CABOMBA ERADICATION PROGRAM
REPORT FOR 2004/05

Northern Territory Government
Department of Natural Resources, Environment and the Arts

CABOMBA REPORT

2004/05

TABLE OF CONTENTS

1. INTRODUCTION

Cabomba, C*abomba caroliniana* is a fully submerged aquatic plant native to the South America. It is commonly found to be a problem in irrigation drains and channels where restrictions to water flow are typical of its impact. Cabomba was first recorded in Australia in 1967, probably as a result of being introduced through the aquarium industry trade.

Since its introduction to Australia, Cabomba is now found in various water storage facilities, farm dams and river systems in an area extending from Victoria north to the Charter's Towers/Townsville region in QLD.

In 1996 Cabomba was first recorded in the Northern Territory at Marlow's Lagoon, Palmerston. After numerous unsuccessful attempts at physical control over a period of several years after a single application of 2,4 – D n-butyl ester, the weed was eradicated.

Cabomba is listed in the top 20 weeds of Australia and as such is a Weed of National Significance. Under the Northern Territory *Weeds Management Act* 2001 the plant is a Class A (to be eradicated) and Class C (not to be introduced to the NT) weed.

On October 21 2004 cabomba was reported and subsequently positively identified in the Darwin River area. This is the second recording for the Northern Territory. Cabomba was found at several locations along an 11 km stretch of the river

Nationally, cabomba has proven to be a very difficult weed to effectively manage once established because of the rate at which it grows, the ease at which it spreads and establishes, and the difficulty of control. Furthermore only a single herbicide is currently registered for use on the species in Australia and the herbicide is not registered for use in potable water supplies. As a result of these issues and associated costs, management programs in most jurisdictions are limited to targeting impact reduction rather than eradication. Given the early stage of invasion and the enormous potential range and impacts for the species in the NT, eradication has been established as a priority.

2. POTENTIAL FOR NORTHERN TERRITORY

Cabomba is a Weed of National Significance because of its potential impact on a wide range of land uses and land use objectives. Cabomba has the potential to impact on the biodiversity and function of wetland and riparian ecosystems, water quality, water storage and distribution infrastructure and also recreation and amenity values.(National Weed Strategy: Cabomba 1999).

Infestations of cabomba have the ability to significantly reduce the capacity of water storage facilities, limiting availability of water supplies. If the storage is a potable water supply, cabomba may increase the cost of treatment. If the storage is for irrigation, increased costs may be incurred for the maintenance of the pumping and delivery infrastructure. Infestations pose a safety hazard and limits to swimming, recreational fishing and boating operations. Cabomba also affects native aquatic plants through competition and exclusion, which in turn influences native fauna.

(Figure 1: Distribution of cabomba, Darwin River, October 2004. Infestation indicated in red)

The single impact of cabomba if established in the Darwin river dam itself could feasibly be a requirement for the establishment of a drinking water supply treatment facility, costing in the order of $40 million, in addition to on going management costs.

It is currently impossible to accurately predict the potential scale of impact for the species in the Top End as accurate modelling programs for the species do not exist.

3. ESTABLISHMENT OF TASK FORCE

Cabomba has the potential to impact on a number of land use objectives, and in turn, control operations have the potential to impact on a number of land use expectations. In the case of the Darwin River infestation, potential existed for off-target herbicide impacts, domestic water supply issues, horticultural water supply issues, aquaculture water supply issues and recreational land use. This wide range of issues led to the formation of a multi agency taskforce in November 2004. Each member of the taskforce was involved in the implementation and/or management of a specific issue in relation to the survey and control program. (see: Table 1). This taskforce was responsible for directing, coordinating and overseeing the Cabomba program.

After initial contact with key stakeholder groups, representatives from the Traditional Owners of the area, the Kungarakany people, Kez Hall and Skei Batton were included in the Task Force. Kungarakany people own land which incorporates the largest single infestation and have registered sacred sites within the control area. Other stakeholder groups, including AFANT, Local Government and the NT Environment Centre were consulted on an as needs basis by the Task Force.

Table 1:

Agency	Business unit	Role
DIPE	- Biodiversity Conservation Unit - Weeds Management Branch - Advisory and Regulatory Services - Marketing and Communication - Transport and Infrastructure - Media and Communication Unit - Office of Environment and Heritage	- Monitoring riparian zone, aquatic fauna and flora. - Survey, control, monitoring, landholder liaison - Water quality monitoring - Media management/contact - Bund wall construction, - Media/public awareness - Environmental monitoring and advice
DBIRD	- Horticulture - Aquatic Pest Management Group -Fisheries and Aquaculture	- Industry liaison - Industry liaison - Industry Liaison
DHCS	- Environmental Health	- Drinking water quality monitoring,
PAW	- Power and Water Corporation	- Domestic water supply, dam quarantine

4. DELINEATING SURVEY

Subsequent to the report in October 2004, and formal identification by the NT Herbarium, a comprehensive riverbank and aerial survey of the freshwater section of Darwin River, its tributaries and two adjacent catchments was conducted to ascertain the exact distribution of the plant in the immediate area. In addition to this an airboat survey of Darwin River and Manton dam was conducted. The result of this survey was that Cabomba was only found at four discrete locations along Darwin River between Leonino Road and Cox Peninsula Road.

In addition to the field survey work, a “Cabomba Hotline” was set up for the purpose of generating public awareness as to the significance of the recording and also to determine the extent of further infestation. The result of the hotline was that Cabomba was positively identified at a further 13 locations in the Darwin area and at one location in Pine Creek. All of these reports were followed up with plants being removed and follow up monitoring of these sites being instigated. All of these positive reports were located in ornamental garden ponds or indoor aquaria and not in the natural environment.

In July 2005 an additional positive finding was made at Nhulunbuy located in a private fish tank. Survey in the area revealed no further infestation outside of "controlled" environments.

5. STRATEGY DEVELOPMENT

Given the limited distribution of Cabomba in Darwin River, the detrimental consequences to Top End waterways and water supplies, and the success of a previous eradication campaign at Marlow's Lagoon, the primary objective of the strategy was to eradicate Cabomba from Darwin River and to stop the further spread of the weed to other waterways. The only short-term option was the use of the herbicide AF Rubbervine Spray™ applied to the water which would make it unusable. The following key elements of the eradication program were implemented:

a) Control

There are a number of options available for the management of Cabomba including water level manipulation, shading, physical removal of plants, herbicides and biological control. To date the options of water level manipulation and physical removal have failed to manage the Cabomba infestation elsewhere in Australia, including the Northern Territory. In the case of the Darwin River site, water level manipulation and physical removal are not viable options due to the size and location of the infestation and potential presence of saltwater crocodiles.

The options for biological control which include the use of Chinese Carp and manatees (*Trichechus* spp.) while having proven successful in some areas overseas are not applicable to the current situation in the NT.

Physical removal of small backyard infestations is proving to be a viable option with positive results being experienced immediately. The exception to this is the Pine Creek site where the production of viable seeds on site has resulted in continual germination of seedlings over an extended period. The discovery of viable seeds at this site was the first record of this occurrence in Australia.

Experience gained through the successful management of the Marlow lagoon infestation in October 2002 indicate that the application of 2,4–D-n-butyl ester appears to be the most suitable option for the management of the species currently available for the Darwin River situation.

b) Extension/Education

Prior to the commencement of control operations an extensive public awareness program was implemented. The purpose of this aspect of the program was to raise public awareness as to the identification of the weed, the potential for the weed to spread in the Northern Territory, the importance of the quarantine aspect of the program in the reduction of weed spread risk, the information on the herbicide to be used in the program and potential for off target damage, the impact on domestic and horticultural water supplies and the potential impact on water users downstream of the control site.

An important element of this extension was the offer of potable water to households which draw water for domestic use from Darwin River.

c) Legislation

Cabomba is declared a Class A (to be eradicated) and Class C (not to be introduced to the NT) plant in the NT under the NT *Weeds Management Act* 2001 and given this, the Minister is able to make a number of undertakings in relation to the management of the species.

Under Section 21 of the Act the Minister declared a quarantine area on 9 November 2004 for a period of two years. There were two aspects to this declaration. Firstly, public physical access to the site was restricted to reduce the likelihood of inadvertent weed spread. Secondly, the pumping of water from the river was prohibited until herbicide levels returned to Australian Drinking Water standards.

d) Bund wall construction

Prior to the implementation of the chemical control program a bund wall was constructed at the lower end of the freshwater section of Darwin River at Cox Peninsula Road. The purpose of the bund was to prevent the flow of Darwin River water containing 2,4-D from entering Darwin Harbour. As the herbicide had not been used before in this context in Top End waterways the effect it may have on non-target organisms was unknown. Furthermore it was expected that oxygen levels in the river would decline to very low levels, and this along with the active herbicide may have an impact on the estuary.

The experience from Queensland where this herbicide has been used extensively and from the literature was that the herbicide would breakdown in a matter of weeks to levels that would be deemed safe to the estuary. The bund wall was a temporary safeguard to contain any impacts of herbicide application to the river.

e) Environmental and water quality monitoring

The concentration of the herbicide 2,4-D and its breakdown products were monitored to determine when it would be safe to release water from the retaining bund into the Middle Arm of Darwin Harbour, and to declare the river water safe for drinking. The method chosen to control Cabomba was based on the assumption of a modest and short-term impact on the water quality and biodiversity of the river. If this assumption was wrong, and there was in fact a major or long-term impact, then a different method may be needed for future control in Darwin River or elsewhere. The monitoring program investigated how serious the impact of the treatment was, and how quickly the water quality and biodiversity returned to normal, by focussing on certain water quality measures and wildlife groups (see table 2). Monitoring programs rely on sampling a small proportion of the study area. Where possible, the monitoring program for each group was designed to provide sufficient statistical power to be a reliable measure of the true state of the entire treatment area.

Table 2: Parameters studied in the monitoring program.

Group	Details
Water Quality	2,4-D, breakdown products, dissolved oxygen, bacteria (E. coli)t
Macro-invertebrates	Samples from the water column, and pool bed in centre and edge
Aquatic plants	Submerged and floating aquatic plants and streamside plants
Fish	Captured by a variety of methods
Crocodiles	By spotlight survey from a boat
Turtles	Captured in traps
Birds	Those foraging on or over the water and in streamside vegetation

6. IMPLEMENTATION OF STRATEGY

a) Control Program

In November 2004 the cabomba infestations on Darwin River comprised of 3 relatively small infestations upstream of the Old Bynoe Road crossing, and a single large continuous infestation located in Lok Landji billabong further downstream approximately 2 kilometres in length.

Details of AF Rubbervine Spray ™ and use are outlined in the table below. The application of AF Rubbervine Spray™ (2,4 – D n – buytl ester) + diatomaceous earth to the cabomba infestations on Darwin River commenced on the 11th of November. Control operations initially targeted small upstream sites where infestation size limited operation to single boats. Upon completion of these sites both control crews moved downstream to the major infestation where more coordinated efforts were required.

Prior to the commencement of herbicide application all landholders drawing water from the river were contacted and appropriate arrangements to supply alternative water were made. All pumping infrastructure in the river was physically disconnected to ensure that no inadvertent use of treated water occurred.

Herbicide: AF Rubbervine Spray ™ (2,4-D n-butyl ester, 800g/lt active ingredient.)

Registration detail: 2,4-D n-butyl ester is registered for the control of C*abomba caroliniana* in an aquatic situation in the Northern Territory. 2,4 D n – butyl ester is NOT registered for use in potable water supplies.

Application rate: 12.5 lt product + 5 kg diatomaceous earth mixed in 200 lt water applied to a megalitre of water.

Restrictions: **(From manufacturers label)** For spot application to scattered Cabomba patches. Add 12.5 L to partly filled 200 lt tank. Pre-mix 5kg diatomaceous earth in 10 lt water and add to tank. Top up, agitate thoroughly. Inject required dose through submerged nozzles into Cabomba biomass. Do not treat whole of water at once. To be used in non-potable water.

Effective dilution: 12.5 lt X 800g/lt = 10,000 g /megalitre

- this equates to 1g/100 lt water
- this equates to 10 mg/ lt of water

Figure: Images above and below illustrating Cabomba control operations on Darwin River November 2004.

The (2) control crews comprised of two Weed Management Officers, one a boat operator and the other a herbicide applicator. Each boat was a punt approximately 12 ‘ in length powered by a three horsepower outboard. Each boat was fitted with a 100 litre tank and a petrol pump used to source river water and apply herbicide.

The front of each boat was fitted with a boom used for application in more open water situations and also a hand held “wand” to be used in more confined situations. Herbicide, fuel, safety equipment and diatomaceous earth were also carried in each boat in order to increase the level of efficiency of control operations. Booms were calibrated in order to allow the application of herbicide to the three dimensional waterbody in an effort to facilitate the application of herbicide to within label recommendations.

Herbicide was specifically applied to visible infestations in order to minimise potential off target impacts and reduce the volume of product applied to the environment. This was a different approach to that used in the Marlow Lagoon exercise in 2002 where water levels and the “controlled” nature of the site allowed complete treatment of the affected water body. At no time was an attempt made to treat the entire water body.

The initial application of herbicide to the entire cabomba infestation took two days. Control operations were very difficult with staff operating in very hot, humid, noisy, cramped conditions for extended periods of time.

On the 17th of November and again on the 25th of November the entire remaining cabomba infestation was re-treated in order to ensure “missed” plants did not remain untreated and potentially allow the re-infestation of the river.

In December 2005 Tom Anderson a weed management scientist from the Queensland Department of Natural Resource Management and Mines visited the Northern Territory in order to assess the situation, review herbicide application techniques and provide potential advice that may be used in finetuning control operations in the future. The overall impression subsequently recorded in his field inspection report indicate that NTG operations appeared to be technically correct, successful and that efforts should continue in order to achieve the objective of eradication.

During the herbicide application period, and between herbicide application events the entire cabomba population was monitored for herbicide impact. General observations were that the application was successful as plant populations, and plant vigour were significantly effected. On December 17th a single untreated cabomba plant was observed remaining in Lok Landji billabong – this plant was subsequently treated.

After each herbicide application the cabomba was observed to “break off” and float in large mats away from infestations connected to the substrate. Efforts were initially made to collect this material however this soon became overwhelming and this material was treated in a manner similar to the “untreated” sites.

In January 2005 a short inspection of the main infestation and the site immediately upstream of the Old Bynoe Road crossing indicated cabomba to be still present at levels approximately 20% of those encountered at the commencement of control operations. In addition to this a significant amount of free floating plant material was observed in the main billabong that had developed “roots”. In some areas, such as the area immediately adjacent to the boat ramp, these plants were observed reattaching themselves to the substrate and resuming normal growth habit.

In May/June 2005 a complete ground based survey of the entire length of Darwin river was conducted in addition to a boat survey of the main billabong. These surveys resulted in indications that the cabomba infestation has further increased to a level of approximately 60 – 70% of that encountered in October 2004.

b) Environmental Monitoring

The Cabomba infestation in Darwin River was confined to four pools along the main channel, but was particularly concentrated in one pool 2.2 long known as Lok Landji billabong. There were also at least four similar (uncontaminated) pools upstream of all of the infested pools, which could act as controls.

Table 3: Sites used for sampling

Pool	Easting	Northing	Length	No of sections	Notes
CON1	712823	8581779	680	3	Control, 1 km downstream of Dam wall
CON2	712829	8582403	470	3	Control
CON3	714262	8585031	510	3	Control
IMP1	714902	8590081	2.2 km	6	Impact, site of main infestation
IMP2	715471	8586848	120 m	1	Impact
IMP3	714015	8585811	320 m	2	Impact

The monitoring program followed a BACI design: Before-After Control-Impact (Bernstein and Zalinski, 1983, Underwood, 1993). Three infested and three control pools were surveyed for birds before and after the 2,4 D treatment (Table 3). All of the pools were biophysically similar: they were approximately 10 m wide, 3 m deep and densely lined by vegetation dominated by *Pandanus aquaticus*. The Before survey took place on the 4th and 5th of November, 2004 and the After survey on the 14th and 15th of December. Both were during the early wet season when conditions are hot and humid but before significant rain has occurred river flow is at its annual minimum. There were a number of rainfall events between the two samples but the river flow and water quality did not change.

Each pool was subdivided into 200 m sections, defined by the distance from the point of entry, using a GPS. 200 m was selected as the section length because this is the approximate scale of the home range of the smaller birds (flycatchers and kingfishers). The end sections of each pool were usually less than 200m in length. Those shorter than 100 m long were ignored, but sections between 100 and 200 m were treated as full sections. There was only one section in the shortest pool and six in the longest, and in total there were nine sections in both the Control and Impact pools. Each pool was traversed in a small boat travelling at about 1 km/h, and two observers recorded all of the birds seen or heard over the water or using the riparian vegetation. The boat was stopped where necessary to aid bird identification. The pool was surveyed on the outward and return trip, and two separate visits were made in one day. Thus, each pool section was surveyed four times, and the two visits were timed so that for each pool, one was close to dawn or dusk and the other closer to midday.

Data were summarized as the mean number of birds of each species recorded in each section. While 62 species of birds were recorded during the two surveys, the analysis concentrated on four species and species groups (Table 4). These were chosen because they were species that were dependent for food on aquatic animals (which may be affected by the treatment) and there were sufficient numbers for analysis. Species were grouped because many individual species had too few records to analyse.

Table 4: species and groups used for the monitoring program.

Species/Group	Description[#]	Total number observed
Shining Flycatcher	Insectivore in riparian and rainforest vegetation	127
Kingfishers	Azure Kingfisher only occurs near water and eats fish. Forest Kingfisher also occurs in woodlands and rivers. The recorded diet includes dragonflies and frogs but not fish.	36
Herons	Nankeen Night-heron and Black Bittern both eat fish and crustaceans	16
Cormorant and Darter	Little Pied Cormorant eats fish and crustaceans and Australian Darter eats mostly fish	14

[#] Dietary information from Marchant and Higgins (1990) and Higgins (1999).

For analysis, the before and after treatment visit to each river section was considered as a sample (n = 36). For each sample abundance was calculated as the mean number of birds recorded over the four surveys for the visit. The data were analysed as a Repeated Measures Anova (Green 1993, Price 2004), with Before/After as the repeated measure and Control/Impact as the independent factor. The analysis tested the effects of Before/After, Control/Impact and their interaction, and a significant interaction was interpreted as evidence of an impact of the 2,4 D treatment. The analyses were conducted using Statistica v 6.0 software.

- Environmental Monitoring Results

All four species groups were recorded in control and impact sites both before and after treatment (Figure 2). This was also the case for pool IMP1, which was the most heavily treated pool. In fact, all four bird groups actually increased slightly in abundance after treatment in the Impact pools. The graphs for each group show that bird abundance is approximately constant across all treatments. For example, the mean number of Shining Flycatchers recorded per section was between 0.7 and 1.2 for all four treatments.

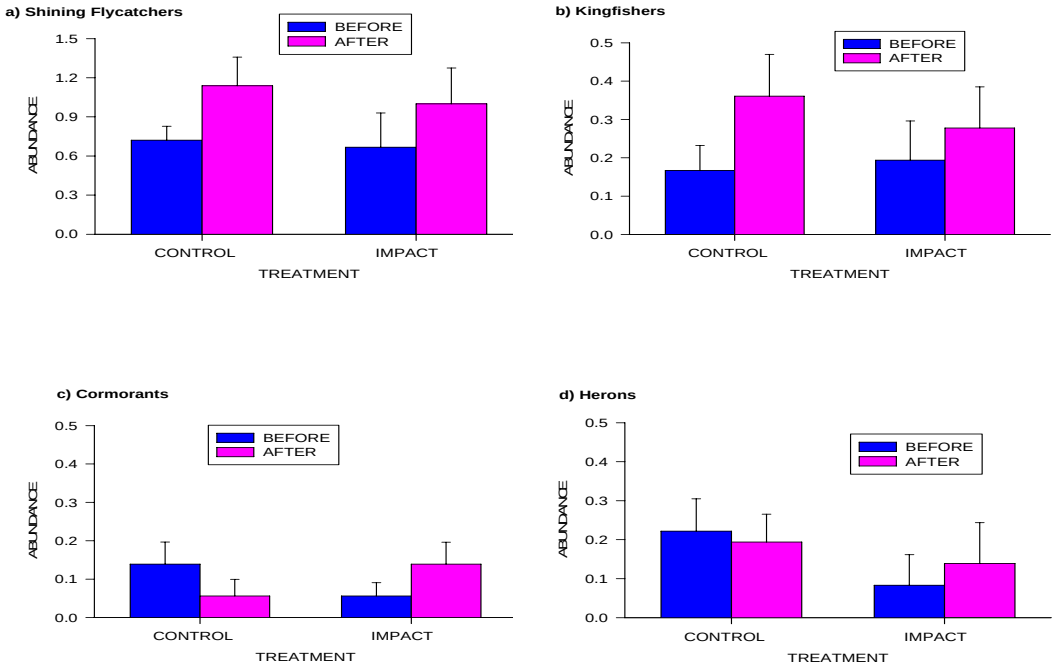

Figure 2: Survey results for four species groups. The abundance measure is the mean number of birds observed per 200 stretch from four repeat observations number of birds observed per 200 stretch from four repeat observations.

There were no statistically significant effects of the two treatments or of their interaction for any of the four bird groups (Figure 2). However, the interaction terms for Shining Flycatchers and kingfishers were significant when the abundance values for the Impact/After samples were reduced to 10% of their actual values (Figure 2). Thus, this monitoring design would have been capable of statistically demonstrating a 90% decline in these two most common species groups.

Vertebrates:

Crocodiles were surveyed in the four treatment and three control pools using a standard spotlight method. The pre-treatment survey was during the week of the 2nd of November and the post-treatment survey on the week of December 13th. The data from the survey has not yet been analysed, but live crocodiles were observed in the treatment pools IMP1 and IMP2 both before and after treatment.

Five Yellow-faced Turtles (*Emydura tanubaraga*) were trapped in Lok Landji pool (the main treatment pool) for temporary removal using 10 crab traps baited with meat for three nights. The same method was used to trap turtles after the treatment, during the week of the 13th December, capturing a further five Yellow-faced Turtles. Note that the original five turtles had not been returned to the river by the time of the second survey.

Macroinvertebrates:

A pre-treatment macro-invertebrate survey was undertaken in the same pools and on the same dates as the other animal monitoring. At the time of writing this report, the invertebrates in the samples were still being identified. A post-treatment sample is intended for June 2005.

Flora:

The same seven pools were monitored as for the birds. Four random plots were sub-sampled at each of the pools. In all there where twenty eight plots measured in the seven sites. Each plot position was recorded using a GPS. Plots were five metre by one metres by water depth.. Each pool was traversed in a canoe. The canoe was stopped to survey the plots and water depth, vegetation cover from bank, dominants from the river bank, vegetation cover from the top of the water column and floristics where recorded. The submerged flora not visible from the surface was sampled by dragging weighted hooks through plots. The same methods where employed at both the Control and Impact pools.

The pre-treatment survey was conducted during the week beginning the 2nd November 2004, and the post-treatment survey during the week of 20th December. A third survey is proposed in November 2005 to determine the extent of recovery of the flora.

The data from the surveys has not yet been analysed. However, the general patterns observed from each plot are summarised in Table 5.

Table 5: Repeated measures results for the treatments and their interactions.

Variable	SS	Df	MS	F	P
Shining Flycatcher					
Intercept	28.0017	1	28.0017	70.8967	0.0000
c_i	0.0851	1	0.0851	0.2154	0.6488
Error	6.3194	16	0.3950		
B_A	1.2656	1	1.2656	1.6364	0.2191
B_A*c_i	0.0156	1	0.0156	0.0202	0.8887
Error	12.3750	16	0.7734		
Kingfisher					
Intercept	2.2500	1	2.2500	12.8317	0.0025
c_i	0.0069	1	0.0069	0.0396	0.8448
Error	2.8056	16	0.1753		
B_A	0.1736	1	0.1736	3.2258	0.0914
B_A*c_i	0.0278	1	0.0278	0.5161	0.4829
Error	0.8611	16	0.0538		
Herons					
Intercept	0.9184	1	0.9184	5.9606	0.0266
c_i	0.0851	1	0.0851	0.5521	0.4682
Error	2.4653	16	0.1541		
B_A	0.0017	1	0.0017	0.0851	0.7742
B_A*c_i	0.0156	1	0.0156	0.7660	0.3944
Error	0.3264	16	0.0204		
Cormorants					
Intercept	0.3403	1	0.3403	7.5385	0.0144
c_i	0.0000	1	0.0000	0.0000	1.0000
Error	0.7222	16	0.0451		
B_A	0.0000	1	0.0000	0.0000	1.0000
B_A*c_i	0.0625	1	0.0625	4.0000	0.0628
Error	0.2500	16	0.0156		

Table 6: The significance of the interaction term when Impact/After abundances are artificially reduced. The values in the table are the F statistic and p value (in parentheses).

Species Group	I/A at 50%	I/A at 25%	I/A at 10%
Shining Flycatcher	1.50 (0.239)	3.76 (0.0703)	5.89 (0.0274)
Kingfishers	2.61 (0.126)	3.86 (0.0671)	4.55 (0.0488)
Cormorants	1.90 (0.187)	0.841 (0.373)	0.369 (0.552)
Herons	0.0220 (0.884)	0.0394 (0.845)	0.124 (0.719)

c) Fish Monitoring

Fish abundance and diversity were assessed using a combination of gillnets and electro-fishing in two treatment sites and four control sites. The combination of methods was required to effectively sample the range of fish in all habitats.

At each site, two 35m multi-panelled gillnets were set at a 45º angle from a bank for a period of two hours, from 3pm to 5pm. A distance of 25m – 100m (depending on length of site) separated the two nets. Sampling with gillnets was replicated after 48 hours at each site.

Two areas within each site were sampled for a ten-minute period using an electro-fisher. The two areas represented either areas of shallow inflow or outflow, or pandanus lined deep edges. Sampling with the electro-fisher was unable to be replicated.

Two of the control sites were upstream of the impact sites on Darwin River, while the other two control sites were located on the Blackmore River which was unaffected by cabomba. These sites were chosen due to their mirroring of the Darwin River control sites.

The majority of captured fish were returned alive to the area of capture, although several specimens from each species were preserved for future reference or to assist in identification.

During the monitoring program there was no evidence of fish kills or detrimental effects on fauna, and there was little or no reduction on dissolved oxygen levels in the estuary.

In addition to these sites water samples were collected from each prawn farm operating in the area on two occasions (22 December 2004 and 2/4 February 2005). Two water samples from each farm were collected under the direction of farm staff. Analyses were performed by National Measurement Institute in Sydney. All results were below the levels of detection for 2,4-D and both breakdown products. This shows that these products did not enter these farms with intake water.

A total of 359 fish from nineteen species and 12 crustaceans from two species were recorded at the six sampling sites (Table 7).

The five most abundant species each contributed more than 5 % to the total number of fish caught and together accounted for 85 % of all fish.

Although the abundance of fish varied from site to site, five of the most abundant fish species were sampled from all the control sites (C1, C2, C3). Four of the same species were sampled in each of the impact sites (IMP1, IMP2, IMP3).

All sites excluding IMP3 had similar species richness. Only 5 species of fish were sampled from IMP3 compared to an average of 10 species from each of the other sites.

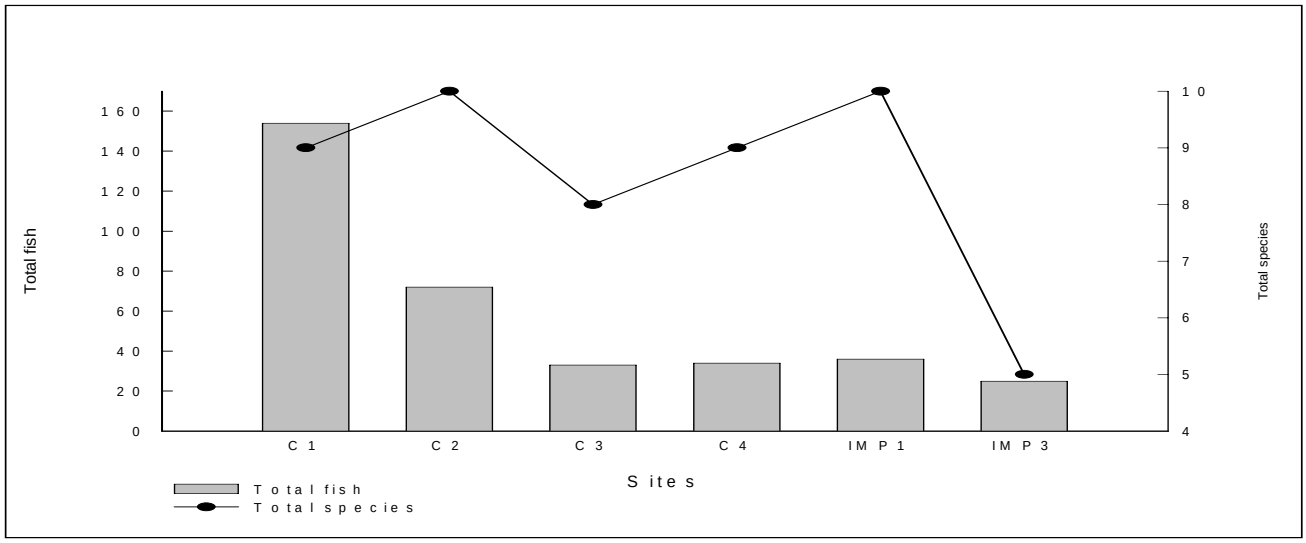

Figure 3 Total numbers of individual fish collected at each site and the number of species recorded from each site sampled

Table 7 Details of the species collected across all sites

Rank	Genus and Species	Common Name	Number of fish (n)	(%)
1	*Nemataolosa erebi*	Bony Bream	192	53.5
2	*Glossamia aprion*	Mouth Almighty	35	9.7
3	*Neosiluris ater*	Eel-tailed Catfish	35	9.7
4	*Amniataba percoides*	Barred Grunter	23	6.4
5	*Melanotaenia splendida australis*	Chequered Rainbowfish	19	5.3
6	*Megalops cyprinoides*	Tarpon	12	3.3
7	*Hypseleotris compressa*	Empire Gudgeon	9	2.5
8	*Craterocephalus stercusmuscarum*	Fly-specked Hardyhead	7	1.9
9	*Ambassis macleayi*	Glassfish	4	1.1
10	*Melanotaenia nigrans*	Black-banded Rainbowfish	4	1.1
11	*Ophisternon gutterale*	Swamp Eel	4	1.1
12	*Oxyeleotris selheimi*	Giant Gudgeon	3	<1
13	*Toxotes chatareus*	Archerfish	3	<1
14	*Glossogobius aureus*	Golden Goby	2	<1
15	*Mogurnda mogurnda*	Purple-spotted Gudgeon	2	<1
16	*Stronylura kreffi*	Freshwater Longtom	2	<1
17	*Denariusa bandata*	Penny Fish	1	<1
18	*Hephaestus fuliginosus*	Sooty Grunter	1	<1
19	*Redigobius chrysosoma*	Crimson-Tipped Gudgeon	1	<1
Total Fish			**359**	
1	*Macrobrachium sp.*	Macrobrachium	10	
2	*Cherax quadricarinatus*	Redclaw	2	
Total crustaceans			**12**	

Table 8 Presence absence of all species sampled from each of the six sites.

	C1	C2	C3	C4	IMP1	IMP3
Nemataolosa erebi	*	*	*	*	*	*
Glossamia aprion	*	*	*	*	*	
Neosiluris ater	*	*	*	*	*	*
Amniataba percoides	*	*	*	*		*
Melanotaenia splendida australis	*	*	*	*	*	*
Megalops cyprinoides	*	*		*		
Hypseleotris compressa			*		*	
Craterocephalus stercusmuscarum	*	*			*	
Ambassis macleayi		*				
Melanotaenia nigrans					*	
Oxyeleotris selheimi			*	*		
Toxotes chatareus	*	*				
Glossogobius aureus					*	
Mogurnda mogurnda		*		*		
Stronylura kreffi					*	
Denariusa bandata					*	
Hephaestus fuliginosus	*					
Redigobius chrysosoma			*			
Ophisternon gutterale				*	*	
Neosiluris sp.						*
Macrobrachium sp.	*		*	*		
Cherax quadricarinatus	*					

d) Water Quality Monitoring

Water quality surveys of Darwin River, adjacent bores and Darwin River estuary were undertaken before, during and after the application of 2,4-D to the Cabomba infestation sites. The following addresses these three water bodies, and focuses on concentrations of 2,4-D, the two breakdown products and dissolved oxygen levels. In the case of Darwin River bacteriological analyses are also presented as this was an important measure addressing drinking water standards.

Two laboratories were used for the analysis of 2,4-D and the breakdown products 4-chlorophenol and 2,4-dichlorophenol. The National Measurement Institute (NMI) in Sydney was used for the measurement of all three compounds to trace levels (1, 0.1 and 0.1 ug/L, respectively). NMI is a NATA accredited laboratory and was used to measure levels of the three compounds to levels well below national guideline levels for drinking water in the Australian Water Quality Guidelines (1996). Reporting time was generally 2 weeks.

The DBIRD laboratory at Berrimah Research Farm was used for the analysis of 2,4-D. This laboratory provided a service with a rapid turnaround time (hours) which was crucial for 'real-time' monitoring of levels in waterways needed to inform decision making by the Taskforce. A detection level of 100 ug/L was initially provided which was adequate to address environmental levels of 2,4-D, and subsequent refinements to methodology enabled levels below drinking water

guidelines (30 ug/L) to be reported. The DBIRD Laboratory also undertook analyses for E.coli, a measure of bacterial numbers in water.
Dissolved oxygen was measured onsite using a Hydrolab Surveyor multi-parameter probe which was calibrated on a weekly basis.

- Groundwater Monitoring:

Prior to the application of 2,4-D (November 2004) bores along Darwin River were selected for monitoring. These bores were selected on the basis of proximity to Cabomba sites and being equipped so that samples could be taken. These were re-sampled in February 2005, along with an additional 5 bores. All samples were sent to NMI in Sydney for low-level analyses of 2,4-D and breakdown products 4-chlorophenol and 2,4-dichlorophenol.

In every sample concentrations of all compounds were below laboratory detection levels stated above. This indicates that there were no traces of 2,4-D or breakdown products before or after the application of herbicide to Darwin River. Note that the Australian Drinking Water Guidelines (1996) state that levels of 2,4-D should be below 30 ug/L and the breakdown product 2,4-dichlorophenol should be at concentrations below 200 ug/L. The laboratory detection level of NMI is well below these recommended drinking water levels. Note that there is no guideline value for 4-chlorophenol.

Table 9 Groundwater monitoring locations.

Location	November 2004	2-3 Feb 2005
Springs d/s Cox Pen. Rd	Sampled	Sampled
RN25905	Sampled	Sampled
RN26278	Sampled	Sampled
RN27459	Sampled	Sampled
RN27723	Sampled	Sampled
RN32661	Sampled	Sampled
RN9134	Sampled	Sampled
RN21865	Not sampled	Sampled
RN25655	Not sampled	Sampled
RN30893	Not sampled	Sampled
RN31095	Not sampled	Sampled
RN33782	Not sampled	Sampled

- Darwin River Monitoring - Pre-treatment:

Sample collection occurred before the application of herbicide to Darwin River to determine pre-treatment river condition. Triplicate water samples were collected from the main infestation site (site 1). Duplicate water samples were collected at the three remaining smaller infestation sites (sites 2, 3 and 4) and single samples were collected from three additional sites (sites A, F and G). Refer to table 10. Samples were analysed for 2,4-D and breakdown products by NMI. All compounds were found to be below the laboratory level of detection.

- Darwin River Monitoring - During Treatment:

The focus of the monitoring effort in Darwin River was Site 1 and the Bund Wall site. The former was the main Cabomba infestation site and the largest pool treated with herbicide. This pool is also the most downstream site and closest to the estuary and the bund wall. The application of herbicide at the other three infestation sites was relatively small, and the herbicide was quickly diluted and transported from the site by water movement.

The following discussion is limited to the main infestation site (Site 1) and the Bund Wall site. These sites are in order of upstream to downstream. Results will focus on levels of 2,4-D, breakdown products and dissolved oxygen.

The following figure (figure 4) shows concentrations of 2,4-D measured at two locations on Site 1 and at the Bund Wall.

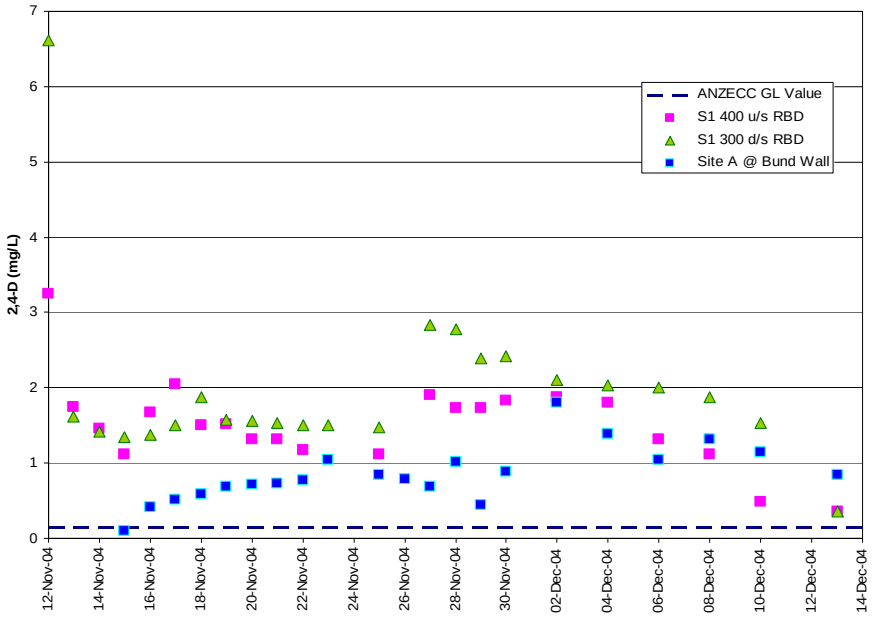

Figure 4

Concentrations of 2,4-D at Site 1 (S1) were very high on 12 November immediately following the first application of herbicide to the edges of the pool. Levels quickly declined as 2,4-D was mixed in to the main water body through wind action. Levels of 2,4-D rose again on 27-28 November after the second application of herbicide to the pool.

Levels of 2,4-D were well above the ANZECC/ARMCANZ (2000) guideline value of 140 ug/L (0.14 mg/L) indicated by the dashed line. This was the case at all four monitoring sites. The expectation was that concentration of 2,4-D would decline as bacterial and chemical processes degraded 2,4-D to the two breakdown products. This did not occur. Confirmation that 2,4-D was not decomposing was indicated by: very low levels of breakdown products measured during the first week of application (4-dichlorophenol was always below 0.1 ug/L; 2,4-dichlorophenol was less than 10 ug/L); and very low concentrations of breakdown products relative to 2,4-D levels in the estuary following release of bund water.

Therefore the only mechanism by which 2,4-D levels declined was through dilution within the pools and dilution with water flowing down the river following early wet season rains. Despite high levels of 2,4-D remaining in this water body for a substantial period of time, there appeared to be little or no impact on the fauna or fauna (refer to other relevant sections).

The second main concern surrounding the application of herbicide was the effect this may have on levels of dissolved oxygen (DO). The following figures (Figures 5 and 6) show levels of dissolved oxygen at two locations in Site 1. Following the application of 2,4-D on 12 November 2004 there was an immediate reduction in levels of dissolved oxygen. Median levels of DO declined to around 2 mg/L from a level of approximately 5 mg/L prior to the application of herbicide. Minimum levels of zero were measured at both locations, especially at 300 m downstream of Reedbeds Road. These very low values were measured towards the bottom of the water body.

Following the rapid initial decline in DO levels an aerator was activated on 14 November 2004. Coincident with this, levels of DO at the 300 m downstream site stabilised, with median values at around 2 mg/L. Minimum levels at the bottom remained at or near zero. At the 400 m upstream site DO levels also stabilised to between 2 and 3 mg/L, and minimum levels usually exceeded 1 mg/L.

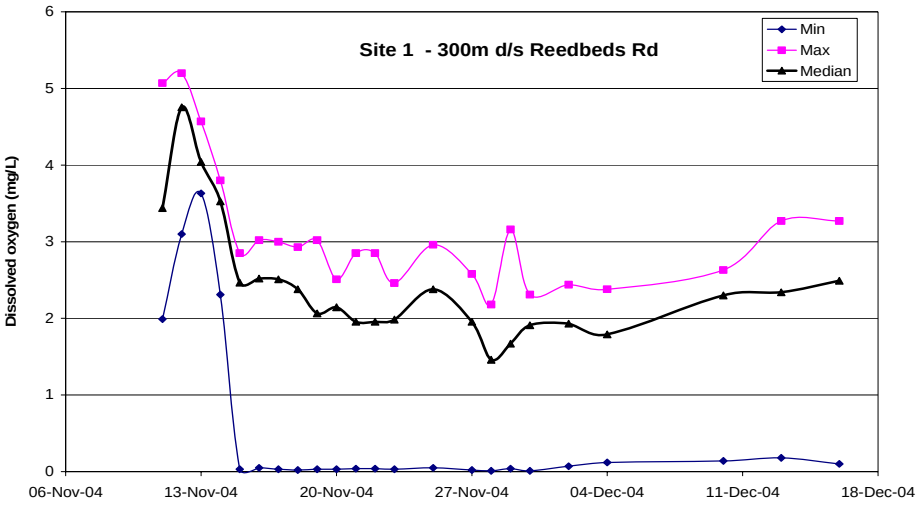

Figure 5

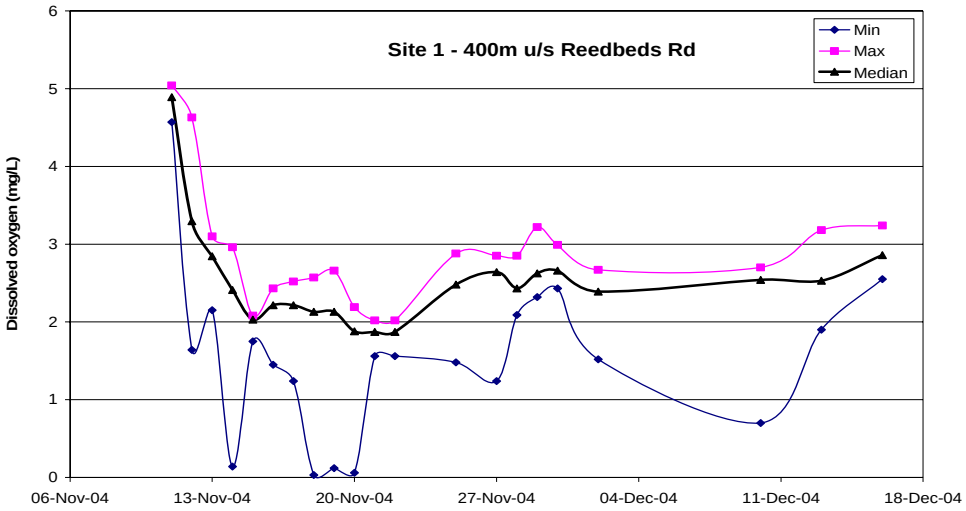

Figure 6

At the bund wall site (see Figure 7) there was a reduction in DO levels coincident with the application of herbicide. Treated water from upstream Site 1 pooled behind the bund wall in an incised location. At this location, saline water from the last high tide was also trapped by the bund wall at the bottom of the pool. This combination of factors resulted in the rapid decline of DO. An aerator was installed at the site and activated on 14 November 2004. There was an immediate increase in DO levels, with minimum values in excess of 3 mg/L, and similar to levels measured before the application of 2,4-D.

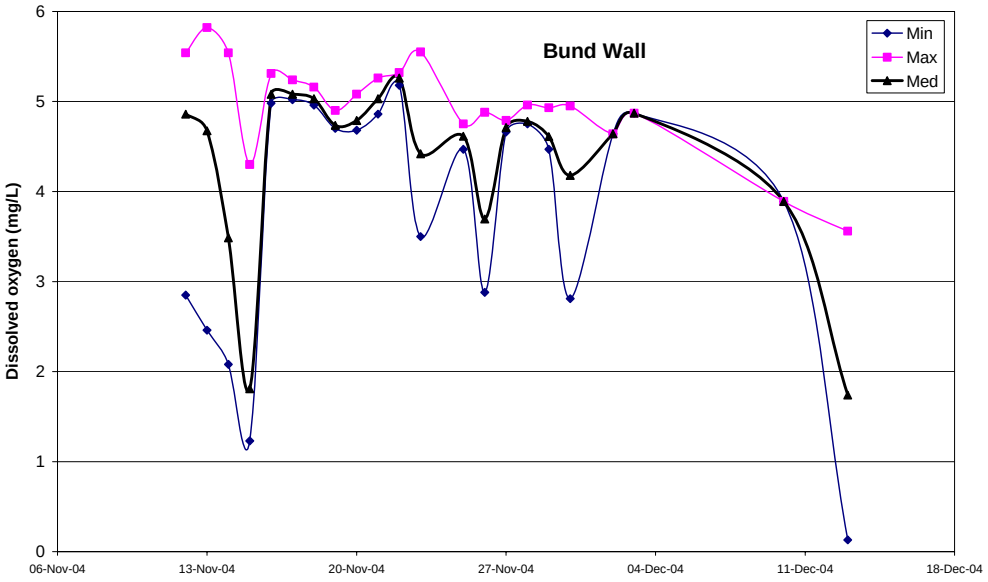

Figure 7

It is not clear from these results what the main mechanism for oxygen reduction is as there are many factors that contribute to this process. This includes the direct effect of herbicide on oxygen levels, the decomposition of plant material, the reduction in flow in Darwin River as the release of water from Darwin River Dam was stopped prior to the application of herbicide, and seasonal reductions in

oxygen levels associated with increasing water temperatures. With respect to the latter, it should be noted that oxygen levels at a control site well upstream of where herbicide was applied had measured oxygen levels of 2-3 mg/L, lower than what was measured at Site 1 immediately before the application of herbicide. This low level is not unusual in Top End waterways and is attributed to low flow and high water temperatures.

As a general rule fish are able to tolerate low DO levels down to 1 mg/L. However, below this value they exhibit signs of stress. In the case of Site 1 at both locations DO levels in excess of 1 mg/L were present in the water body. Throughout the time the river was treated with 2,4-D observations on the river showed there were no fish kills or loss of other animals. Other sections in this report deal with impact on flora and fauna more closely.

- Darwin River Monitoring - Post Treatment:

Post treatment analysis of Darwin River water was undertaken to ensure that levels of herbicide and breakdown products were below the recommended health guideline values. Landholders in the quarantine area were not permitted to reconnect pumps and extract water for domestic use until river water was declared usable.

To assist in determining suitable water quality in Darwin River a protocol was developed and endorsed by the Chief Health Officer, Department of Health and Community Services. In summary, the Australian Drinking Water Guideline (1996) (ADWG) state the maximum permitted levels to be 0.03 mg/L for 2,4-D and 0.2 mg/L for 2,4-dichlorophenol. There are no guideline values for 4-chlorophenol. In addition to these compounds water samples were also collected for the analysis of *E. coli*, a bacteriological indicator of water quality. ADWG state that potable water should contain no *E. coli*.

The protocol required that water samples were collected in duplicate at the herbicide application sites, and collection was to occur on two consecutive occasions at least 1 day apart. In addition, landholders were not allowed to reconnect to the river unless all analyses showed acceptable levels of 2,4-D, 2,4-dichlorophenol and *E.coli* both upstream and downstream of each pump site.

To satisfy this condition water samples were collected at sites and occasions. summarised in Table 10. Note that the collection of water samples commenced one week after the removal of the bund wall to allow a period of flushing of the river.

On every sample occasion the concentrations of 2,4-D, 2,4-dichlorophenol and 4-chlorophenol were below the level of detection of 1, 0.1 and 0.1 ug/L respectively. The exception was at Site G when sampled on the first occasion on 14 December 2004 when 2,4-dichlorophenol was detected at a concentration of 0.11 ug/L. It was not detected on the repeat sampling on 17 December 2004. This concentration of compound is well within the Australian Drinking Water Guidelines (1996) value of 0.2 mg/L (200 ug/L).

The other important indicator of water quality was *E.coli*. It was detected on every occasion. The following table summarises *E.coli* counts. Note no samples (NS)

were collected from the three bottom locations at Site 1 which requires specialised equipment for sample collection which is not available.

Table 10 Summary of site location and sample dates. Refer to text for explanation of site codes.

Site	Sample Date 1	Sample Date 2
Control Site 1	14 December 2004	17 December 2004
Site 4 U/S	14 December 2004	17 December 2004
Site 4 D/S	14 December 2004	17 December 2004
Site 3 U/S	14 December 2004	17 December 2004
Site 3 D/S	14 December 2004	17 December 2004
Site G	14 December 2004	17 December 2004
Site G	5 January 2005	6 January 2005
Site 2 U/S	5 January 2005	6 January 2005
Site 2 D/S	5 January 2005	6 January 2005
Site F	5 January 2005	6 January 2005
Kinbachers Pump Site	5 January 2005	6 January 2005
Site C	5 January 2005	6 January 2005
Site 1 900m U/S RBR Top	5 January 2005	6 January 2005
Site 1 900m U/S RBR Bot	5 January 2005	6 January 2005
Site 1 400m U/S RBR Top	5 January 2005	6 January 2005
Site 1 400m U/S RBR Bot	5 January 2005	6 January 2005
Site 1 300m D/S RBR Top	5 January 2005	6 January 2005
Site 1 300m D/S RBR Bot	5 January 2005	6 January 2005
Kez Hall Pump Site	5 January 2005	6 January 2005

The following should be noted with respect to the table:

- U/S refers to the upper reach of the infestation sites and D/S refers to the lower end of the reach.
- Sampling of the river was undertaken in two stages: December 2004 and January 2005. This was due to viable Cabomba being found at Site 2 and herbicide re-applied during the December sample collection. There was insufficient time to allow the river to clear of herbicide and water samples re-analysed before the Christmas break when NMI closes.
- Site 1 is the main infestation site and is a very large water body. To ensure the entire water body was adequately sampled three locations along the waterhole were selected, and these were sampled at the top and the bottom (a total of 6 water samples on each occasion).

On every sample occasion the concentrations of 2,4-D, 2,4-dichlorophenol and 4-chlorophenol were below the level of detection of 1, 0.1 and 0.1 ug/L respectively. The exception was at Site G when sampled on the first occasion on 14 December 2004 when 2,4-dichlorophenol was detected at a concentration of 0.11 ug/L. It was

not detected on the repeat sampling on 17 December 2004. This concentration of compound is well within the Australian Drinking Water Guidelines (1996) value of 0.2 mg/L (200 ug/L).

The other important indicator of water quality was *E.coli* (Table 11). Note no samples (NS) were collected from the three bottom locations at Site 1 which requires specialised equipment for sample collection which is not available.

Table 11 *E.coli* counts (per 100 mL) in Darwin River post-treatment. Sample dates coincide with dates in Table 10.

Site	Sample Date 1	Sample Date 2
Control Site 1	4	4
Site 4 U/S	740	150
Site 4 D/S	130	198
Site 3 U/S	40	86
Site 3 D/S	60	70
Site G	20	24
Site G	130	50
Site 2 U/S	180	80
Site 2 D/S	100	80
Site F	190	60
Kinbachers Pump Site	200	70
Site C	150	60
Site 1 900m U/S RBR Top	110	70
Site 1 900m U/S RBR Bot	NS	NS
Site 1 400m U/S RBR Top	80	60
Site 1 400m U/S RBR Bot	NS	NS
Site 1 300m D/S RBR Top	90	90
Site 1 300m D/S RBR Bot	NS	NS
Kez Hall Pump Site	40	60

This table shows that *E.coli* was present in the water on all occasions. This is not surprising as the river is an untreated water source, and as such is no different to any other untreated waterway.

The post treatment water quality survey provided the information needed to determine which landholders could reconnect to Darwin River and resume water extraction for extraction purposes. This occurred in two stages due to the re-application of herbicide at Site 2 mid-way through this sampling process. Before Christmas 2004 5 landholders above Site 3 were allowed to reconnect pumps to the river. The remaining 7 were allowed to reconnect in January 2005. On all occasions the water was declared safe of 2,4-D and the breakdown product 2,4-dichlorophenol. However, a warning was issued for residents to treat the drinking water due to the presence of *E. coli* being detected.

- Darwin River Estuary:

The purpose of the bund wall constructed across Darwin River at Cox Peninsula Road was to prevent Darwin River water containing 2,4-D flowing to Darwin Harbour. By mid-December the bund wall had neared capacity and was continuing to fill with the advent of the wet season. The decision was made to remove the

bund wall before the onset of monsoonal rains destabilised the wall. This required the controlled release of Darwin River water containing 2,4-D to Darwin Harbour.

A monitoring program was developed to determine the effects of this discharge on the estuary. This was formalised through Waste Discharge Licence 119 issued by the Office of Environment and Heritage, DIPE. The license specifies water release conditions and a monitoring program for the estuary for the compliance report associated with WDL 119. This report provides details of the monitoring program and all chemical and physical results. A brief summary of the monitoring program is presented below.

The monitoring program required estuarine waters to be analysed for 2,4-D, breakdown products and other in-situ water quality measures on one occasion before the release of bund water, and analyses repeated on four occasions after the release of bund water. Sites in the estuary were located at: Darwin River, Blackmore River, the confluence of these rivers, Adam Body Prawn Farm and Phelps Prawn Farm in Middle Arm. The initial concentration of 2,4-D in the bund water averaged 1,000 ug/L.

Figure 8 summarises concentrations of 2,4-D measured at the five sites. The main finding is that concentrations of 2,4-D rapidly decreased after release of bund water to levels below those recommended in the ANZECC Water Quality Guidelines of 140 ug/L (the most stringent level). The breakdown product 4-chlorophenol was not detected in the estuary, and traces of 2,4-dichlorophenol were found. This shows that there was virtually no degradation of 2,4-D.

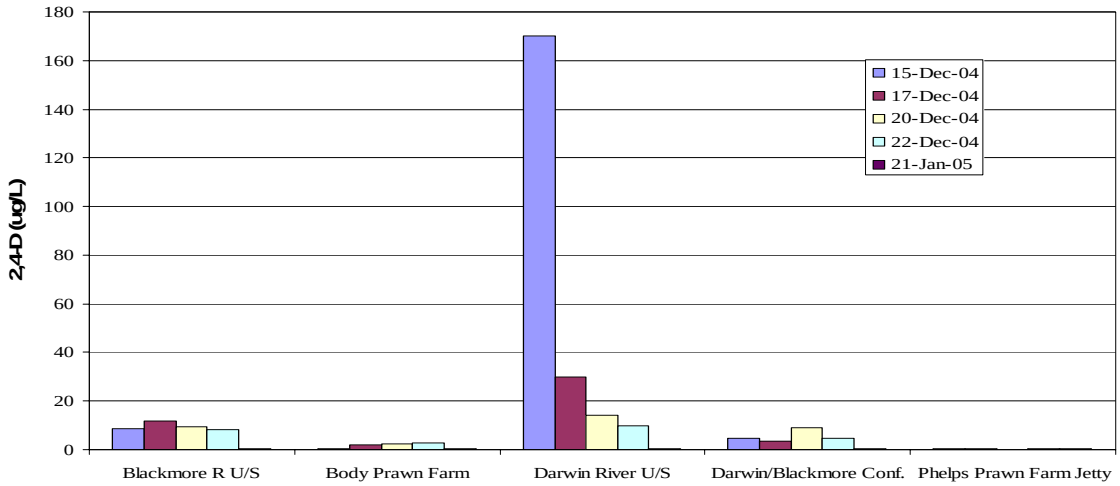

Figure 8. Concentrations of 2,4-D at the five estuary monitoring sites on five occasions after the release of bund water.

During the monitoring program there was no evidence of fish kills or detrimental effects on fauna, and there was little or no reduction on dissolved oxygen levels in the estuary.

In addition to these sites water samples were collected from each prawn farm operating in the area on two occasions (22 December 2004 and 2/4 February 2005). Two water samples from each farm were collected by government officers under the direction of farm staff. Analyses were performed by NMI in Sydney. All

results were below the levels of detection for 2,4-D and both breakdown products. This shows that these products did not enter these farms with intake water.

- Conclusions of Water Quality Monitoring

The two main issues of the effect of 2,4-D on water quality relate to the degradation of the herbicide to the two breakdown products 4-dichlorophenol and 2,4-dichlorophenol, and to the effect of 2,4-D on dissolved oxygen levels.

Contrary to what was reported in the literature and from experience with herbicide application in Queensland, 2,4-D did not appear to breakdown in Darwin River or in the estuary. The expectation was that herbicide breakdown would occur within a matter of weeks. The bund wall was built with the intention of containing treated Darwin River water for sufficient time so that natural breakdown reduced concentrations to below 140 ug/L, the most stringent ANZECC Water Quality Guideline value. At this point treated water could be released to Darwin Harbour. Reductions in concentrations of 2,4-D were associated with inflows of water, and mixing of treated waters with untreated waters, that is, dilution.

It is not clear why 2,4-D did not breakdown in Darwin River. The experience in Queensland is that the herbicide decomposes in the order of a week. It may be that the bacteria which are thought to facilitate this process are not present in sufficient numbers to effectively reduce 2,4-D concentrations.

Levels of dissolved oxygen were affected by the application of 2,4-D. Oxygen levels declined in the main infestation site (Site 1, both locations) and at the bund wall. At both these main sites the effect on oxygen levels was almost immediate. It is unclear what the mode of action is. These include: decomposing Cabomba resulting in an increased oxygen demand in the water; or 2,4-D affecting photosynthetic plants (eg plankton) which caused a reduction in oxygen production. Compounding these factors is the reduction in river flow associated with the application of herbicide, and the seasonal rise in water temperatures which results in natural reductions in Dissolved oxygen levels. The use of aerators to limit declines on DO was successful, especially in the smaller water body at the bund wall.

7. CONCLUSIONS

1. The multiple applications of AF Rubbervine Spray™ achieved an estimated knockdown of 99.99% of the living cabomba in Darwin River. Since achieving this knockdown cabomba has re-established. It is not yet known whether this re-establishment is a result of re-growth from root stock or germination of seeds.

2. The effect of AF Rubbervine Spray™ on non-target organisms during this program appears to be minimal. Monitoring of water dependant birdlife showed no significant detrimental impacts of AFRS. Fish surveys show total numbers of fish and number of species at the main infestation site (and the main 2,4 – D application site) to be similar to 2 out of the 4 control sites. There were also no sightings of dead fish along the river and in the main infestation site.

3. Concentrations of 2,4 – D at the main infestation site was sustained at 1,000 – 2,000 ug/l for a period of just over a month. This level is far in excess of 140 ug/l adopted by the Taskforce as an environmentally safe level. The level of 140 ug/l was the most stringent set by the Australian Water Quality Guidelines.

4. Concentrations of 2,4 – D in Darwin River did not decay as expected. Reductions in concentrations were associated with dilution rather than decay. This was due to lack of suitable micro-organisms which facilitate the decay process.

5. The main water quality impact in Darwin River was marked reductions in levels of dissolved oxygen. How much of this was attributed to 2,4 – D directly, or to reductions in flows and the bund wall is unknown. The use of aerators at two sites appeared to be effective in maintaining oxygen levels in the water at more immediate locations.

6: Monitoring of groundwater showed no presence of 2,4 – D or its breakdown products.

7. Monitoring of 2,4 – D in Darwin River estuary after the release of bund water showed rapid dilution of herbicide, and little or no effect on water quality. There were no observed fish deaths during water quality monitoring surveys, suggesting that fish life in the estuary had similar tolerances to the herbicide as freshwater species.

8: It cost the NTG $90,000 for delivery of potable water to landholders, most of which went to rambutan farm for irrigation purposes.

9. The production of viable seed has been confirmed at the Pine Creek site however, to date, this has not been confirmed at Darwin River although seed production is occurring. Currently it is too early to make a conclusion in relation to this issue however a number of considerations will need to be made if this occurs, these include: length of time until seed production occurs from germination, viability of seed produced and germinability of seed produced.

REFERENCES

Australian Drinking Water Guidelines (1996)

ANZECC/ARMCANZ (2000) Guidelines

Australian Water Quality Guidelines (1996)