Elimination of *Aedes aegypti* in northern Australia, 2004-2006

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ABSTRACT: The Northern Territory (NT) of Australia is currently free of the dengue mosquito *Aedes (Stegomyia) aegypti* (L). However, on 17 February 2004, two *Ae. aegypti* adults were captured in two routine CO_2-baited encephalitis virus surveillance traps in Tennant Creek, located 990 km south of Darwin in the NT. The detection triggered an immediate survey and control response undertaken by the NT Department of Health and Community Services, followed by a Commonwealth of Australia-funded *Ae. aegypti* elimination program. This report details the methods and results of the detection and subsequent elimination activities that were carried out between 2004 and 2006, returning the NT to its dengue vector-free status. There have been very few successful *Ae. aegypti* elimination programs in the world. This purposeful mosquito elimination for Australia was officially declared on 5 April 2006. ***Journal of Vector Ecology* 45 (1): 118-126. 2020.**

Keyword Index: Australia, mosquito surveillance, elimination, vector control, *Aedes aegypti*, mosquito-borne disease.

INTRODUCTION

Aedes aegypti is a vector for arbovirus-borne diseases such as dengue, Zika, chikungunya, and yellow fever, with dengue outbreaks causing high mortality in many tropical countries (Gubler 1998, Stanaway et al. 2016, Christofferson 2016). Although dengue is not currently endemic in Australia, the vector is well established in central and far northern Queensland (Qld), where importations of the virus in infected travelers has led to dengue outbreaks (Hanna et al. 2003, Russell and Kay 2004, Ritchie et al. 2013).

Aedes aegypti was also widely established in the Northern Territory (NT) until the 1940s, where it was recorded from Darwin, the principal city on the north coast, to Newcastle Waters in the south, before it disappeared between 1956 and 1974 (Whelan 1981). Its disappearance may be due to various factors, including the widespread reticulation of water during and soon after World War II and the coincidental removal of rainwater tanks (O'Gower 1956, O'Gower 1958, Lee et al. 1987). The same appeared to have occurred in the city of Brisbane, Qld, where the introduction of reticulated water and the reduction in the number of rainwater tanks played a major role in the disappearance of *Ae. aegypti* (Trewin et al. 2017).

Since 1974, regular and comprehensive mosquito surveillance and control has been conducted by the NT Department of Health and Community Services (DHCS) Medical Entomology (ME) unit, including exotic vector surveillance in major NT centers and at NT first points of entry in liaison with the Department of Agriculture and Water Resources (Whelan 1991, Whelan 2007). Despite numerous dengue mosquito interceptions in Darwin port areas, the mosquito has failed to establish, due to successful interception responses (Whelan 1991, Kurucz and Pettit 2013). However, on 17 February 2004, *Ae. aegypti* was detected in two routine CO_2-baited (1 kg dry ice) encephalitis virus surveillance traps (Rohe and Fall 1979), herein after referred to as EVS traps, in Tennant Creek (TCK), which is south of any previous occurrence of this species in the NT (Figure 1). The detection indicated a possible town-wide establishment in an arid region of Australia thought to be free of this vector.

This report details the initial *Ae. aegypti* detection response and subsequent surveillance and control activities carried out during the successful dengue mosquito elimination program in TCK between 2004 and 2006.

MATERIALS AND METHODS

Aedes aegypti detection and initial response (17 February to 17 April 2004)

Tennant Creek is a small town located in an arid region 990 km south of Darwin, comprised of 1,107 properties, including urban residential, industrial, rural, and vacant lots, in addition to seven Aboriginal communities consisting of 107 residential properties.

On 17 February 2004, a total of two female *Ae. aegypti* were collected in two separate EVS traps located 900 m apart, indicating a widespread establishment in the town. The ME team commenced immediate *Ae. aegypti* surveillance and control measures between 22 February and 17 April 2004, with support from the DHCS Centre for Disease Control and Environmental Health, Western Australian Health and Australian Defence Force staff, with weekly teams comprising of up to eight staff traveling to TCK during this period. ME staff were appointed authorized officers under NT Public Health Act, with surveillance and control activities supported by the local Barkly Regional Council, the Julalikari Council and Anyinginyi Congress.

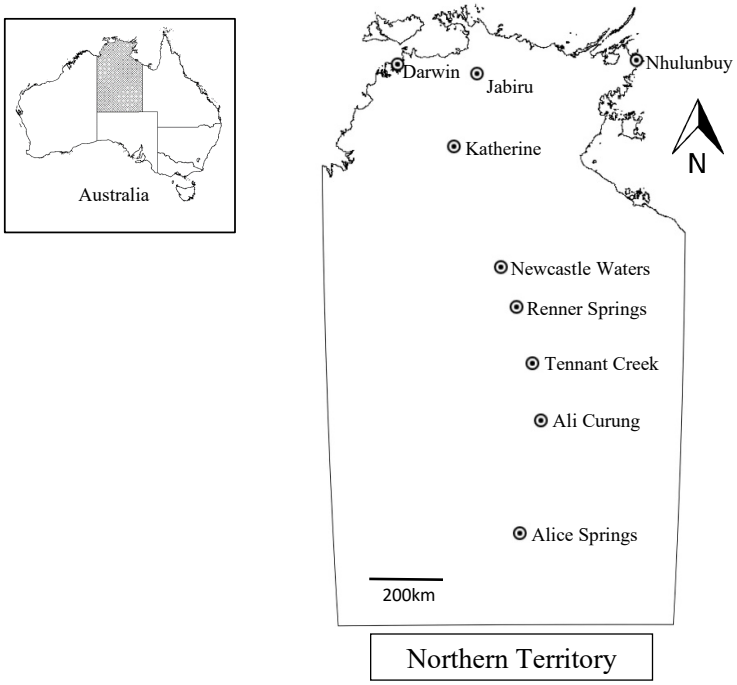


Figure 1. Map showing the location of Tennant Creek in the Northern Territory.

Adult mosquito and ovitrap surveillance

To assess the distribution and abundance of *Ae. aegypti*, a total of 59 EVS traps were set overnight at *ad hoc* locations throughout TCK during the initial response period. Trap sites included high breeding or importation risk areas, such as the airport, bus, and railway terminals, plant nurseries, trucking and tire yards, the refuse landfill, and properties where *Ae. aegypti* larvae were found during property inspections. In addition, 30 sticky ovitraps (Ritchie at al. 2003) were set for five days around the town on 23 February, and 100 lethal ovitraps (Ritchie 2005, Williams et al. 2007) were set between 28 February and 16 April 2004, with each lethal ovitrap trap containing two s-methoprene pellets (Altosid®), a hormone growth regulator to prevent the traps from becoming larval mosquito sites. The lethal traps' red velour strips were impregnated with bifenthrin (Bistar®), a residual pyrethroid insecticide, with each trap also containing half a lucerne pellet (Natty Cat Litter™) or 10 ml of hay infusion to attract egg-laying mosquitoes. Surveys of the nearest towns along the Stuart Highway were carried out to assess whether *Ae. aegypti* had spread from TCK, with sticky ovitraps set in Ali Curung (nine) and Mungkarta (three) on 27 February, Elliott (nine) on 16 March, and Renner Springs (four) on 22 March 2004.

Property-by-property receptacle survey and control

A first and subsequent second round of property inspections in TCK were commenced to survey and treat any receptacles potentially breeding *Ae. aegypti.* A total of 1,959 properties was visited between 22 February and 17 April 2004. A standardized protocol for entry, surveillance, and treatment of properties was established, and a field collection form used to capture all survey and control activities during every property inspection. All potential *Ae. aegypti* breeding sites were surveyed, with every section of a property inspected, including unsealed pipe fence posts, dense garden beds, ornamental ponds, animal water receptacles, roof gutters and down pipes, every tire in a pile of tires, old car bodies, and all manner of receptacles. Larval inspections were conducted using white painted soup dippers and pipettes or lengths of clear plastic tubing or pale buckets for obtaining samples of water. Larval mosquito samples were pipetted into clear plastic vials labelled in the field and stored later in the day in 70% alcohol. The field collection form consisted of data on property location and access, in and outdoor potential and actual breeding receptacles, presence and condition of rainwater tanks or septic tanks, number of larval samples taken, receptacle treatments, and the need for follow-up visits.

Field data were entered into an Excel spreadsheet, with data from *Ae. aegypti* positive properties entered into an existing ME Access database. Properties surveyed were marked on a series of NT cadastre maps, using lot numbers for property identification, and color-coded to indicate properties surveyed, properties positive for *Ae. aegypti,* and properties with no access. Results from individual maps were transferred daily to an operational overview map.

All actual and potential *Ae. aegypti* breeding sites were treated with bifenthrin or s-methoprene pellets or briquettes determined by the nature of the breeding site. Actual breeding receptacles were further treated with household bleach/ detergent as a 1% chlorine solution by pressure sprayer to kill mosquito eggs (Lamche and Whelan 2003). Receptacles associated with animals, used for human food consumption, and roof gutters were treated with bleach or s-methoprene, or both. Disused fishponds were treated with s-methoprene.

Bifenthrin was applied using a 5 liter handheld pressure sprayer, while bleach was applied using 1 liter handheld spray bottles. Two local pest control companies were commissioned to treat some industrial premises, including car wreckers' yards and receptacles at the TCK refuse landfill. In addition, bifenthrin was applied as a barrier spray to potential *Ae.*

aegypti harborage sites at relevant properties, including low vegetation and house walls near door entrances, around the outdoor laundry area, and other dark and moist areas close to entrances.

Following initial treatment, property owners and occupiers were supplied with a 350g 'Mortein' barrier outdoor surface spray can (manufacturer: Reckitt Benckiser NZ) (active ingredients: 3.5g/kg tetramethrin, 0.6g/kg lambda-cyhalothrin) and were requested to re-treat disused backyard receptacles four weeks after initial treatments to ensure continuous control between official treatment rounds.

The Alice Springs Town Council fogged all suburban streets in TCK in the afternoon and evening of 27 February to control any *Ae. aegypti* on the wing, using a vehicle-mounted Leco ULV fogging machine. A mixture of bioresmethrin (Reslin®) and diesel was dispensed at the label rate.

Rainwater and septic tank treatments and repairs

Unsealed rainwater tanks encountered during property inspections were treated with s-methoprene briquettes at a rate of one briquette per 5,000 liters. Property owners refusing this treatment were advised to carry out treatment with 100 ml kerosene. As Altosid® briquettes were not registered for rainwater tank treatment in 2004, an interim permit from the Australian Pesticides and Veterinary Medicines Authority was issued, with the product officially registered for this purpose in April, 2005. During March and April, 2004, a total of 96 rainwater tanks were inspected, with 70 receiving insecticide treatment. In addition, stainless steel mesh (1 mm aperture) and silicone was used to mosquito-proof any unsealed rainwater tank, including overflow and down pipes.

Unsealed septic systems were treated with s-methoprene pellets or fenthion (Baytex 550®), with the tank opening and associated pipes treated with bifenthrin to prevent mosquito ingress/egress. Fenthion was flushed into the septic tank via the toilet or a gully trap at a rate of 10 ml/treatment. Where required, septic absorption trench breather pipes and gully traps were sealed with aluminium fly mesh (1 mm aperture). In cases of extensive septic system damage, the property owner was requested to arrange for repairs.

Inspection and treatment of stormwater drains, road-side entry pits, telecommunication pits, and manholes

The majority of TCK's storm water drainage system consists of underground concrete-lined pipes that receive storm water from side entry pits along the roadside curbing, with water directed ultimately into one of two large open drains that lead to flood-out areas to the south of the town. All storm drain outfalls with dry season water presence were investigated for upstream sources of water, and attempts were made to reduce discharge into drains where possible. There were no natural waterways that directly receive water from the drainage system. Side-entry pits holding water were treated with liquid temephos (Abate 100E®) in late March by the Barkly Shire Council to control any potential *Ae. aegypti* breeding in the storm water drainage system, with a total of 40 liters of insecticide mix applied. To prevent subsequent breeding, roadside entry-pits were also treated with s-methoprene pellets or briquettes, with bifenthrin applied to the inner surfaces within the pits to prevent adult mosquito ingress/egress. The outfalls of large drains with water were inspected by crawling in the drains or inspecting manholes where possible to determine the presence of obstructions to drainage. In addition, an initial six telecommunications pits and 16 communications manholes were inspected for mosquito breeding, with 14 sites treated with s-methoprene briquettes and internal pit surfaces sprayed with bifenthrin.

Public communication

Media releases were issued to inform the public of the presence of *Ae. aegypti* in TCK, with a community service announcement on 25 February 2004 advising of imminent property inspections and fogging on 27 February. Further public awareness was raised through adapted Queensland Health dengue public awareness fact sheets made available to DHCS offices, local newspaper articles, public information and display sessions, and notices displayed at public notice boards, the health clinic, and local shop windows. Posters to prevent receptacles possibly containing *Ae. aegypti* eggs from being transported out of TCK were also displayed and later replaced by official road signs on roads in and out of the town.

A mail drop advised residents of a Barkly Shire Council hard-rubbish curbside collection on 21 March to reduce potential breeding receptacles, with hard-rubbish treated and buried at the local landfill. In addition, a CDC hotline was established within the first response week to address public queries, receive reports on backyard mosquito breeding, and receive advice on suitable property inspection times.

Mosquito identification and genetic analysis

All adult and larval mosquitoes collected during surveys were identified by ME staff using taxonomic keys (Lee et al. 1987), with lethal traps assessed by microscope for the presence of mosquito eggs. All data were captured in Excel spreadsheets and the existing Access database. Adult and larval specimens were sent to the University of Technology in Sydney for DNA analysis to determine the probable origin of the incursion (Beebe et al. 2005).

Aedes aegypti elimination program funding

Human resource, travel, and consumable costs of $211,700 were absorbed by DHCS during the initial response period. A request for $1 m financial support for a 12-month *Ae. aegypti* elimination program to commence in November, 2004 was forwarded by the NT Health Minister to the Commonwealth Department of Health and Ageing (DoHA). The application had support from the National Arbovirus and Malaria Advisory Committee, as the elimination of *Ae. aegypti* was considered of national importance. The budget submission was approved by DoHA and accepted by the NT Government on 15 June 2004, with the condition for the NT Government to match the $1 m with in-kind contributions. An official agreement was established, securing project funds to 30 June 2005.

Towards the end of 2004/05, a request for additional funding of $500,000 was forwarded to DoHA for program

continuation through a second wet season to detect any possible wet season-initiated breeding of *Ae. aegypti* from cryptic sources. A lower amount of $300,000 was received from DoHA for 2005/06, which enabled program continuation to confirm the elimination after two wet seasons without evidence of *Ae. aegypti*, however, with reduced human resource capacity.

Interim dry season *Aedes aegypti* surveillance (18 April to 21 November 2004)

Until the *Ae. aegypti* elimination program funds became available on 1 July 2004, only limited vector surveillance and control operations were conducted in the early to mid-dry season, with field elimination program activities fully commenced on 20 November 2004 (start of wet season) to maximize the likelihood of *Ae. aegypti* detection following rain. To keep residents informed, program updates were provided via radio interviews and notices in the local paper, with the start of the elimination program field activities announced on 19 November 2004. Lethal ovitrap surveillance continued, with a total of 109 and 53 traps set, respectively, by local EH or ME staff. Properties previously not accessed during the initial response period were followed up, with inspections and treatments also carried out on premises close to EVS or lethal ovitraps positive for *Ae. aegypti*, with a total of 60 properties inspected and treated. Rainwater tanks encountered during inspections were also surveyed. A second hard-rubbish curb-side collection was carried out by the Barkly Shire Council in October. To enhance overall exotic vector surveillance in the NT, routine ovitrap programs were established in Darwin rural, Palmerston, Jabiru, Katherine, and Alice Springs, in addition to existing programs in Darwin urban, Alyangula, and Nhulunbuy.

***Aedes aegypti* elimination program (22 November 2004 to 30 June 2006)**

The official program team was recruited and trained by November and consisted of a Darwin-based Program Director, Manager, and Technical Officer, and a TCK-based Senior Technical Officer and eight locally recruited TCK Field Officers. Office space, IT equipment, vehicles, insecticide storage space, and consumables, including insecticides, and surveillance and control equipment were also organized. EVS trapping was continued throughout the elimination program, with a total of 612 traps set. A further 177 sticky and 310 lethal ovitraps were set between June and September 2005.

Property-by-property receptacle survey and control

Between 22 November 2004 and 31 March 2006, six rounds of property inspections and control were conducted, with the aim to inspect all properties every six to eight weeks, coinciding with the time the residual insecticides remained active. However, due to access issues for some properties, inspection rounds overlapped.

During the elimination program, any detection of receptacle-breeding adult mosquitoes in traps, including *Cx. quinquefasciatus* and *Aedes* species, was considered an indication of an unknown source of mosquito breeding until follow-up adjacent property receptacle surveys could establish a possible source. The end of March was considered the end of the rainy season period, with 31 March 2006 the end of two wet seasons without evidence of *Ae. aegypti*. To confirm the absence of *Ae. aegypti* from other areas in the NT, a further 19 towns or communities were inspected in the Alice Springs, Barkly, and Katherine regions during the elimination program.

Rainwater and septic tank treatments and repairs

Septic systems present in 142 Aboriginal community buildings around TCK were inspected between August and September, 2005, with Julalikari Council, EH, and NT Department of Housing advised on maintenance required. In the interim, unsealed septics were treated with s-methoprene and bifenthrin. Inspection, treatment, and sealing of rainwater tanks on 133 properties was achieved between December 2004 and March 2006.

Inspection and treatment of stormwater drains, road-side entry pits, telecommunication pits, and manholes

The TCK stormwater drainage system was treated five times during the elimination phase (February, April, May/June 2005, September/October 2005, January 2006) using bifenthrin and s-methoprene. Silt blockages at outfalls and up-pipes were removed by cable attached bucket lines and earthmoving equipment by a local contractor to ensure free water flow. In addition, 511 and 432 telecommunication pits and manholes were surveyed for mosquito breeding and treated with bifenthrin and s-methoprene between 22-25 February 2005 and 23-28 February 2006, respectively, in liaison with telecommunication staff assigned to the program.

TCK landfill

Large areas at the TCK landfill contained uncovered dry and wet waste, including used tires and other discarded receptacles from the town. To remedy the situation, $50,000 of the *Ae. aegypti* elimination funds were dedicated toward a cooperative project between the Barkly Shire Council and the NT Department of Infrastructure, Planning and Logistics between March and June, 2005, to renovate the landfill into separate dry and wet waste areas, and waste coverage on a regular basis.

Public communication

Public communication continued through regular media releases of results and the need for vigilance and appeals for cooperation, including the weekly mosquito comic strip 'Millie Mozzie' produced for another program by the NT government and printed in the local TCK paper advising of public actions required.

RESULTS

***Aedes aegypti* initial response (17 February to 17 April 2004)**

Of the 59 EVS traps set during the initial response period, 11 traps were positive for *Ae. aegypti*. While none of the 30

Table 1. Number of properties and receptacles positive for mosquito larvae in Tennant Creek between 22 February 2004 and 31 March 2006.

Period of *Ae. aegypti* survey and control activities	Dates	Number of properties surveyed for *Ae. aegypti*	Number of properties with mosquito larvae	Number of times properties positive for larvae of receptacle breeding mosquitoes											Number of receptacles with mosquito larvae	Number of times receptacles positive for receptacle breeding mosquitoes										
				Aedes (Stg) aegypti	*Aedes (Stg) katherinensis*	*Aedes (Fin) notoscriptus*	*Aedes (Mac) tremulus*	*Anopheles (Cel) amictus*	*Anopheles (Cel) annulipes*	*Culex (Cux) annulirostris*	*Culex (Cux) crinicauda*	*Culex (Cui) pullus*	*Culex (Cux) quinquefasciatus*	*Tripteroides (Pol) punctolateralis*		*Aedes (Stg) aegypti*	*Aedes (Stg) katherinensis*	*Aedes (Fin) notoscriptus*	*Aedes (Mac) tremulus*	*Anopheles (Cel) amictus*	*Anopheles (Cel) annulipes*	*Culex (Cux) annulirostris*	*Culex (Cux) crinicauda*	*Culex (Cui) pullus*	*Culex (Cux) quinquefasciatus*	*Tripteroides (Pol) punctolateralis*
Initial response period (2004)	22/02/04 to 17/04/04	1959	116	88	0	10	24	0	4	3	1	7	56	3	140	110	0	10	26	0	4	3	1	8	63	3
Dry season (2004)	18/04/04 to 21/11/04	60	5	4	0	1	4	0	0	0	0	0	2	0	6	4	0	1	4	0	0	0	0	0	2	0
Elimination program (to end of 2004)	22/11/04 to 31/12/04	1052	73	2	0	1	20	0	3	0	0	0	57	1	102	2	0	1	26	0	4	0	0	0	77	1
Elimination program (2005, 1st quarter)	1/01/05 to 31/03/05	2523	91	0	0	1	33	0	16	7	0	0	59	0	117	0	0	2	41	0	16	8	0	0	75	0
Elimination program (2005, 2nd quarter)	1/04/05 to 30/06/05	1643	71	0	0	0	5	0	21	14	0	0	51	0	94	0	0	0	6	0	30	16	0	0	66	0
Elimination program (2005, 3rd quarter)	1/07/05 to 30/09/05	88	36	0	1	0	1	0	2	1	0	0	34	0	48	0	1	0	1	0	2	1	0	0	46	0
Elimination program (2005, 4th quarter)	1/10/05 to 31/12/05	974	160	0	0	0	6	0	7	5	0	0	154	0	239	0	0	0	7	0	8	6	0	0	233	0
Elimination program (2006, 1st quarter)	1/01/06 to 31/03/06	304	70	0	0	0	3	1	4	6	0	9	63	0	120	0	0	0	3	1	6	10	0	11	105	0
Totals		**8603**	**622**	**94**	**1**	**13**	**96**	**1**	**57**	**36**	**1**	**16**	**476**	**4**	**866**	**116**	**1**	**14**	**114**	**1**	**70**	**44**	**1**	**19**	**667**	**4**

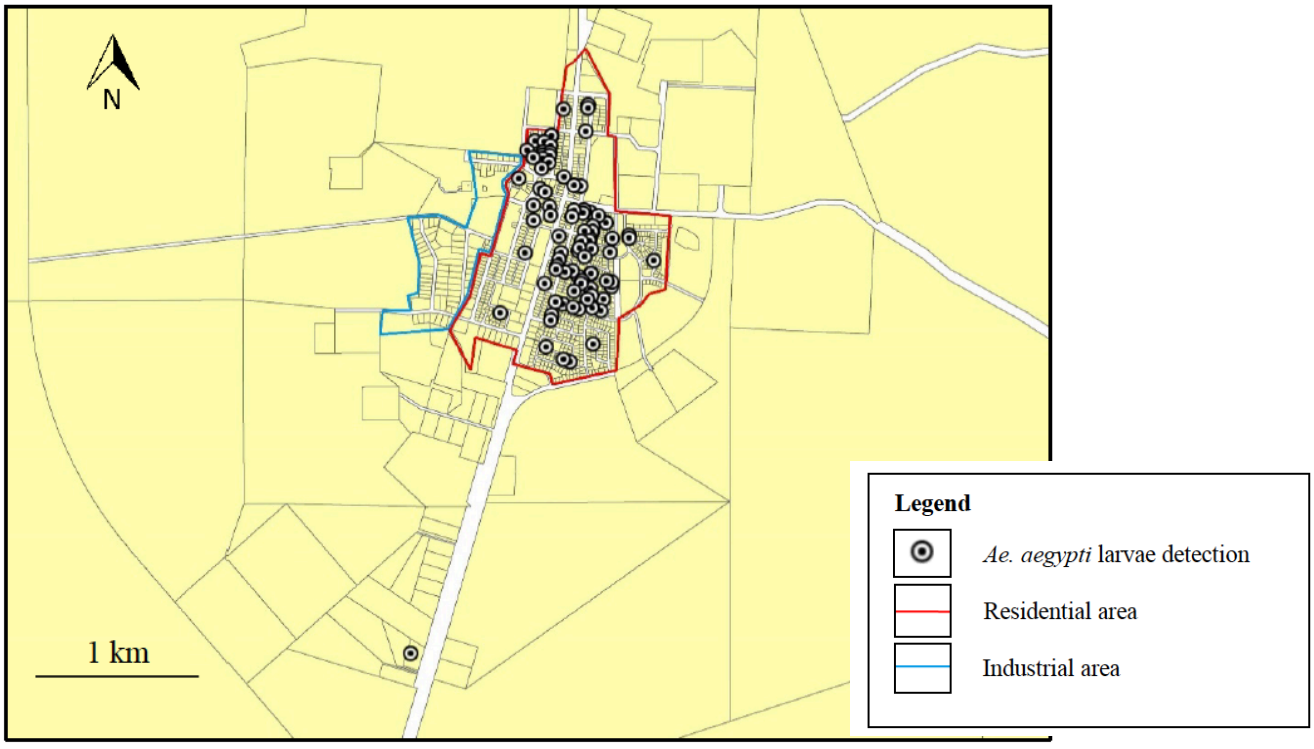


Figure 2. Tennant Creek properties positive for *Ae. aegypti* larvae during the initial response period.

sticky ovitraps set in TCK collected adult *Ae. aegypti*, eight of these were positive for *Ae. tremulus*, *Ae. normanensis,* and *Culex annulirostris*. None of the 25 sticky ovitraps set in the nearby communities collected adult mosquitoes. Of the 100 lethal ovitraps set, three were positive for mosquito eggs, with *Ae. aegypti* larvae reared from a trap collected on 16 April 2004.

During the initial response period, a total of 116 properties out of 1,959 were found breeding mosquitoes, with 88 properties positive for *Ae. aegypti* in 110 receptacles (Table 1, Figure 2). Positive receptacles included a 'Canadian' canoe, a synthetic cover on a utility car, children's toys, and pot plant drip trays. *Aedes aegypti* breeding was restricted to urban areas, except for one receptacle positive on a hobby farm located 3 km south of TCK. During investigations on subsequent property inspections, the majority of 'Mortein' barrier spray cans distributed to property owners were not used as advised.

Of the six telecommunication pits and 16 communication manholes inspected, two pits and five manholes held water, with two manholes positive for *Culex quinquefasciatus*. No *Ae. aegypti* breeding was found in any of the inspected rainwater tanks, septic tanks, road-side entry pits, telecommunication pits, or manholes. The mtDNA cytochrome oxidase 1 (CO1) gene that was used as a genetic marker in the DNA analyses of *Ae. aegypti* specimens confirmed their origin to be from Cairns, northern Qld (Beebe et al. 2005).

Interim dry season *Aedes aegypti* surveillance (18 April to 21 November 2004)

During this period, *Ae. aegypti* adults were captured in four out of the 53 EVS traps set, with the last adult *Ae. aegypti* collected on 4 November 2004. Of the 60 properties inspected, four had *Ae. aegypti*, with four out of six receptacles with larvae found positive for this species (Table 1). Two of the positive EVS traps triggered property inspections in the immediate vicinity, leading to the detection of two positive properties for *Ae. aegypti* breeding. One of the positive receptacles was a rainwater tank inspected on 18 October 2004. All 109 lethal ovitraps set in TCK during this period were negative for mosquito eggs. None of the enhanced ovitraps set in other NT regions were positive for *Ae. aegypti*.

***Aedes aegypti* elimination program (22 November 2004 to 30 June 2006)**

None of the EVS, lethal, or sticky ovitraps set during this period were positive for *Ae. aegypti*. Due to the overlap of property inspection rounds, actual round time frames and inspections were from 23 November to 29 December 2004 (round 1 – 997 properties inspected), 24 December 2004 to 8 March 2005 (round 2 – 1479 properties inspected), 1 February to 18 April 2005 (round 3 – 1343 properties inspected), 23 February to 16 May 2005 (round 4 – 1333 properties inspected), 27 May to 11 October 2005 (round 5 – 157 properties inspected) and 17 October 2005 to 31 March 2006 (round 6 – 1275 properties inspected).

Results are shown for each quarter of the elimination program in Table 1. *Aedes aegypti* larvae were only found on two properties in TCK between 22 November and 31 December 2004, with two receptacles positive for this species out of a total of 102 found breeding mosquitoes (Table 1). The last *Ae. aegypti* larvae were detected in a plastic crate on 30 November 2004. During the remaining elimination program, a further 5,532 properties were inspected, with a total of 618 receptacles found breeding mosquitoes other than *Ae. aegypti* (Table 1).

Of the 116 receptacles breeding *Ae. aegypti* in TCK, the majority were water storage receptacles (31), garden accoutrements (28), domestic or commercial usage receptacles (26), or discarded household items (13). Of these, receptacles most frequently found positive for *Ae. aegypti* were dog bowls (15), buckets (11), pot planters (9), pot plant drip trays (9), drums (9), bird baths (8), and tires (6). Receptacles less frequently with *Ae. aegypti* included evaporative air-conditioners, discarded coolers, boats, spas, tarpaulins, plastic shells, and general rubbish. During the entire program, only one rainwater tank was positive for *Ae. aegypti* larvae. Of the 511 telecommunication pits and communication manholes inspected in 2005, six were holding water, with one breeding mosquitoes, while in 2006 water was found in 11 pits and five manholes out of the 432 inspected, with one pit with larvae. No *Ae. aegypti* were found in the telecommunication pits or manholes.

DISCUSSION

The likely mode of transport of the *Ae. aegypti* incursion to TCK was as eggs in receptacles transported from Cairns in northern Qld, as evidenced by the types of receptacles found positive, including a canoe, a utility car tray cover, and pot plant drip trays that are all readily transportable receptacles, and DNA studies (Beebe et al. 2005). Following the detection, initial surveys showed that the species was widely distributed throughout the town, with 8% of the 1,107 properties surveyed in the first initial round being positive for the vector. Vazquez-Prokopec et al. 2010 suggested that a delayed detection response could potentially increase the number of dengue cases and costs to health systems exponentially. Thus, the decision to conduct a swift and dedicated elimination program to prevent its spread and to return the NT to its *Ae. aegypti*-free status has most likely resulted in a very large public health benefit in following years by preventing dengue establishment in the NT. The initial response was very quick and thorough, which was the key to a rapid reduction in infested receptacles and halting the spread of the vector in the town.

The pattern of *Ae. aegypti* positives in the initial period (Figure 2) indicates a relative lack of occurrence in the south-east residential area and in the western industrial area, indicating that the distribution was still in the process of expanding and that the introduction was relatively recent and probably not more than one-year-old. The property-by-property inspection and elimination measures had to include every single property, and without this attention to every property, eradication could not have been achieved.

Elimination was formally declared on 5 April 2006, just

over two years after it was first detected, without a subsequent detection until 2011, when *Ae. aegypti* was again detected in a TCK ovitrap, and successfully eliminated (Kurucz and Pettit 2013). However, while the genetic *Ae. aegypti* haplotype was the same during both incursions, the 2011 detection was assessed as a separate incursion from the detection in 2004, as it is highly likely that the continuous surveillance in the form of weekly EVS traps and annual backyard surveys carried out by the ME unit would have detected an ongoing presence of this species between 2006 and 2011 (Pettit and Kurucz 2014).

The steep reduction from 110 receptacles positive for *Ae. aegypti* in the first initial round, to only four, and then the last two positive receptacles during the dry season and at the end of 2004, respectively, was the key to achieving elimination. This indicates that at least three complete rounds of inspection and treatment are required for success in *Ae aegypti* elimination programs. The success can be attributed to the combined effect of residual insecticide applications to harborage areas and receptacles, the elimination of eggs in positive receptacles, and the immediate response by experienced staff. While the program received overall support from residents enabling property access, the response by many residents to assist with household insecticide applications was disappointing. However, this might have been largely due to a lack of a dedicated public relations strategy on this aspect at an early stage during the elimination program.

Rainwater tanks were initially suspected to be important *Ae. aegypti* breeding sites, especially during the dry season. However, other receptacles, particularly ones associated with animals, drums, pot plant drip trays, buckets, and tires were the most important breeding sites. Even during the dry season, when little rain occurred, there were ample receptacles with water that were breeding endemic mosquitoes, indicating there was no obstacle to dry season breeding and survival of *Ae. aegypti* in the town. The fact that *Ae. aegypti* was not found in storm drains, roadside entry pits, telecommunication pits, or septic tanks indicated a distinct preference for domestic receptacles, with frequent detection of *Ae. aegypti* in pot plant bases, garden accoutrements, tires, and buckets, indicating a potential mode of spread from one infested community to another when such items are transferred.

The elimination program appears to have also eliminated other receptacle breeding species. *Aedes notoscriptus* was last collected on 25 March 2004, *Ae. katherinensis* on 9 September 2005, and *Tripteroides punctolateralis* in Nov/Dec 2004 (Table 1). *Tripteroides punctolateralis* was collected again on 24 November 2011, *Ae. notoscriptus was* collected again on 12 February 2016, while *Ae. katherinensis* has not been recollected to date. The reappearance of *Ae. notoscriptus* and *Tr. punctolateralis* is probably from receptacles imported from other NT towns in much the same way as *Ae. aegypti* can be reintroduced from Qld. The failure to significantly reduce numbers of both *Ae. tremulus* and *Cx. quinquefasciatus* reflects possible non-receptacle underground storm-drain breeding sites for the latter and unlocated cryptic receptacle and possibly tree hole breeding sites for the former.

The intense adult mosquito trapping showed that EVS traps were the most efficient in detecting *Ae. aegypti* compared to lethal or sticky ovitraps. EVS trapping was also useful in detecting close-by *Ae. aegypti* breeding sites through detection of adult specimens. However, it is of concern that the presence of *Ae. aegypti* in TCK was only detected in EVS traps when the vector was already widely distributed throughout the town, indicating that this trap type is not as efficient for exotic vector surveillance compared to CO_2-baited Biogents® Sentinel traps or Gravid *Aedes* traps, the current mainstay for exotic mosquito surveillance, which were not available at the time of the 2004 incursion.

New surveillance and control measures introduced in recent years may influence how a similar response would be approached under current circumstances. The use of molecular testing and subsequent expenses for adult/larval/egg collections (Montgomery et al. 2017) may be a quick, easier, and cheaper surveillance tool, especially for larval presence, by taking water samples in rainwater tanks, other receptacles, and storm water drain systems. If the incursion had not been eliminated, *Ae. aegypti* would have become firmly established in TCK and could have soon spread to other NT locations, including Darwin, by road transport as adults or as eggs in receptacles (Whelan et al. 2001, Whelan et al. 2003). The occurrence of a vector of dengue in the high rainfall tropical city of Darwin, with its demonstrated vulnerability and receptivity for the vector and dengue disease (Whelan 1981, Kurucz 2013, Whelan et al. 2012) , would inevitably lead to outbreaks of dengue, with considerable public health impacts.

To date, there have been very few reported successful attempts to eliminate established populations of *Ae. aegypti* in the world (Gubler 1989, Gubler 1998), including the campaign in South America directed by the Pan American Sanitary Board from 1946 to 1970 resulting in *Ae. aegypti* elimination in a number of countries (Severo 1955, Soper 1963, Schliessmann and Calheiros 1974), and a program in the 1980s based on intensive insecticidal treatment and source reduction, which led to elimination of the vector from large areas of Cuba (Gubler 1989, Kouri et al. 1989). However, these programs were not sustained in the long term, similar to the initial elimination success around the Singapore airport (Chan 1972), and elimination attempts in Singapore in 1973, which achieved a 15-year low dengue incidence but failed to achieve elimination (Ooi et al. 2006).

The only long-term successful, purposeful elimination appears to be the elimination of the long-term establishment of *Ae. aegypti* from Brisbane Qld post-World War II by health legislation and the removal of rainwater tanks as perennial breeding sites (Trewin et al. 2017). However, the Brisbane program was not to eliminate a recent introduction, but rather a longer-term approach in NSW and southern Qld to eliminate the species in an otherwise suboptimal environment (Trewin et al. 2017). Elimination of *Ae aegypti* has recently been documented in the Torres Strait in Qld, when it was undetectable on most of the islands in the Torres Strait where it was previously present, but this was probably caused by displacement by the exotic species *Ae. albopictus* (Muzari et al. 2019), and not the result of a specific program for its elimination. The elimination presented here is the first

one in Australia of the elimination of a recently established population of *Ae. aegypti* by a comprehensive and verified short-term program to prevent further spread to other towns and cities.

The elimination, as well as the subsequent *Ae. aegypti* introductions and elimination successes on Groote Eylandt (a higher rainfall area in the tropic region) in the NT in 2008 (Whelan et al. 2009) and again in TCK in 2014 (Whelan et al. 2011, Pettit and Kurucz 2014), are examples of successful top-down programs where elimination is government directed and implemented, with similar response methods used following all three detections. While Gubler argued that a top-down approach (primarily by the government) without a combination of a bottom-up approach (primarily with the people) cannot work in mosquito control or elimination programs (Gubler 1989), this program has demonstrated that success, at least in Australia, and potentially in other places, as outlined by current occurrence of *Ae. aegypti* and the maps of potential spread (Kraemer et al. 2015, 2019, Ryan et al. 2019), indicates that a comprehensive, well-funded and well-executed top-down program, that also involves publicity and engagement with the people, can achieve success. The *Aedes* surveillance and control programs in the NT have been able to maintain an *Ae. aegypti*-free status in a demonstrated vulnerable and receptive geographic area for over 40 years, despite its historical presence and repeated importations (Whelan 1981, Beebe et al. 2009, Williams et al. 2010, Whelan et al. 2012, Warchot et al. 2020). The surveillance and elimination methods described here are possibly applicable to other regions in the world to eliminate or reduce the footprint of *Ae. aegypti.* Elimination programs require a comprehensive and organized task, rapid funding, experienced staff, and program activities for at least two consecutive rainy seasons to confirm the absence of the vector after the second season.

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