris*. There was one unknown isolate from *Culex annulirostris* trapped on

21/Mar/12.

Penrith: trapping was undertaken at the three sites of Glenmore Park, Emu Heights and Waterside, with mostly 'low' mosquito numbers being trapped. No arboviral isolates were yielded.

Ryde: as per every season Wharf Road trapped the most mosquitoes for any of the sites at Ryde as it is closer to the *Aedes vigilax* breeding ground in Homebush Bay. However, this for season, it was *Culex sitiens* that dominated this sites' collections. Maze Park had many 'high' collections and these were dominated by *Aedes notoscriptus*. Most other sites trapped mainly 'low' mosquito numbers. Virus isolation was undertaken mainly from Lambert Park and there was one RRV isolate from *Coquillettidia linealis* trapped on 27/Feb/12. Despite increased mosquito trapping and virus testing, no further isolates were yielded.

Sydney Olympic Park: mosquito monitoring at this location has been occurring for a number of years and just one site was regularly included in the processing for arbovirus surveillance. Mosquito collections varied between 'low' to 'high' throughout the season and dominated by *Culex annulirostris* and *Culex sitiens*. No arboviral isolates were yielded.

Appendix 2. THE MOSQUITOES

The following briefly details the main mosquito species collected in NSW.

	<p style="text-align: center;">The Common Domestic Mosquito, <i>Aedes notoscriptus.</i></p> <p>A common species that breed in a variety of natural and artificial containers around the home. It is the main vector of dog heartworm and laboratory studies shows it be an excellent transmitter both of RRV and BFV.</p>
	<p style="text-align: center;">The Bushland Mosquito, <i>Aedes procax.</i></p> <p>Common throughout coastal NSW. This species breeds in bushland freshwater ground. Numerous isolates of BFV have been recovered from this species and it is probably involved in the transmission of the virus.</p>
	<p style="text-align: center;">The Northern Saltmarsh Mosquito, <i>Aedes vigilax.</i></p> <p>The most important species along coastal NSW. This species breeds on the mud flats behind saltmarshes and can be extremely abundant and a serious nuisance biter. It is the main vector for RRV and BFV along the coast.</p>
	<p style="text-align: center;">The Common Australian Anopheline, <i>Anopheles annulipes.</i></p> <p>A mosquito from throughout NSW, but is most common in the irrigated region of the Murrumbidgee where it can be collected in the 1000's. Despite its abundance, it is not thought to be a serious disease vector.</p>
	<p style="text-align: center;">The Common Marsh Mosquito, <i>Coquillettidia linealis.</i></p> <p>Found throughout NSW but especially in areas with freshwater marshes such as the Port Stephens area. Both BFV & RRV have been isolated from this species and is probably involved in some transmission.</p>
	<p style="text-align: center;">The Common Banded Mosquito, <i>Culex annulirostris.</i></p> <p>The species is common in the NSW inland regions that have intense irrigation. This species is highly efficient at transmitting most viruses and is responsible for the spreading of most of the arboviruses to humans inland.</p>

Appendix 3. THE VIRUSES

Alphaviruses

Barmah Forest virus (BFV): disease from this virus is clinically similar to that of RRV disease, although BFV disease tends to be associated with a more florid rash and a shorter duration of clinical severity. This is an emerging disease and is increasingly being recognised in NSW, with around 3-400 cases annually. However, serological misdiagnosis of this condition appears to be common and there are doubts about many of the reported cases from the winter months. Despite being first isolated from an inland region, cases of BFV disease tend to occur mainly in coastal regions in NSW. The main vector in NSW is *Aedes vigilax* although other species are involved, notably *Aedes procax*. In 2010-2011 there was a small epidemic (but largest to date) from BFV across inland NSW.

Ross River virus (RRV): this virus causes RRV disease and is the most common arbovirus affecting humans in NSW and Australia. For the state, there are around 700 cases per season. A wide variety of symptoms may occur from rashes with mild fever, to arthritis that can last from months to occasionally years. The virus occurs in both inland and coastal rural regions. The main vectors are *Culex annulirostris* (inland) and *Aedes vigilax* (coast), although other mosquitoes are undoubtedly involved in the transmission of the virus as isolates have been made from many species.

Sindbis virus (SINV): this is an extremely widespread virus throughout the world and occurs in all mainland states of Australia. In contrast with Africa and Europe where outbreaks have been reported, disease from SINV is relatively uncommon in Australia; only 24 infections were notified in NSW from Jul/1995-Jun/2003 (Doggett 2004). Symptoms of disease include fever and rash. Birds are the main host, although other animals can be infected, including macropods, cattle, dogs and humans. The virus has been isolated from many mosquito species, but most notably *Culex annulirostris* in southeastern Australia.

Flaviruses

Alfuy virus (ALFV): no clinical disease has been associated with this virus and it has not been isolated from southeastern Australia.

Edge Hill virus (EHV): a single case of presumptive infection with EHV has been described, with symptoms including myalgia, arthralgia and muscle fatigue. *Aedes vigilax* has yielded most of the EHV isolates in southeast Australia, although it has been isolated from several other mosquito species. The virus is quite common, with isolates from most years. The vertebrate hosts may be wallabies and bandicoots, but studies are limited.

Kokobera virus (KOKV): only three cases of illness associated with KOKV infection have been reported and all were from southeast Australia. Symptoms included mild fever, aches and pains in the joints, and severe headaches and lethargy. Symptoms were still being reported by the patients five months after onset. This virus historically

was only known from inland regions of NSW until it was detected in a mosquito trapped from the coastal region in 2009-2010. *Culex annulirostris* appears to be the principal vector.

Kunjin virus (KUNV): disease from this virus is uncommon, with only two cases being notified from 1995-2003 (Doggett 2004). Historically, activity has been confined to the inland region of NSW where it is detected every few years; however, in the summer of 2010-2011, the virus made it to the coast, which resulted in an outbreak amongst horses with a number of animal deaths resulting. *Culex annulirostris* appears to be the main vector.

Murray Valley Encephalitis (MVEV): activity of this virus is rare in south-eastern Australia and the last epidemic occurred in 1974. However, since the year 2000 there has been five seasons when MVEV activity has been detected within the state: 2000-2001, 2003-2004, 2007-2008, 2010-2011 and 2011-2012. With the latest two seasons of activity, there were two human cases. The virus occurs only in inland regions of the state and symptoms are variable, from mild to severe with permanent impaired neurological functions, to sometimes fatal. *Culex annulirostris* is the main vector.

Stratford virus (STRV): there have been very few documented symptomatic patients, only three described to date and symptoms included fever, arthritis and lethargy. The virus has mostly been isolated from coastal NSW, particularly from the saltmarsh mosquito, *Aedes vigilax*, although recent isolates from the Sydney metropolitan area have been from *Aedes notoscriptus* and *Aedes procax*. This is a common virus, being isolated most years.

Appendix 4. ABBREVIATIONS

AHS	Area Health Service
BFV	Barmah Forest virus
BOM	Bureau of Meteorology
CC	Central Coast Public Health Unit
CS	Central Sydney Public Health Unit
EHV	Edge Hill virus
FW	Far West Public Health Unit
GM	Greater Murray Public Health Unit
GODSEND	Graphical Online Data Surveillance and Evaluation for Notifiable Diseases
HUN	Hunter Public Health Unit
IgG	Immunoglobulin G (a type of antibody)
IgM	Immunoglobulin M (a type of antibody)
ILL	Illawarra Public Health Unit
IOD	Indian Ocean Dipole
ICPMR	Institute for Clinical Microbiology and Medical Research
MAC	Macquarie Public Health Unit
MNC	Mid North Coast Public Health Unit
MVEV	Murray Valley Encephalitis virus
MW	Mid West Public Health Unit
NE	New England Public Health Unit
NR	Northern Rivers Public Health Unit
NS	Northern Sydney Public Health Unit
KOKV	Kokobera virus
KUNV	Kunjin virus
PHU	Public Health Unit
RRV	Ross River virus
SA	Southern Area Public Health Unit
SES	South Eastern Sydney Public Health Unit
SINV	Sindbis virus
SLA	Statistical Local Area
SO	Southern Oscillation
STRV	Stratford virus
SWS	Public Health Unit
TC	Tropical Cyclone
WEN	Public Health Unit
WS	Western Sydney Public Health Unit
VADCP	Victorian Arbovirus Disease Control Program
Virus?	Virus unknown (not BFV, RRV, SINV, EHV, KOKV, KUNV, MVEV, STRV)

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