APPENDIX F
Mosquito Risk Assessment
Mosquito Risk Assessment:
Ploughman’s Creek Constructed Wetlands &
Stormwater Harvesting Project

Prepared for Orange City Council
August 2009

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EXECUTIVE SUMMARY

- A Mosquito Risk Assessment (MRA) was commissioned by Orange City Council for the construction of five stormwater treatment wetlands proposed as part of the Ploughmans Creek stormwater harvesting project.

- Due to the timing of the MRA, it was not possible to undertake any onsite mosquito population sampling to assess the abundance and relative pest risk of extant mosquito populations. No existing mosquito data for the Orange district is currently available. Reference sites were used from three inland NSW locations where mosquito population data is available, Forbes, Leeton and Griffith.

- Predictions based on mosquito population data available from Forbes, Leeton and Griffith, and taking into account differences in climate, topography and landuse of the four regions, it is predicted that Orange currently experiences low levels of pest mosquito activity.

- Mosquito-borne disease caused by Ross River virus and Barmah Forest virus is a concern for communities of inland NSW. However, the number of human infections within the Orange region appears to be relatively low compared to other inland regions.

- The species that pose the greatest risk for the newly constructed wetlands are *Coquillettidia linealis*, *Culex annulirostris* and *Culex quinquefasciatus*. While each species has certain biological and ecological requirements, all three have the potential to colonize the constructed wetlands and generate large populations if favourable conditions are created. While these species may be associated with the constructed wetlands, they may not necessarily become serious pest concerns.

- To reduce the risk of mosquito production, wetlands should be designed and constructed with consideration of factors that may contribute to increase mosquito risk. A key element will be the design and maintenance of shallow water macrophyte zones as these represent the most likely components of the constructed wetlands to provide favourable conditions for mosquitoes.

- It is strongly recommended that a mosquito monitoring program be implemented over the coming years to determine change in local mosquito populations, identify any nuisance-biting species of public health importance and assist the development of any mosquito management strategies that may be required. Such a monitoring program will be key to identify, or disassociate, any actual or perceived mosquito problems associated with the wetlands or surrounding habitats.
INTRODUCTION

Mosquito risk assessments are designed to provide information on actual and/or potential mosquito populations, identify significant larval habitats and provide comments on their significance. Based on this information, and considering the nature of local mosquito habitats, the assessment should provide comments and recommendations for mosquito management strategies*. In situations where new residential and/or recreational developments are proposed, the MRA should provide information on the likely nuisance biting and public health risks posed to current and future residential communities.

Mosquitoes are a natural component of aquatic ecosystems and their presence in a wetland does not necessarily result in serious nuisance-biting or public health impacts. However, unusually large mosquito populations can be associated with constructed water bodies and, throughout inland regions of NSW, elevate risks of mosquito-borne disease during the warmer months.

As urban centres grow in many regions, an expanding residential and tourist population has increased the potential exposure of the community to nuisance-biting impacts and mosquito-borne disease risks through the transmission of arboviruses. While not life threatening, the human disease caused by these pathogens can have a significant impact on the community and have, potentially, an adverse impact on tourism, property prices and public amenity of recreational facilities.

A Mosquito Risk Assessment was carried out on the proposed constructed wetlands to be built as part of the Orange City Council’s Ploughman’s Creek stormwater harvesting project. This document is designed to determine the relative abundance of current mosquito populations and the likely impacts posed by those mosquito populations.

Given the timing of this MRA, it was not possible to undertake mosquito population surveys. As a result, information on local mosquito populations has been based on historic data from nearby locations and general mosquito fauna data for inland regions of NSW.

There are five constructed wetlands proposed for the stormwater harvesting project but it is beyond the scope of this document to provide detailed assessments of the individual wetlands. This document is designed to provide an assessment of overall risk associated with these wetlands and general information that can be applied to the design and management of each wetland. Without further site-specific information, it is not possible to provide more detailed information at this point.

* This Mosquito Risk Assessment has been prepared in accordance with the brief provided by Orange City Council and is for the use only by this client. The Department of Medical Entomology, Westmead Hospital, accepts no responsibility for its use by other parties. The use of brand names and any mention or listing of commercial products or services in this article does not imply endorsement by the Department of Medical Entomology, or discrimination against similar products or services not mentioned.
BACKGROUND: MOSQUITO BIOLOGY

Mosquitoes are small blood sucking insects that belong to the family of flies called Culicidae (Order Diptera) and there are more than 300 different species in Australia with each species closely associated with particular habitats.

Mosquitoes have a relatively short but complex life cycle consisting of eggs, four aquatic larval stages (instars), an aquatic pupal stage and a terrestrial adult stage. Mosquitoes are dependent on water, with the immature stage totally aquatic, and without access to free-standing water of some kind, the larvae cannot complete their development to the adult phase.

A gravid adult female mosquito will typically lay eggs either on the water surface (usually with eggs in the form of a floating raft) or on a frequently inundated substrate (usually singularly or in small groups). The ‘oviposition sites’ may include frequently inundated soil or vegetation at the edge of a wetland, soil or leaf litter where temporary pools form after rainfall or inside water holding containers (eg. tins, tyres etc).

While some mosquito eggs (such as those laid by Aedes or Verrallina species) can be desiccation resistant and remain unhatched for many months before being inundated by tides or rainfall, most eggs (particularly those laid by Culex and Anopheles species) will hatch within 2-3 days. On hatching, the young larvae (commonly called wrigglers) feed continuously on aquatic particulate matter and grow through four different instars or moults. The larvae of some mosquito species have developed specialised mouthparts and are predatory, feeding on other mosquito larvae and aquatic invertebrates. The final larval stage (4th instar) develops into a pupa (commonly called tumbler) from which the adult mosquito emerges approximately 2 days later. During summer, it generally takes seven to ten days from the hatching of larvae to the emergence of adults.

On average, a female mosquito may live approximately 2-3 weeks but the male’s lifespan is much shorter. Adult mosquitoes are most active from dusk until dawn, seeking refuge during the day in cool and humid habitats such as well-vegetated areas or under houses. Some pest species, however, can be active during the day and disperse many kilometres from larval habitats.

Within their lifetime, both adult male and female mosquitoes will feed on nectar and plant fluids, but it is only the female that will seek a blood meal required to provide protein for egg development. While many mosquitoes are generalist feeders, some specialise in feeding on humans, mammals, birds or amphibians. Host seeking females respond to a range of stimuli, the most important being carbon dioxide.
THE STUDY SITE

Study site

The Orange district is located on the Central Tablelands of NSW, approximately 250km west of Sydney. Orange City Council services a residential population of over 30,000 and the area is becoming increasingly popular as a tourist destination.

To address issues of water shortages, Orange City Council has investigated strategies to harvest flows from waterways in the region including Blackman’s Creek and Ploughman’s Creek. As a component of the Ploughman’s Creek stormwater harvesting project, five constructed wetlands are proposed to be built across the two stages of the project. Stage 1 includes Cargo Road, Escort Way, Sommerset Park and Burrendog Way wetlands while Stage 2 will include Beer Road wetland. The location of these five wetlands in relation to waterways and residential areas of Orange City Council are shown in Figure 1.

Reference locations

In the absence of site-specific mosquito population sampling or the availability of historic mosquito abundance data for the Orange district, information must be gathered from reference locations elsewhere in NSW.

The NSW Arbovirus Surveillance and Mosquito Monitoring Program commenced in the summer of 1984/85 with the intention of measuring the temporal and spatial abundance of major arbovirus vectors from across the state with particular emphasis on those areas where mosquito-borne disease is regularly reported. The total number of trapping locations has varied from year to year. However, there is long-term population and arbovirus activity data available from many sites throughout coastal and inland NSW.

The closest site to Orange is Forbes. At this location, mosquito populations were sampled at up to three trap sites and on up to 20 occasions per year from 1995-1996 through until 2000-2001. Two additional inland sites that have had long-term mosquito monitoring programs are Leeton and Griffith. While there are some differences in the climate and local land use between Leeton and Griffith and Forbes and Orange, the data collected provides a guide to the mosquito fauna of inland NSW.

Full details on these three reference site locations, trapping methodology used and background on mosquito and arbovirus activity over recent years are provided on the website of the NSW Arbovirus Surveillance and Mosquito Monitoring Program [http://www.arbovirus.health.nsw.gov.au].
Figure 1. Location of the five proposed wetlands to be constructed, Orange City Council, NSW.
Climate and rainfall

There are a number of key climatic differences between Orange and the three reference sites that must be taken into account when comparing the abundance and diversity of mosquito populations across the four locations. These climatic differences are important considerations and extrapolation of data from the reference sites to the Orange district should be done with caution.

Notwithstanding the distance between Orange and the three reference locations (approximately 100km to Forbes, 350km to Leeton and 350km to Griffith), there are differences in elevation (Orange 950m compared to Forbes, 230m, Leeton 140m and Griffith 135m), rainfall and temperature.

The long-term average total monthly rainfall for Orange and the three reference locations is shown in Figure 2. Of note is the substantially greater rainfall recorded at Orange compared with the reference locations. This rainfall distribution would typically indicate that Orange may be predisposed to more favourable mosquito habitats when compared to the other three locations. However, given the absence of large floodplains and/or wetlands associated with the major waterways, favourable habitats for major pest mosquitoes are not common in the region.

The relatively high elevation of the Orange district results in generally cooler temperatures compared to the three reference locations as indicated in Figure 3. While mean daily maximum and minimum temperatures per month are relatively similar for Forbes, Leeton and Griffith, temperatures recorded at Orange between December and March each year are generally 4-6°C cooler.

Studies have shown that temperature is a significant predictive factor for mosquito activity and, even in the presence of favourable mosquito habitats and high rainfall, mosquito populations, particularly the most common freshwater species found in NSW, will not reach substantial levels at cool climates.

The comparison of climate between Orange and the three reference locations highlights some notable differences. While the higher rainfall may predispose the region to potential increases of mosquito populations, the cooler climate may limit the magnitude of population increases. The use of the three reference locations provides important information in the absence of local mosquito data. However, the extrapolation of mosquito risk from the data provided by reference locations should be approached with due caution.
FIGURE 2. Mean long-term total monthly rainfall for four key inland mosquito monitoring locations in NSW including Orange, Forbes, Griffith and Leeton.

FIGURE 3. Mean long-term daily maximum and minimum temperatures per month for four key inland mosquito monitoring locations in NSW including Orange, Forbes, Griffith and Leeton.
MOSQUITO POPULATIONS OF THE ORANGE REGION

While many regions of NSW have been part of the NSW Health Arbovirus Surveillance and Mosquito Monitoring Program, there has not been any mosquito sampling undertaken within the area covered by this mosquito risk assessment. No recent scientific studies or technical reports could be found that may provide quantitative data on the extant mosquito fauna.

Although the previous section has identified that the cooler temperatures of the Orange district is a possible limiting factor in the potential for large mosquito populations. In the absence of any mosquito fauna surveys, it is difficult to make predictions regarding the abundance of potential pest species. However, from information on mosquito fauna from other regions within inland NSW some predictions of general mosquito fauna can be made.

An assessment of adult mosquito collections undertaken at Forbes has identified a total of 15 mosquito species (Table 1), with the species of greatest concern, due to likely nuisance biting impacts and potential public health risks, *Anopheles annulipes*, *Coquillettidia linealis*, *Culex annulirostris*, *Culex quinquefasciatus* and *Aedes notoscriptus*. These species are associated with a range of different habitats including natural and urban freshwater environments. The actual impacts of these species is expected to vary with the relative abundance of populations dependent on factors such as the quantity and distribution of rainfall, flooding events, species specific dispersal patterns, prevailing winds and the distribution of appropriate refuge sites. An important factor in determining future mosquito diversity and abundance, as well as pest impacts, may be the creation of new wetlands if they represent a substantial change in the availability of potential mosquito habitat.

Compared to the abundance of mosquitoes at other locations such as Leeton and Griffith, mosquito abundance at Forbes is considered relatively low (Figure 4). The substantially greater abundance of major pest species such as *An. annulipes* and *Cx. annulirostris* at Leeton and Griffith compared to Forbes is the result, primarily, of available habitat provided by agriculture but also higher mean temperatures. Both Forbes and Orange are surrounded by different types of agriculture compared to Leeton and Griffith where flatland crops associated with flooding irrigation (e.g. rice) are more likely to provide opportunities for mosquito production. In addition, Orange and Forbes are located at a higher elevation than Leeton and Griffith and resulting in generally cooler temperatures, this is particularly the case for Orange that experiences a much cooler climate than Forbes.

This data suggests that the species recorded from Forbes provides a reliable indicator of mosquito diversity in the Orange region. However, the substantially higher rainfall and cooler temperatures of Orange make it difficult to confidently predict the abundance of pest species in this region. While the cooler temperatures may limit the abundance of mosquitoes, the higher rainfall may provide more favourable mosquito habitats. It is difficult to make an assessment on current
productivity of aquatic habitats but the steep topography throughout much of the Orange region indicates there are few opportunities for low-lying floodplains or wetlands. The absence of substantial bushland areas, where mosquitoes associated with ephemeral ground pools created following rainfall can be productive, is also likely to limit the abundance of mosquitoes.

Overall, this information suggests that the mosquito species most likely to represent current and future pest impacts in the Orange region are:

- **Culex annulirostris** is a medium sized, light to dark coloured mosquito with a banded proboscis. Larvae are commonly collected from a range of freshwater habitats from flooded grasslands to permanent, well-vegetated wetlands. This mosquito is of increasing concern as constructed freshwater wetlands are increasingly incorporated into urban coastal developments.

- **Coquillettidia linealis** is a medium sized, dark mosquito and has the potential to be an important pest close to extensive natural or constructed well vegetated freshwater wetlands. The larval biology of this species differs markedly from most other mosquitoes in that the larvae have a modified siphon that attaches to the roots and/or stems of aquatic vegetation.

- **Culex quinquefasciatus** is a medium sized pale brownish mosquito and another very common pest species in urban areas. The larvae of this mosquito are usually associated with, habitats with a high organic content such as drains, sullage pits, septic tanks and other water holding and water storage areas. This species is more likely to be associated with a constructed wetland if there are high levels of organic content within inflows.

- **Anopheles annulipes** is a medium sized, spindly grey mosquito. The larvae are typically associated with freshwater wetlands, particularly those where filamentous algae forms thick mats on the water surface.

- **Aedes notoscriptus** is a small to medium sized dark mosquito with conspicuous pale banded legs. This mosquito is often a serious nuisance pest in residential areas. The larvae are usually associated with small water holding containers around dwellings such as tins, pots, ornamental ponds, blocked guttering and tyres, as well as water holding plants (e.g. bromeliads) and tree holes.

It is likely that there are many other species of mosquito present in the greater Orange region. However, these species may only be found at very low abundances or may only be collected following particularly favourable climatic conditions. The eggs of many *Aedes* spp. are desiccation resistant and can remain in soil or on rocks or debris for many years until flooding occurs. These species may cause some short-term nuisance-biting impacts but do not represent the same potential pest risks as other species.

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<td>213</td>
<td>612</td>
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FIGURE 4. Mean trap night abundance of five common mosquito pests collected at Forbes, Griffith and Leeton as part of the NSW Arbovirus Surveillance and Mosquito Monitoring Program.
MOSQUITO RISK AND CONSTRUCTED WETLANDS

The areas of Australia likely to be at greatest ‘mosquito-risk’ from constructed wetlands are those where natural wetlands are non-existent or few and, generally, there are relatively small mosquito populations. In such areas, particularly in arid or semiarid regions, the establishment of wetlands can provide a high-risk situation with habitat for pest and vector mosquitoes, and perhaps for vertebrate reservoirs of mosquito-borne pathogens, that presents a major public health concern to local communities. Elsewhere, in regions with greater inherent mosquito habitat, constructed wetlands can still be a health concern, providing additional breeding and harbourage sites and bringing vector mosquitoes into closer association with pathogen reservoirs such as birds that are roosting or nesting, or mammals visiting for water or shelter.

In the absence of site-specific mosquito sampling, due caution must be exercised when assessing the potential future pest impacts associated with the constructed wetlands based on off-site mosquito data. However, while the magnitude of mosquito populations may be difficult to predict, the mosquito species of greatest concern in the local area can be identified.

The three species most likely to colonize the constructed wetlands are *Cx. annulirostris*, *Cq. linealis* and *An. annulipes*. Of the mosquito species most likely to exist in the local area, these three species are best suited to the newly constructed wetlands. However, each of these three species exhibits unique biological and ecological requirements that may influence the rate of colonization of wetlands and generation of problematic populations.

It is important to note that currently, there does not appear to be any substantial wetland areas that already support populations of these species. While the mosquitoes may be present in the Orange region, there is no evidence from aerial photography that substantial habitats occur close to the residential areas. There are, however, some areas along Ploughman’s Creek that appear to be potential mosquito habitats but, as the constructed wetlands (particularly Escort Way Wetland, Sommerset Park Wetland and Burendong Way Wetland) will replace these existing habitats, there may be no significant net increase in potential mosquito habitat.

As previously stated, it is difficult to assess the current mosquito productivity of areas along Ploughman’s Creek based on aerial photography alone. While there appears to be some moderately large areas of macrophytes, many factors will influence how favourable they are for mosquitoes and, as a consequence, make it difficult to determine if they currently represent a locally significant mosquito habitat.

As there are unlikely to be large existing populations from which colonization of newly constructed wetlands can occur, the five newly constructed wetlands may represent a significant increase in potential mosquito habitats and, as a consequence, contribute to overall increases in mosquito populations.
Nuisance biting

It is extremely difficult to quantify the impact of nuisance biting in a specific area due to the spatial and temporal fluctuation in mosquito abundance resulting from the mix of different mosquito species, their respective habitats and the environmental and/or climatic conditions that trigger population increases. The NSW Mosquito Monitoring and Arbovirus Surveillance Program identifies average trap densities of adult mosquitoes greater than 100 per trap as high with over 1,000 per trap as very high to extreme when exceeding 100,000 per trap.

At Forbes, adult mosquito collections rarely exceeded 100 mosquitoes per trap night and there is currently no evidence to suggest that mosquito populations at Orange are substantially higher. However, even in areas where there is a low baseline of mosquito activity, even small increases in relative abundance may result in noticeable increases in nuisance-biting rates.

The tolerance level of individuals to nuisance-biting varies substantially and is often dependent on the extant mosquito populations and previous personal experiences. However, there are strong indicators that nuisance biting alone can have negative impacts on a homeowner's standard of living as well as the economic impacts on residential, recreational and tourist developments.

In areas where there is a change in the contact between mosquitoes and humans, where residential developments encroach on wetlands or newly constructed or rehabilitated wetlands increase mosquito populations, pest impacts may be more noticeable. Even in regions where mosquito populations are considered low, it is the relative change in local mosquito abundance and/or diversity that is often the concern for local communities.

Public health risks

Ross River virus (RRV), Barmah Forest virus (BFV) and Murray Valley encephalitis virus (MVE) are the most serious disease-causing pathogens spread by mosquitoes in Australia and are classified as notifiable diseases by NSW Health. There are other arboviruses transmitted by mosquitoes but these rarely cause human disease in Australia.

There are, on average, approximately 5,000 human cases of RRV and BFV per year across Australia. While the symptoms can vary greatly between individuals, and include fever and rash, infection with either of these viruses may result in a condition known as polyarthritis with arthritic pain in the ankles, fingers, knees and wrists. Generally, the rash tends to be more florid with BFV infection but the arthritic pain is greater with RRV infection.
TABLE 4. A summary of mosquito species most likely to occur at Orange and known arboviruses previously isolated from elsewhere in NSW and the human biting risk they represent.

<table>
<thead>
<tr>
<th>Mosquito species</th>
<th>Ross River virus</th>
<th>Barmah Forest virus</th>
<th>Murray Valley encephalitis virus</th>
<th>Other arboviruses¹</th>
<th>Biting pest²</th>
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<tbody>
<tr>
<td>Aedes alboannulatus</td>
<td>+</td>
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<td>Aedes alternans</td>
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<td>Aedes notoscriptus</td>
<td>+</td>
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<td>Culex orbostensis</td>
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<td>Mansonia uniformis</td>
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¹ Other arboviruses include Stratford virus, Edge Hill virus, Kunjin virus, Kokobera virus and Sindbis virus. These viruses, although known to cause mild human disease, are not considered serious public health risks
² Mosquito has been documented as a human biting species (Russell 1990)

Both viruses have the potential to be spread by mosquitoes likely to be present in the Orange region (Table 4), particularly *Cx. annulirostris*, generally considered to be one of the most important pest species of inland regions. The transmission cycles of these viruses require the presence of suitable reservoir hosts such as native macropods (i.e. kangaroos and wallabies or waterbirds), and there is a greater risk of disease transmission in rural and semi-rural areas compared with major urban centres.

It is important to note that while *Cx. annulirostris* is generally considered to be the major vector of arboviruses from inland regions, mosquito species expected to be both present in Orange and potentially associated with the constructed wetlands, such as *An. annulipes* and *Cq. linealis*, are known to carry and transmit disease-causing pathogens. One of the key nuisance-biting pests and vectors of disease, *Ae. notoscriptus*, is likely to be present in Orange but this species will not be directly associated with the constructed wetlands.
While BFV tends to be more prevalent in coastal regions, RRV is, generally, equally common in both inland and coastal regions as indicated by human notification data. From 1994 to 2008, an average of 24 cases per year of BFV were reported from inland areas compared to 298 cases from coastal regions while, over the same period, 257 cases of RRV were reported from inland regions compared to 404 cases from coastal regions.

The most important factors influencing the number of cases from each region are differences in local mosquito fauna (e.g. the coastal regional generally supports a more diverse mosquito fauna), human population density (e.g. residential and tourist populations are generally concentrated closer to more extensive mosquito habitats in coastal regions) and climatic conditions (e.g. coastal regions are less likely to suffer from extended drought conditions and, in addition, population increases of some mosquito species are strongly influenced by tidal flooding of estuaries as well as rainfall).

Within inland regions, the quantity of human disease notifications can vary greatly. Greater Western Area Health Service releases public health warnings to increase awareness of arbovirus activity and provide assistance with personal protection strategies. The Greater Western Area Health Service covers a large area and included Orange which was previously within the Mid-Western Area Health Service jurisdiction prior to amalgamation in 2006.

Between the years 1994-1995 and 2003-2004, there was more activity of RRV compared to BFV in the Mid-Western Area Health Service (Table 5). During the 2007-2008 monitoring period, the notification rates for the Orange region were less than 100/100,000 while the rates for nearby Forbes was 111.7/100,000, and Dubbo was 160.4/100,000. When compared to notification rates over the same period from other inland locations such as Bourke (678.7/100,000), Deniliquin (336.9/100,000) and Leeton (270.7/100,000), the relative risk of mosquito-borne disease in the greater Orange region appears relatively low.

The symptoms of MVE infection can vary from mild to severe to fatal, with symptoms almost invariably including a sudden onset of fever, anorexia and headache, while vomiting, nausea, diarrhoea and dizziness may also be experienced along with lethargy and irritability. Drowsiness, confusion, convulsions and neck stiffness can be experienced a few days after the onset of initial symptoms. The disease can be fatal and many who survive the encephalitic syndrome have some residual mental or functional disability.
TABLE 5. Annual number of human cases and notification rates of Ross River virus and Barmah Forest virus disease from Mid-Western Area Health Service, 1994-1995.

<table>
<thead>
<tr>
<th>Season</th>
<th>Ross River virus</th>
<th>Barmah Forest virus</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. Cases</td>
<td>Rate per 100,000</td>
</tr>
<tr>
<td>1994-1995</td>
<td>0</td>
<td>0.0</td>
</tr>
<tr>
<td>1995-1996</td>
<td>21</td>
<td>12.7</td>
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<td>1996-1997</td>
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<td>3.0</td>
</tr>
<tr>
<td>2003-2004</td>
<td>17</td>
<td>10.1</td>
</tr>
</tbody>
</table>

* The area covered by the Mid-Western Area Health Service is currently included in the Greater Western Area Health Service.

The virus has a natural endemic cycle in northern Australia, which involves water birds as the vertebrate host and the freshwater mosquito Cx. annulirostris as the major vector. Epidemic activity of the viruses in the southeast of Australia is rare and has been associated with excessive rainfall and flooding, which increases bird and mosquito populations, but it is still uncertain whether the virus is introduced occasionally to the southeast from the north or whether it is endemic in inland areas at undetectable levels and only become evident with periods of intense bird and mosquito breeding.

Sentinel chicken flocks are maintained at various locations in western NSW and weekly blood tests are undertaken in conjunction with the NSW Arbovirus Surveillance and Mosquito Monitoring Program. Despite occasional positive test results from these chicken flocks and a very small number of isolates of MVE identified from mosquitoes collected in western NSW (Griffith, Leeton and Macquarie Marshes), there has only been one confirmed human of MVE since the last major epidemic in 1974. There is no evidence to suggest that there is a serious risk of MVE in the Orange region.
MOSQUITO RISK ASSESSMENT AND MONITORING

It is important that for the most reliable assessment of mosquito population change following the construction and/or modification of wetlands, sampling is undertaken before the construction, modification or rehabilitation is undertaken and again. However, this is not always possible for many reasons and, importantly, if such sampling is to be done, the services of a professional entomologist be engaged to provide advice on the most effective monitoring program.

A mosquito-monitoring program will provide the only reliable, quantitative measure of mosquito abundance and diversity at the site. Mosquito populations vary between and within seasons with changes to temperature and rainfall (both quantity and temporal distribution) and management of mosquitoes is most effectively undertaken in response to the results of monitoring of immature and adult populations. It is important that specimens are correctly identified by an experienced entomologist as often the most abundant mosquitoes may not be directly associated with the wetland habitats and alternative management strategies may be required. Unfortunately, many flying and aquatic insects can be misidentified as mosquitoes and trigger community complaints and unnecessary control agent applications.

Monitoring programs can be tailored to suit site-specific conditions and may not be required to be ongoing for many years. They can be complex, with sampling of adult and immature mosquito populations but at a minimum, adult mosquito sampling should be undertaken as it provides the best guide to actual and potential pest impacts and the species responsible.

For local authorities charged with the task of managing the constructed wetlands, and the associated mosquitoes, it is important to make accurate assessments of the relative productivity of mosquitoes from the wetlands and the resulting actual, potential and perceived pest impacts. Complaints regarding mosquitoes are generally directed towards council and the responsibility for mosquito control is with local council. However, while complaints from the community may provide an indication of increased mosquito activity and warrant further investigation, no mosquito control activities should be undertaken without an entomological survey of the area to confirm the abundance of local mosquito species.

In some locations, nuisance-biting impacts may be reported in residential areas close to newly constructed wetlands. However, entomological investigations may identify the major pest species as one associated, not with the wetland itself but with nearby bushland or backyard habitats. The domestic mosquito *Ae. notoscriptus* can be a serious pest but the species is not found in natural or constructed wetlands. The larvae of this species are associated with small water holding containers in backyards. Consequently, if this species is the source of nuisance-biting impacts, a community education program will be more effective at managing the pest impacts as opposed to control agent application of habitat modification of the wetlands.
Proposed mosquito monitoring program for Orange

It is strongly recommended that a mosquito monitoring program is undertaken over the coming years to assess the impact of the constructed wetlands on the relative abundance of local mosquito species and their potential pest and public health impacts.

Following is a proposed mosquito monitoring program for the Orange region that is simple, relatively cost-effective and will provide some baseline data and preliminary assessment of mosquito populations and their response to the constructed wetlands.

It is proposed that sampling during the period January – March 2010 would provide some baseline information on mosquito activity. Additional sampling during the same period (January-March) in the summer following completion of wetlands and again three years after completion of the wetlands would allow for an effective assessment of both actual and potential mosquito impacts.

1. Timing

With consideration given to long-term rainfall patterns, temperature and ecological requirements of the potential pest species in the region, a mosquito monitoring program would be most effective if targeted within the period between January and March. Outside this period, mosquitoes may still be active but data collected may be unreliable as short-term climatic conditions may have a greater impact on mosquito populations. No sampling should be undertaken between the months of May and November as very little mosquito activity would normally be expected.

2. Traps and trapping network

Adult mosquitoes can be collected using Encephalitis Vector Surveillance (EVS) traps. These small battery operated traps are baited with carbon dioxide (usually in the form of dry-ice) and attract female, host seeking mosquitoes. The traps are effective as they collect a subsample of the mosquito fauna that are most likely to cause pest impacts while also minimizing the collection of non-target insects that are often collected in traditional light traps.

Depending on the expected completion dates of constructed wetlands, the number of trap sites can be prioritized if budgetary constraints limit the number traps available (or number of wetlands targeted) but, as a minimum, at least two traps should be operated at each wetland (or proposed wetland site) on each occasion.

3. Trapping frequency

Trapping frequency will be dependent on the available funds but sampling on at least two occasions during the January – March period would be considered a minimum. Each sampling occasion may consist of 2-3 consecutive nights of trapping with up to 3 traps per wetland. This replicate trapping, if repeated twice or three times will
provide a suitable measure of mosquito abundance with which to compare to the post-construction period.

4. Measuring change

It is difficult to directly assess pest impacts (e.g. nuisance-biting rates) with adult mosquito abundance. However, in the assessment of change to mosquito populations associated with the constructed wetlands, a comparison of both species richness and diversity should be undertaken. A comparison of mean trap night abundance of key pest wetland species (i.e. *An. annulipes*, *Cx. annulirostris*, *Cq. linealis*, *Cx. quinquefasciatus*) between years and against non-wetland pest species (i.e. *Ae. notoscriptus*) will provide an indication of any change to localized pest impacts.

5. Response to change

If a change is detected that may indicate increased mosquito risk, strategies should be considered that may reduce that risk associated with the wetlands. It is important that any changes in mosquito populations are investigated to determine if they are the result of favourable conditions within the wetlands or unseasonal climatic or environmental factors. A number of strategies for addressing mosquito management are outlined in the following section.
MOSQUITO MANAGEMENT

Constructed wetland design

Constructed freshwater wetlands often contain various component zones and each of these may represent a relative high or low risk of mosquito productivity. These various components can be variously classified, but are often described as follows:

- inlet zone with water control structures, and GPT and detention basin to trap larger pollutants and store surcharge stormwater, and perhaps an energy dissipater (e.g. riffle zone) to prevent erosion,
- deep-water zone and sedimentation pond to manage high stormwater flows and capture settleable solids, and with submerged plants for water quality,
- littoral zone with edge plants for bank protection water quality, habitat creation, recreation and aesthetics,
- macrophyte zone with reed beds for water quality,
- open water zones with islands for habitat and aesthetics,
- deep-water zone for water storage and further sedimentation, and with submerged plants for further quality improvement, habitat and aesthetics,
- outlet zone with water control structures to manage water levels, spillways and weirs to protect wetland during high flow and maintain ponded water during low flow.

There are a number of publications available that discuss the “mosquito-sensitive” design elements of the various constructed wetland components in detail (see Russell and Kuginis 1998 and Russell 2001). However, the most important component that holds the greatest risk for creating favourable mosquito habitats is the shallow macrophyte zone. The productivity of this habitat will be dependent on water depth, vegetation type and vegetation density.

There is little information available on species-specific relationships between mosquitoes and aquatic vegetation in Australia. However, the presence of aquatic macrophytes has been well documented as one of the primary factor influencing the suitability of wetlands for mosquito production. Shallow water macrophyte zones are the most common site of mosquito production in constructed wetlands and studies in North America have identified positive correlations between vegetation density and the abundance of mosquito larvae.

Aquatic macrophytes primarily provide refuge for immature stages of mosquitoes from predators (e.g. fish, macroinvertebrates) and wind generated wave action. The refuge that is provided by the vegetation can vary with the types of plant present. Plants with a single, thick culm may not provide the same degree of protection as a plant with multiple stems. Fast growing invasive species such as *Typha* spp. and *Phragmites* spp. can quickly spread through the wetland and form dense stands. Studies in North America have shown that a complex matrix of aquatic vegetation is beneficial for wetland function and the minimisation of mosquito populations but
when invasive species create large uniformly vegetated areas, wetland function is reduced and mosquito production is more likely to increase. Unfortunately, without an appropriate vegetation management plan, even if wetlands are initially planted with a complex matrix of native species, invasive species can quickly move in a take over the wetland.

In addition to the physical refuge created for mosquitoes by aquatic vegetation, the plants seasonally shed material that can contribute to an increase in the organic content of the wetland. The decaying vegetation can create further refuges but the decomposition of this material can increase the organic content of the wetland and improve conditions for species of mosquito that prefer the highly organic conditions such as *Cx. quinquefasciatus* and *Cx. molestus*. Within these densely vegetated areas, dissolved oxygen concentrations are relatively low and may reduce the survival of predatory fish and macroinvertebrates, further enhancing conditions for mosquito production.

There are some species of mosquito that have a direct relationship with aquatic macrophytes. *Coquillettidia* and *Mansonia* species (both likely to be present in Orange) have modified body parts that attach directly to plant material for oxygen exchange rather than the water surface like the majority of mosquito species (such as *Anopheles*, *Aedes*, *Culex* and *Verrallina* species).

The larvae of mosquito species such as *Cq. linealis*, attach to the stems and exposed roots of emergent macrophytes. The larval development time for these species can be substantial longer than normal lasting many months rather than under 10 days which is typical of most mosquitoes. As the larvae are attached to submersent vegetation, they are rarely collected in standard larval monitoring and little is known of the appropriate control strategies available for these species. While *Coquillettidia* species prefer to attach to roots amongst sediments and the lower end of stems, *Mansonia* species are thought to be more strongly associated with floating vegetation.

Specifications for depths and margins of vegetated wetland ponds, to prevent emergent vegetation contributing to mosquito problems, should approximate a steep (>30°) edge and a deep (>1.3 m) bottom. Higher incidence of mosquitoes has been associated with smaller (<0.2 ha) rather than larger retention basins, particularly when water depths are less than 0.3m. With larger ponds, profiling the bottom (to a slope of 0.01%) with the depth greatest at the inflow end can be advisable to prevent/concentrate pooling, particularly if there are periods of low flow, although if the flow is low enough to have the water disappear by infiltration, evaporation and/or transpiration within a week, then a lower forward slope may be appropriate. Poor drainage through inadequate grading, and poorly constructed outlet or recirculation structures, provide for mosquito problems; pools should be provided in deep sections for predators to survive periods of low flow, drawdown or drainage. Additionally, wetlands should not ‘leak’ water to pool nearby and create mosquito habitats separate from the wetland and inaccessible to predators.
The manipulation of water levels in the wetlands is also a useful tool in managing mosquito populations. The ability to drain wetlands strategically in response to increasing mosquito production is useful but it is important to note that while the life cycles of major pest species such as *Cx. annulirostris* and *Cq. linealis* may be interrupted, populations of species that prefer habitats prone to temporary drying and reflooding (e.g. *Aedes* spp.) may potentially increase. It is also important to consider the effect such a strategy may have on the vegetation itself and other aspects of wetland function and/or purpose.

The production of mosquitoes from stormwater inlet pits, gross pollutant traps or surface storage areas (e.g. bioretention swales) can be sometimes be problematic. For above ground water storage, mosquito production can be avoided by ensuring that the structures are self draining, have the siltation depth shallow enough to encourage evaporative drying, and that the accumulation of organic material is maintained at low levels.

Sub-surface water flow and storage structures are generally not suitable sites for mosquito production unless the carrying capacity of the drainage system is regularly exceeded and standing water pools at the surface for more than 5 days. If mosquitoes (particularly *Cx. quinquefasciatus*) can gain access to the standing water in the void spaces between the gravel, some mosquito breeding may occur with the suitability of the habitat increasing over time with sedimentation and vegetation growth.

**Mosquito control: Biological control**

A number of organisms have been investigated to determine their suitability as effective predators of mosquito larvae. These include invertebrate (e.g. Diptera and Coleopteran larvae, Crustaceans, Notonectids, Odonates) and vertebrate (fish) predators. In urban environments, fish are often used to control mosquito production in ornamental ponds and constructed wetlands, while Odonates and Copepod crustaceans have been used to control container-breeding mosquitoes.

The ‘mosquitofish’ *Gambusia holbrooki* was introduced to Australia from North America at the beginning of the 1900s and has since spread to most of the waterways in Australia. There is some debate as to the effectiveness of *G. holbrooki* as a mosquito control agent, but where they are less effective it is often because of obstructive vegetation and in such circumstances alternative species are similarly ineffective. The ‘mosquitofish’ has been implicated in significant adverse impacts on aquatic native fauna, particularly other fish and amphibians, and under no circumstances should *G. holbrooki*, or other exotic fish or non-endemic native fish, be released into aquatic habitats for mosquito control.

A number of native fish has been identified that may be appropriate for mosquito control in Australia. A few examples of native species known to consume mosquito
larvae are *Pseudomugil signifer* (Pacific Blue-eye), *Hypseleotris compressa* (Empire Gudgeon) and *Hypseleotris galii* (Firetail Gudgeon). While native fish introductions alone will not significantly reduce mosquito populations, they do provide an important component of integrated pest management and have been shown to provide a valuable link to the wider community promoting environmentally sensitive mosquito management. However, the frequently polluted water of urban wetlands, the ‘obstructiveness’ of heavy vegetation, and the widespread distribution and local proliferation of *G. holbrooki* all serve to limit the likely success of native fish for mosquito control.

Tadpoles are often mistakenly promoted as effective predators of mosquito larvae, but most are general herbivores and suspension feeders and there is little evidence that effective control of mosquito larvae can be achieved through the use of tadpoles alone. Recent studies have shown that, although tadpoles of *Limnodynastes peronii*, *Limnodynastes tasmaniensis*, *Litoria aurea* and *Litoria peronii* consume mosquito larvae in small containers, there is no evidence that they provide an effective level of mosquito control.

Given the trend towards more ‘environmentally sensitive’ control strategies, the use of potential use of predators such as dragonfly adults, frogs and spiders against adult mosquitoes is often raised, but there is no evidence they can have any significant impact on pest mosquito populations. In particular, the installation of ‘bat houses’ to maintain local populations of insectivorous bats has become a popular proposal for biological control of adult mosquitoes. There is evidence that insectivorous bat species are active in and exhibit feeding behaviour close to known mosquito habitats but there is no indication that mosquitoes are a preferred food source or that the bats can significantly reduce the mosquito populations. While providing increased refuges and habitat for bats may be advantageous for bat conservation, it would be unwise to link bat conservation to mosquito control as the sole objective of this strategy, and there is no scientific evidence from anywhere in the world that insectivorous bats or birds have any significant impact on adult mosquito populations.

**Mosquito control: Larvicides**

The only strategy available to completely remove the risk of mosquito impacts is to undertake a mosquito control program in the wetlands. While there is no indication that the mosquito risk is sufficient to warrant the implementation of a mosquito control program at the present time, or at least until local mosquito surveys have been undertaken, it is important that the options available are discussed.

It should be expected that mosquitoes will be active during the warmer months and that they are an integral part of many wetland ecosystems. Mosquitoes are an important component of the wetland ecosystem with mosquito larvae providing food for birds, fish and macroinvertebrates and adult mosquitoes providing food for birds,
bats, frogs, reptiles and spiders. While there is strong evidence that larvicides, when applied correctly and at recommended rates, have minimal impact on non-target organisms, there is little information available as to the impact of large scale mosquito treatment programs and the resulting removal of mosquitoes, over the long term, from local ecosystems.

Temephos is an organophosphate compound that has been used in mosquito control since the early 1950’s, but although it is highly effective it is not totally selective for mosquitoes and may have toxic effects on non-target organisms such as birds, fish and some invertebrates - particularly in estuarine habitats. Additionally, there have been concerns regarding the development of resistance in target species and, combined with the potential for non-target and environmental impacts, local mosquito control authorities have reduced the use of temephos in favour of alternative control agents. However, for the treatment of water holding containers where contact with non-target organisms is minimal, such as tyre piles, this product may be appropriate and is available in liquid or granular formulations.

The bacterium Bacillus thuringiensis israelensis (Bti) produces a protein crystal which contains a number of microscopic pro-toxins that when ingested are capable of destroying the gut wall and killing mosquito larvae within 12 hours. Commercial formulations of the bacterial culture product are available, with liquid Bti formulations most often used. The greatest benefit of Bti is that it is highly specific to mosquito larvae and very few non-target effects have been recorded when the product is applied at recommended rates. Unlike chemical insecticides (e.g. household fly spray) that kill insects on contact, this larvicide must be ingested by the mosquito larvae. It is only under the specific conditions within the gut of the mosquito larvae that the toxins are released and break the lining of the gut.

In Australia, it has been used successfully to reduce pest mosquito populations in both saline and freshwater habitats. The product does, however, have some disadvantages in that the efficacy is reduced in habitats with a high organic content (so it is less likely to be effective in some wetlands, stormwater systems and wastewater structures), and when larval populations are very young (i.e. 1st instar) or nearing pupation (i.e. late 4th instar larvae that have stopped feeding and thus will not ingest the Bti). As the product has no residual activity, there can be only a very small window of opportunity for effective treatment of larval populations. Generally, Bti is not considered the most suitable control agent for the type of wetlands proposed for construction in Orange.

The insect growth regulator, s-methoprene is a synthetic mimic of the juvenile hormone produced by insect endocrine systems and has been shown to be an effective control agent of pest mosquitoes without adversely affecting non-target organisms. When absorbed by the larvae, development is interrupted and larvae fail to successfully develop to adults, usually dying in the pupal stage. A side benefit of s-methoprene is that it retains mosquito larvae and pupae in the aquatic ecosystem long enough to provide food for predators.
The product is available in three formulations, a liquid, and slow-release solid pellets and briquets. The advantage of the pellets and briquets is that they are ‘slow-release’ formulations and offer extended periods of control, reducing the need to treat wetlands or stormwater structures on numerous occasions during the ‘mosquito season’. This product would be considered the most suitable control agent for the constructed wetlands should mosquito populations increase to problematic levels.

**Terrestrial vegetation management and buffer zones**

Harbourage sites contribute to an increase in the pest impacts of mosquitoes by acting as cool humid refuges from which mosquitoes can disperse. As well as contributing to favourable habitat conditions that assist increased longevity of mosquitoes (and consequently increasing the risk of disease transmission), these harbourage sites can act as “stepping stones” (a number of clumps are close together) or “bridges (a continuous corridor of vegetation) that facilitate the movement of mosquitoes into areas they would not normally disperse due to a naturally short flight range.

The use of buffer zones, vegetated or non-vegetated areas between wetlands and/or harbourage sites and residential/recreational areas has been proposed as a potential strategy to reduce mosquito impacts. However, there is ongoing debate as to the design of such buffer zones and their relative effectiveness.

While the concept of “buffer zones” does not offer a single effective strategy to reduce mosquito impacts, the basic principal of reducing harbourage sites close to residential areas is a worthwhile consideration. Minimising dense vegetation close to residential areas, particularly immediately surrounding buildings, will reduce the density of mosquitoes in close proximity to residents.

With the exception of Beer Road Wetland, the proposed wetlands are very close to existing residential areas (generally less than 100m) and the use of buffer zones is not considered a suitable management strategy.

**Adult mosquito control: Adulticides & barrier treatments**

The routine application of adulticides is not a viable management strategy and should only be considered during disease epidemics. Although there are products available that may be useful against adult mosquitoes these products are not effective for the control of mosquitoes over a large area, can be expensive and may have substantial non-target impacts.

A recently proposed mosquito mitigation strategy is the use of barriers treated with a residual insecticide, bifenthrin (trade name BISTAR). Bifenthrin is a contact poison with high insect toxicity but low mammalian toxicity and provides a residual layer of pesticide that kills landing and/or resting mosquitoes. It is currently registered for the
control of mosquitoes and biting flies around dwellings as a control agent for treating mosquito resting places (internal & external areas of domestic, commercial, public and industrial buildings).

Initial field trials have shown positive results with a reduction in mosquito abundance observed where the product has been applied. However, there are warnings on the label that the product is toxic to bees, fish & aquatic organisms and the use of this product adjacent to a nature reserve may not be considered appropriate. There are no published reports available on the impact to non-biting insects but as this product is a contact poison, it is likely that many of these non-target insects may be killed if exposed to the product.

**Domestic mosquito traps and other devices**

A number of commercial trapping units are available that utilise attractants (e.g. light, heat, carbon dioxide, odour) to draw in and catch or kill adult mosquitoes. There are many different types of units available and, while most will collect mosquitoes, there is no quantitative evidence that they can significantly reduce nuisance biting impacts in areas close to productive breeding habitats. Units that use light alone to attract mosquitoes (e.g. Blue light electrocutors) have been shown to have little impact on nuisance biting rates and often kill many more harmless insects than mosquitoes.

The units that use carbon dioxide as the main attractant may offer some limited protection but a network of multiple traps would probably need to be employed to protect the residential and/or recreational areas from exposure to mosquitoes and there is no information available in Australia regarding the design of effective trapping networks.

There is a range of ‘sonic repeller’ units commercially available ranging from small battery operated key chain devices to large units that plug directly into power points. The general principal is that female mosquitoes are repelled by sound of the male mosquito wing beats as they avoid multiple matings. However, there is no scientific basis to these claims and scientific trials have repeatedly shown that these units are not effective.

There are a number of electronic units available (for both indoor and outdoor use) that release insecticides from slow release mats or liquids. These units can be very effective as the pyrethroids (e.g. allethrin) kill mosquitoes rather than simply repelling them. Alternative products, particularly ‘coils’ and ‘sticks’, containing natural products such as tea tree oil or citronella will only repel mosquitoes and are not as effective.

**Personal protection and education**

While some mosquito activity should be expected in outdoor areas, the entry of mosquitoes into buildings can often have significantly greater nuisance impacts. Fly screens of an appropriate mesh size should be fitted to windows and doors where
possible and maintained regularly. It should also be ensured that there are no entry points via air conditioning ducts, ventilation structures or other connections between indoor and outdoor areas.

As with residential area located close to wetlands, it should be expected that some mosquitoes will be active during the summer months. Residents and visitors should be encouraged to undertake personal protection measures to reduce the impact of nuisance biting and the risk of arbovirus disease transmission.

Avoidance of wetland areas during the peak periods of mosquito activity (i.e. dusk and dawn) and the use of personal insect repellents (formulations containing DEET or Picaridin or mosquito coils containing insecticide) are also highly recommended to minimise mosquito impacts. There are many products available that contain plant extracts and essential oils as their active ingredient. These products have been shown to offer protection for only a short period of time.

It is important to note that mosquitoes produced from residential and rural properties can also contribute to localized mosquito impacts. The community should be informed of these risks as part of community education programs.

Mosquito production from septic tanks, usually of *Cx. quinquefasciatus*, can be problematic and occurs when the mosquitoes can access the interior of the tank through a damaged or open top, or unscreened vents. While modern fibreglass tanks are less likely to pose a significant mosquito risk, the older style concrete tanks are much more likely to allow access by mosquitoes, particularly if allowed to fall into a state of disrepair.

The maintenance of a property free of as many water holding containers as possible will reduce available habitats for mosquitoes such as *Ae. notoscriptus*. Many of the potential habitats could be classed as rubbish and are best disposed of appropriately. However, while other items in the property may need to be stored and/or covered so that they are not filled following rainfall events, it is important that these coverings themselves do not allow pooling of rainfall and subsequent mosquito production. These mosquitoes only require small volumes of water and the least likely objects can be productive habitats, such as garden ornaments, open fence posts and children’s toys. Any object that can hold standing water for more than five days should be considered a potential larval site.

As the Australian community becomes increasingly aware of water conservation issues, the use of domestic water hoarding strategies is raising concern regarding the threat of increased mosquito populations. While modern rainwater tanks should pose minimal risk and older tanks, if properly screened, should not support mosquitoes, the use of buckets, bins, open tanks and other large containers to store rainwater and runoff is of concern.
CONCLUSIONS & RECOMMENDATIONS

- It is difficult to make accurate assessments of extant mosquito populations at Orange in the absence of current or historic site-specific data. However, based on a comparison to three reference locations in NSW, Forbes, Leeton and Griffith, and with consideration given to the differences in climate (including rainfall and temperature) between the locations, it is reasonable to make the assessment that, although potential pest mosquitoes will be present in the local area, the abundance of those species is generally considered low and that the period of peak mosquito activity is limited to the months between January and March each year.

- The most important pest species in the local area are most likely An. annulipes, Cq. linealis, Cx. annulirostris, Cx. quinquefasciatus and Ae. notoscriptus. These five species are known nuisance-biting pests and vectors of disease-causing pathogens in some regions of NSW. With the exception of Ae. notoscriptus that is closely associated with backyard water holding containers, the other four species all have the potential to colonise the proposed constructed wetlands.

- The productivity of the constructed wetlands will be highly dependant on their final design, particularly the macrophyte zones that have the greatest potential for providing favourable mosquito habitats. There is still uncertainty regarding the likelihood of problematic mosquito populations being produced in a location like Orange that experiences a cool climate. However, given the relatively high rainfall compared to other inland sites and that there appears to be no substantial mosquito habitats currently in the local area, the constructed wetlands may represent a significant increase in available mosquito habitat. The resulting increases in mosquito populations may not be substantial but given what may be a relatively low baseline level of mosquito activity, nuisance-biting impacts in residential areas close to the wetlands may be noticeable.

- It is strongly recommended that mosquito sampling in undertaken during the January-March 2010 period in order to establish the levels of extant mosquito activity. This sampling can be used to make more accurate assessments of future mosquito risk and inform the design and management of the wetlands. Additional mosquito population sampling following the completion of wetlands will allow a quantitative assessment of mosquito population change.
REFERENCES


