



WARRELL CREEK TO URUNGA:

GIANT BARRED FROG MANAGEMENT STRATEGY

DECEMBER 2014



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1.0 Introduction

1.1 Background to the Project

The Pacific Highway Upgrade Program is a joint commitment by the Australian and New South Wales (NSW) governments to improve the standard and safety of the Pacific Highway between Hexham and the Queensland border. The Pacific Highway Upgrade Program includes the upgrade of the Pacific Highway between Warrell Creek and Urunga (WC2U) comprised of approximately 42 kilometres of dual carriageway road that would bypass the towns of Warrell Creek, Macksville, Nambucca Heads and Urunga on the Mid North Coast of NSW. The WC2U Project has been divided into two construction stages and includes the following:

- Stage 1 consisting of the northern 22.5 kilometres of the Project between Nambucca Heads and Urunga (NH2U).
- Stage 2 consisting of the southern 19.5 kilometres of the Project between Warrell Creek and Nambucca Heads (WC2NH).

The Environmental Assessment (EA) prepared for the Warrell Creek to Urunga Pacific Highway Upgrade project identified potential habitat for the Giant Barred Frog at several creeks and drainage lines in the northern half of the study area, through Nambucca, Little Newry and Newry State Forests (SKM 2010). The EA identified the proposal as having the potential to impact on this species as it would directly traverse streams and rivers across the study area. This was investigated in further detail with the Roads and Maritime Services (RMS) engaging Lewis Ecological Surveys to perform field surveys of the project route and based on these results and any updated information from desktop surveys formulate a Giant Barred Frog management strategy for the Upgrade.

1.2 Order of Precedence

In the event of any inconsistency, ambiguity or discrepancy between this Management Plan and the Flora and Fauna Management Plan for the Warrell Creek to Nambucca Heads Pacific Highway upgrade project, the following order of precedence must apply:

1. This Giant Barred Frog Management Strategy; followed by
2. The Flora and Fauna Management Plan for the Warrell Creek to Nambucca Heads Pacific Highway upgrade project (AFJV 2014).

1.3 Objectives of the Management Strategy

There are two key objectives of this management strategy, firstly, to demonstrate through the life of the Project that mitigation has maintained or improved the population size and habitat of the Giant Barred Frog at all sites where a population is present, and secondly, in the event significant negative changes are detected in the habitat of the Giant Barred Frog the Department of Environment (DoE) and Environmental Protection Agency (EPA) will be advised and agreed adaptive actions taken will be undertaken within three months to reverse the negative trend.

1.4 Subject Species – Giant Barred Frog (*Mixophyes iteratus*)

1.4.1 Description

The Giant Barred Frog (*Mixophyes iteratus*) is a large, dark-olive green to black coloured frog that grows to 115 mm. It has a pointed snout and a broad lateral band of dark spots dividing the dark dorsal surface from the white or pale yellow, ventral surface (underside). The limbs have dark crossbars. The hind side of the thighs are black with large yellow spots. Two joints of the fourth toe are free of web (Cogger 2000). The skin is finely granular above but smooth below. The call of the male Giant Barred Frog is a deep guttural grunt (OEH 2014).

Giant Barred Frog tadpoles are large and grow to over 100 mm in length. They are deep-bodied and ovoid, with a tail length twice that of the body. The tadpole's eyes are dorsolateral. The tadpoles are coloured yellow-brown above with dark spots and a dark patch at the base of tail. The underside is silver-white. The intestinal mass is obscured but the heart and lungs are visible from below (except near metamorphosis). The tail is thick and muscular (Anstis 2002). Fins are low and opaque with dark flecking (except the anterior half of the ventral fin; Meyer *et al.* 2001).

1.4.2 Distribution

The Giant Barred Frog is currently known from mid to low altitudes below 610 m above sea level (Hines *et al.* 2004), along the Coast and ranges from south-eastern Queensland to the Hawkesbury River in NSW. North-eastern NSW, particularly the Coffs Harbour-Dorrigo area, is now a stronghold (Figure 1-1). Considered to have disappeared south of the Hawkesbury and there are no recent records from the Blue Mountains (Hines and SEQTFRT 2002). Between Port Macquarie and Urunga the species appears to be patchily distributed with some confirmed recent locations from upper Warrell Creek and in smaller fast flowing streams in Way Way State Forest (Lewis 2014; Figure 1-1).



Plate 1-1. Giant Barred Frog (*Mixophyes iteratus*).

1.4.3 Habitat and Ecology

The Giant Barred Frogs forage and live amongst deep, damp leaf litter in rainforests, moist eucalypt forest and nearby dry eucalypt forest, at elevations below 1000 m. Whilst it has been observed to prefer a closed forest canopy with a relatively light cover of vegetation at ground level (Aland and Wood 2013), they have been found in cleared or disturbed areas, for example cattle farms with vegetated riparian strips and regenerated logged areas (Ingram and McDonald 1993; Hero and Shoo undated and cited in Hines *et al.* 2004; Lemckert and Brassil 2000; Lewis and Rohweder 2005). Many sites where the Giant Barred Frog is known to occur are the lower reaches of streams which have been affected by major disturbances such as clearing, timber harvesting and urban development in their headwaters (Hines *et al.* 1999).

Giant Barred Frogs breed around shallow, flowing rocky streams as well as deeper slower moving rivers from late spring to summer. Females lay eggs onto moist creek banks or rocks above water level, from where tadpoles drop into the water when hatched. Tadpoles grow to a length of 80–100 mm and take up to 14 months before changing into frogs. When not breeding, the frogs remain within 50 m of the stream edge (Streatfield 1999). They feed primarily on large insects and spiders.

1.4.4 Conservation Status

In NSW, the Giant Barred Frog is currently listed as Endangered pursuant to the NSW *Threatened Species Conservation Act* (1995) and Commonwealth *Environmental Protection and Biodiversity Conservation Act* (1999).

1.5 Initial Targeted Field Survey Program

1.5.1 Warrell Creek to Nambucca Heads

Initial targeted field surveys were performed between November 2011 and January /February 2013. During this time, all of the freshwater creeks considered as either semi-permanent or permanent in nature were surveyed regardless of the extent of riparian vegetation. Some neighbouring dams close to streams were also surveyed in light of recent findings on the Sapphire to Woolgoolga Project. The Nambucca River and Newee Creek were not considered suitable frog habitat due to their saline nature and were omitted from the field sampling program. Despite Butchers Creek initially being identified as containing potential habitat during the Environmental Assessment and during the first round of field surveys in 2011/12, further field surveys later in 2013 rendered there was little likelihood of Butchers Creek supporting populations of Giant Barred Frog. A reference site in Way Way State Forest (E:494538 N:6596076) was used to demonstrate conditions were suitable for the detection of Giant Barred Frogs throughout the sampling period (i.e. 2011-2013).

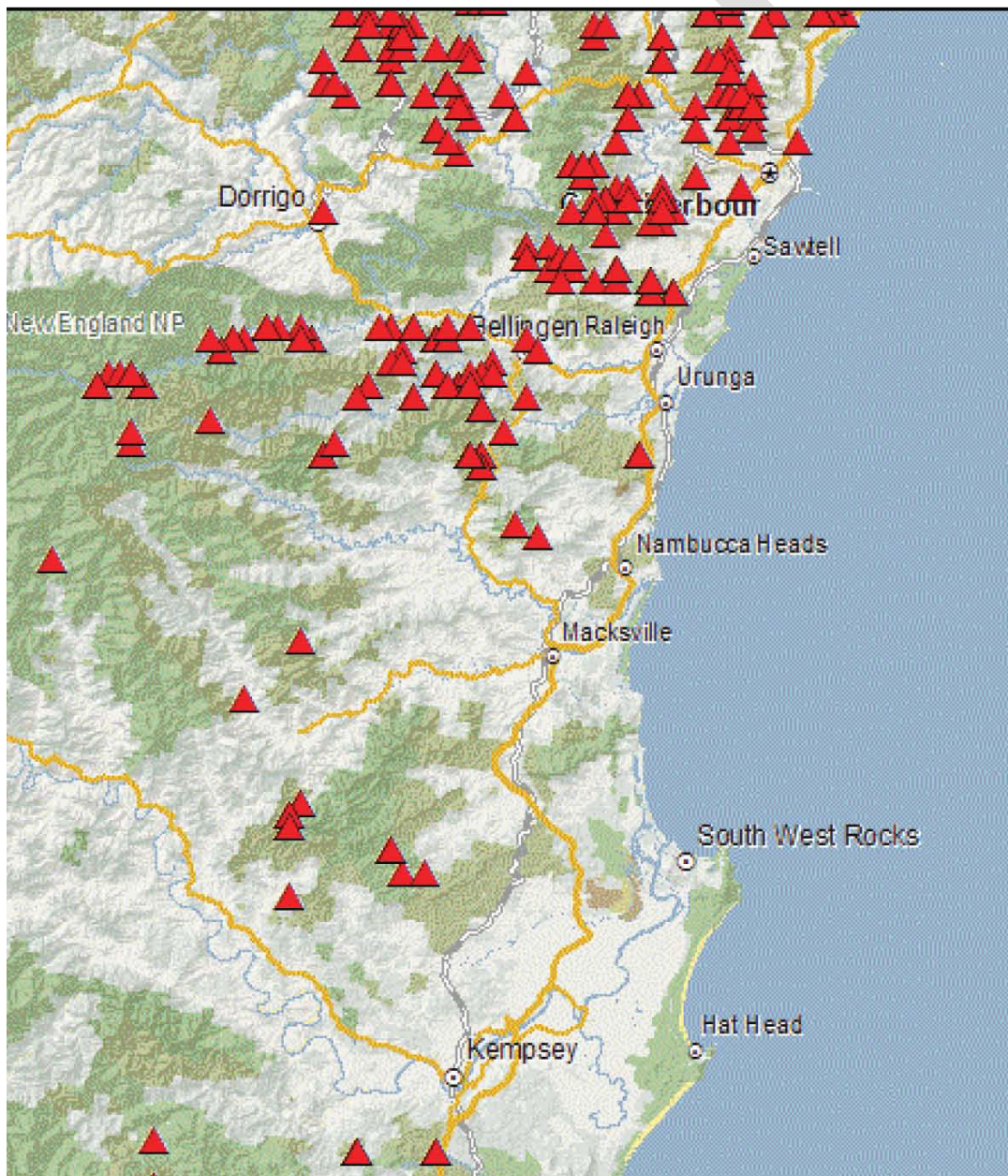


Figure 1-1. Localised distribution of Giant Barred Frog between Kempsey and Urunga.

Note - Triangles represent approximate location as sensitive 2 species.

Surveys were undertaken when the survey site received >10 mm of rainfall in 24 hours with field surveys performed within 7 days of this event. Surveys during heavy rainfall events/flooding were abandoned or repeated again at a later date when flooding had subsided. All sites were surveyed on at least two occasions during suitable conditions with additional surveys performed at sites which had an increased likelihood of supporting Giant Barred Frog. For example, Butchers Creek (ch. 43300) was surveyed during the summer of 2011 and 2012 and on three separate occasions during spring 2013.

1.5.2 Nambucca Heads to Urunga

Initial targeted Field surveys were performed in the same manner as described in Section 1.3.1 for the Nambucca Heads to Urunga section of the Upgrade between November 2011 and 2013. Despite some areas initially being identified as containing potential habitat during the Environmental Assessment and during the first round of field surveys in 2011/12 some subsequent field surveys later in 2013 rendered there was little likelihood of these areas supporting populations of Giant Barred Frog. They included Boggy Creek and McGraths Creek which had been subject to an additional three repeated surveys performed in December-February of 2012/13. The Kalang River and Bellingen River and Deep Creek were not considered suitable frog habitat due to their saline nature and were omitted from the field sampling program.

2.0 Results of the Initial Targeted Surveys

2.1 Warrell Creek to Nambucca Heads

Initial targeted field surveys resulted in the detection of only one Giant Barred Frog at Upper Warrell Creek (ch. 42565). At this location, one adult female (approximate snout-vent 120 mm) was recorded on the edge of the RMS project boundary (Figure 1-2). The individual was approximately 10 m from the water's edge and completely exposed above the leaf litter close to overhanging vegetation (i.e. *Lomandra longifolia*). Giant Barred Frogs were always recorded at the Way Way State Forest reference site and thus demonstrating that conditions were suitable to enable the detection of this species.

Following detection of the Giant Barred Frog at Upper Warrell Creek pre-construction baseline monitoring was undertaken to describe the population and existing habitat condition in more detail. The pre-construction baseline monitoring report is available in Appendix B.

2.2 Nambucca Heads to Urunga

No Giant Barred Frog populations were recorded in this section of the Upgrade. Despite some areas initially being identified as containing potential habitat during the Environmental Assessment and during the first round of field surveys in 2011/12, subsequent field surveys later in 2012/2013 (December-February) rendered there was little likelihood of these areas supporting populations of Giant Barred Frog.

2.3 Discussion of Initial Targeted Surveys

Field surveys confirmed the presence of Giant Barred Frogs in the Warrell Creek to Urunga study area with a population being identified at the southern limit of the Upgrade (Upper Warrell Creek ch. 42565). Despite some areas being initially identified as containing potential habitat in the Environmental Assessment, the surveys performed by Lewis Ecological Surveys found either no evidence to support this earlier assertion or employed a field survey program that was considered rigorous enough to confirm the presence or absence of Giant Barred Frog. For example, Butchers Creek (ch. 43330), Boggy Creek (ch. 62765) and McGraths Creek (ch. 71965) were surveyed on two occasions in 2011/12 and repeated again with three additional surveys between September and November 2013 for Butchers Creek and between December and February of 2012/13 for Boggy Creek and McGraths Creek. Normally three surveys would be sufficient to obtain a confidence interval at or above 95%. For example, surveys performed in the Bungawalbin Catchment consistently yielded Giant Barred Frog on the first and second occasion whilst the third visit to a site rarely yielded additional new locations for frogs (Lewis and Rohweder 2005). The absence of frogs following five surveys at those sites believed to provide some potential habitat for Giant Barred Frogs would indicate with a high degree of probability that they do not exist at these locations, a fact also supported with the apparent absence of historic records for the immediate area (see Figure 1-1).

A reference site was used throughout the field sampling as a means to demonstrate that conditions were suitable for the detection of Giant Barred Frogs. Whilst locating such a site proved initially problematic, largely due to the fact that no sites could be located close to the project the Way Way site was a useful indicator to demonstrate the prevailing abiotic conditions were always suitable when field sampling was undertaken. This site was, however, different from many of the sites being sampled along the project route because it was a rocky fast flowing stream within a continuous tract of forest unaffected by agriculture.

2.4 Areas Subject to Giant Barred Frog Management

A Giant Barred Frog population was only recorded at Upper Warrell Creek (ch. 42565) with this area identified for management. No other areas within the Upgrade Project have been proposed for Giant Barred Frog management; however, provisions are available within this management strategy to allow for unexpected finds procedures and the actions therein.

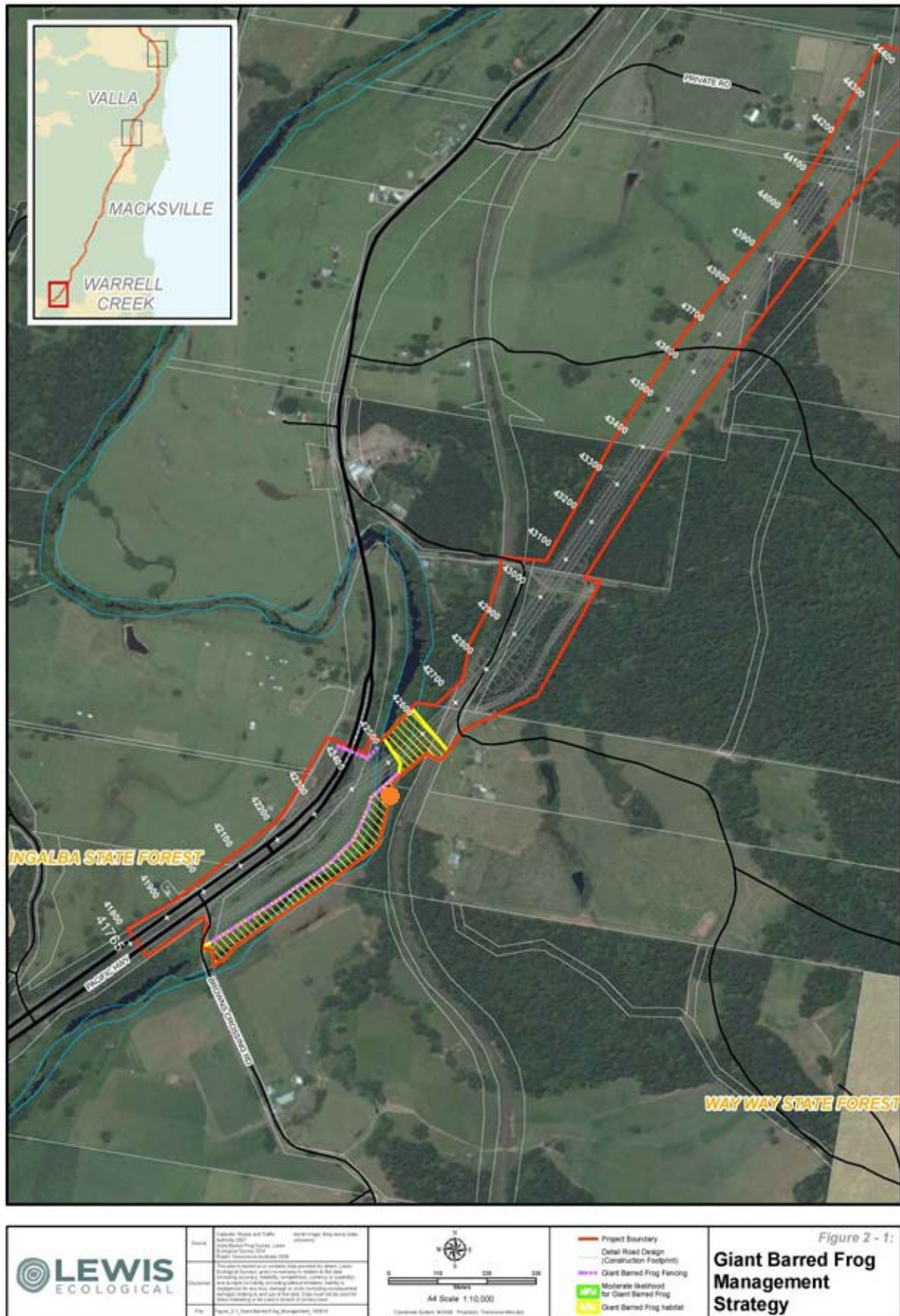


Figure 1-2. Location of the Giant Barred Frog records (orange circle) during initial field surveys of the Upper Warrell Creek site.

3.0 Pre-construction Management Measures

3.1 Overview of Activities

Pre-construction activities would involve the following works:

- Field survey;
- Water quality monitoring;
- Translocation of threatened plants;
- Geotechnical investigations;
- Completion of utility relocations; and
- Construction of sites accesses.

3.2 Timing

Pre-construction works are to be undertaken up until the commencement of construction stage works which are scheduled to commence in January 2015.

3.3 Summary of Potential Impacts

Pre-construction activities may have the following potential impacts to the Giant Barred Frog:

- Mortality to Giant Barred Frog from pre-construction activities; and
- Spread of Chytrid fungus.

3.4 Main Goals for Management

There are four main goals for the management of Giant Barred Frogs including:

- No loss of known Giant Barred Frog habitat from pre-construction activities;
- No injury/mortality to Giant Barred Frog from pre-construction activities;
- Minimise the spread of Chytrid fungus during pre-construction activities; and
- Ensure that appropriate habitat offsets have been identified for Giant Barred Frog conservation.

3.5 Mitigation Measures

3.5.1 Detailed Design Considerations

As detailed design progresses, a number of factors will be addressed to minimise potential impacts on the Giant Barred Frog. These include:

- Avoiding and minimising vegetation removal where feasible and reasonable;
- Protection of existing known habitat (*see* Section 3.5.2);
- Review and enhance where relevant the proposed temporary frog fencing to reduce the likelihood of road kills;
- Review and enhance the landscape and rehabilitation plan as well other temporary seeding schedules to maintain or enhance habitat connectivity.

3.5.2 Protection of Existing Habitat

Upper Warrell Creek at ch. 42565 should be protected from pre construction and construction related works other than what is considered essential. The locating of access tracks, utilities redistribution, car parking facilities and other ancillary works including topsoil stock piles, lay down areas, wash down bays, site shedding and compound sites should not be located in this area. This approach will be in accordance with MCoA:

C1. The Proponent shall employ all feasible and reasonable measures to minimise the clearing of native vegetation to

the greatest extent practicable during the construction of the project

C27 Unless otherwise approved by the Director General in accordance with this condition, the sites for ancillary facilities associated with the construction of the project shall (c) be located in areas of low ecological significance and require minimal clearing of native vegetation (not beyond that already required by the project).

The protection of the identified areas should include the demarcation of clearing limits and signage identifying these areas as 'no go' zones.

3.5.3 Controls on Habitat Clearing (Pre-construction)

During the pre-construction stage of the Project (prior to approval of the CEMP) only clearing defined as 'minor' (see Approval Instrument Definitions for "construction") can be undertaken, unless approval is sought from the Director-General. Prior to any clearing taking place, a suitably qualified Project Ecologist will undertake an inspection of vegetation to be cleared to determine that only 'minor clearing' is to be undertaken. Minor clearing will be defined as the following:

- Vegetation that does not include mature trees >150 mm diameter at breast height (DBH);
- Vegetation that does not comprise known threatened fauna habitat. In the case of the Giant Barred Frog, this is defined as Upper Warrell Creek at ch. 42565 and neighbouring riparian vegetation for distances of up to 75 m (Figure 2-1).
- Areas of vegetation that have ecological constraints (e.g. threatened flora habitat/ areas of endangered ecological communities).

3.5.4 Pre-construction Baseline Monitoring (Upper Warrell Ck)

Pre-construction baseline monitoring has been undertaken to obtain data on the local Giant Barred Frog population at Upper Warrell Ck (ch. 42565). These surveys were undertaken in Spring 2013, Summer 2014 and Autumn 2014 with each survey occurring within 7 days of a suitable rainfall event defined here as >10 mm in 24 hours and the ambient air temperature was >18°C at the commencement of the survey and not lower than 14.4 °C during the survey. The Bureau of Meteorology (BOM) weather stations at Macksville Country Club (059018) and Fisherman's Reach (059143) were used as reference points to identify a rainfall event and provide a guide for prevailing ambient air temperatures. No surveys were performed during heavy rainfall events/flooding. This sampling rationale has considered other studies which have examined the environmental conditions on the activity of Giant Barred Frogs (e.g. Koch and Hero 2007). Further details are provided in Appendix B of this management strategy.

3.6 Performance Measures and Corrective Actions

The performance measures and corrective actions for the pre construction management of Giant Barred Frogs is summarised in Table 3-1. This table also describes how the identified mitigation measures are to be monitored, the timing and frequency of monitoring, who is responsible for implementing the measures, the performance thresholds that each goal is measured against and the corrective actions if the performance thresholds are triggered.

Table 3-1. Pre-construction management goals, mitigation measures and their timing, performance thresholds and corrective actions during the pre-construction works.

Management Goal	Mitigation/ Control Measure	Monitoring / Timing Frequency	Responsibility	Performance Threshold	Corrective Actions if Deviation from performance Criteria
No loss of known Giant Barred Frog habitat from pre-construction activities	No areas of Giant Barred Frog habitat to be cleared during preconstruction	Constraints maps to include Giant Barred Frog habitat mapping	AFJV (Design team)/ suitably qualified Project Ecologist	No Giant Barred Frog habitat to be cleared during preconstruction	Consideration of additional offsets for habitat loss
	All ancillary sites to be located outside of mapped Giant Barred Frog habitat.	Ecological assessments to be prepared for ancillary sites to verify minimal impacts to Giant Barred Frog habitat	AFJV (Environmental team)/ suitably qualified Project Ecologist	No areas of mapped Giant Barred Frog habitat to be impacted by the ancillary facilities	Consideration of additional offsets for habitat loss
No injury/mortality to Giant Barred Frog from pre-construction activities	No areas of Giant Barred Frog habitat to be cleared during preconstruction	Constraints maps to include Giant Barred Frog habitat mapping	AFJV (Design team)/ suitably qualified Project Ecologist	No Giant Barred Frog habitat to be cleared during preconstruction	Consideration of additional offsets for habitat loss
	All ancillary sites to be located outside of mapped Giant Barred Frog habitat.	Ecological assessments to be prepared for ancillary sites to verify minimal impacts to Giant Barred Frog habitat	AFJV (Environmental team)/ suitably qualified Project Ecologist	No areas of mapped Giant Barred Frog habitat to be impacted by the ancillary facilities	Consideration of additional offsets for habitat loss
Minimise the spread of Chytrid fungus during pre-construction activities	No areas of Giant Barred Frog habitat to be accessed during preconstruction	Constraints maps to include Giant Barred Frog habitat mapping	AFJV (Design team)/ suitably qualified Project Ecologist	No Giant Barred Frog habitat to be cleared during preconstruction	Consideration of additional offsets for habitat loss
	All ancillary sites to be located outside of mapped Giant Barred Frog habitat.	Ecological assessments to be prepared for ancillary sites to verify minimal impacts to Giant Barred Frog habitat	AFJV (Environmental team)/ suitably qualified Project Ecologist	No areas of mapped Giant Barred Frog habitat to be impacted by the ancillary facilities	Consideration of additional offsets for habitat loss
Ensure that appropriate habitat offsets have been identified for Giant Barred Frog conservation	Perform field surveys at nominated biodiversity offset sites	Spring and Summer 2014	Roads and Maritime	Giant Barred Frog potential habitat identified in the nominated biodiversity offset sites	Located additional areas and survey for Giant Barred Frog

4.0 Construction Management Measures

4.1 Timing

Construction works are scheduled to commence in January 2015 and are expected to be completed in late 2017.

4.2 Summary of Potential Impacts

The construction stage works have the following potential impacts on Giant Barred Frog:

- Loss of known Giant Barred Frog habitat to accommodate the Project over Upper Warrell Creek;
- Injury/ mortality to individuals during the clearing and subsequent construction works; and
- Fragmentation of habitat.

4.3 Main Goals for Management

The main goals for Giant Barred Frog management during construction include:

- Minimise the loss of known Giant Barred Frog habitat during clearing and grubbing operations;
- Minimise road kill during construction activities;
- No injury/ mortality to Giant Barred Frog from construction activities;
- Undertake habitat rehabilitation works within identified areas of the Project Site to create or improve existing Giant Barred Frog habitat.

4.4 Mitigation Measures

4.4.1 Pre-clearing Surveys at Sites with Known Giant Barred Frog Habitat

Pre-clearing surveys will provide an additional safeguard to reduce direct mortality to individual frogs during the clearing and grubbing phase of the project. At known Giant Barred Frog sites (Upper Warrell Creek) the following pre-clearing survey procedure shall be undertaken.

4.4.2 Early Works – Establishing Site Controls (Temporary Frog Fencing)

- a) The works area for the temporary fencing is inspected/searched by the Project Ecologist immediately prior to installing the temporary fencing. The search should use active techniques such as raking the leaf litter, call broadcast (this species will readily call during the day) and inspections around tussocks (i.e. *Lomandra* clumps in particular) and logs.
- b) Temporary frog fencing installed for up to 200 m either side of the stream (minimum 900 mm high above ground and buried to a depth of 50-100 mm)¹. Where the terrestrial habitat bordering the stream is cleared land (i.e. Upper Warrell Creek ch. 700) this may be reduced to 100 m. In each instance a return wing (5 m in length) will be installed to reduce frogs breaching the fence.
- c) Fencing to be installed and inspected/signed off by the ecologist with Giant Barred Frog experience or a suitably qualified person who has successfully detected this species on at least 10 occasions at different sites. This procedure should form part of the pre clearing/ground disturbance checklist/permit.

¹ It is acknowledged that installation of the fence itself will represent ground/vegetation disturbance and as such it should be subject to a pre clearing active search survey and the works supervised by the Project Ecologist.

- d) Fencing will be installed at least 5 days prior to the scheduled clearing date so that active searches can be performed within the clearing footprint (see below).
- e) All this is to be in place at least 5 days prior to nominated clearing start date.
- f) Daily inspections of temporary frog exclusion fencing shall be undertaken following completion of pre-clearing survey (as below) up until the installation of the permanent Giant Barred Frog fencing

4.5.3 Pre-clearing Survey for Giant Barred Frogs

- a) Within 6 weeks of scheduled clearing/ground disturbance operations, the Project Ecologist will perform pre-clearing surveys over a minimum of two non-consecutive nights (i.e. before clearing commences). Surveys during heavy rainfall events/flooding are not supported and should be abandoned or repeated again at a later date when flooding has subsided. Pre-clearing surveys in known Giant Barred Frog habitat areas are not to take place during winter periods or other periods of likely dormancy including extended dry weather periods (i.e. more than 7 nights without a rainfall event of greater than 10 mm in 24 hrs).
- b) Surveys to last 1 person hour per hectare of habitat to be disturbed/removed and involve the use of call broadcast, spotlighting and active searches of litter, debris and logs. For any individuals that have their home range within the construction site they will be temporarily relocated during construction. Relocation points will be minimised as much as practical from collection point (see below).
- c) All Giant Barred Frogs captured will be relocated to the nearest side of the clearing limit with information collected on sex, breeding condition and snout-vent length. Alternative relocation sites may be considered provided they occur within the same drainage. As a general rule frogs should not be relocated further than 100 m from the capture site which should theoretically remain within an individual's home range.
- d) Frogs with a snout-vent length >40 mm will be PIT² tagged to document the performance measure of this as a suitable relocation strategy. Juvenile/sub adult frogs may be marked in accordance with the animal care and ethics licence of the Project Ecologist or frog expert. Toe-clipping is one possible method, however, not all animal care and ethics committees support this approach.
- e) A frog hygiene protocol will be adopted at sites with Giant Barred Frog (see Appendix D). This protocol will be in accordance with Department of Environment and Climate Change DECC (now OEH) Hygiene protocol for the control of disease in frogs Information Circular Number 6 (see DECC 2008).
- f) In the instance of flooding in the area and flood water breaches the exclusion fencing, the Project Ecologist with Giant Barred Frog experience or frog expert to be consulted regarding replacement of fencing.

4.5.4 Clearing Supervision in Giant Barred Frog areas

- a) At the Upper Warrell Creek site (ch. 42565) the clearing and grubbing activities will be supervised by the Project Ecologist until such a time they are confident no Giant Barred Frogs remain within the work site.
- b) Captured frogs will be treated as per 4.5.3 c) and 4.5.3 d).
- c) The need to perform additional night time surveys will be at the discretion of the Project Ecologist or frog expert. For example, only part of the site may have been cleared or more suitable weather conditions present an increased opportunity to detect frogs.

² Passive Integrated Transponder (i.e. microchip as used to mark and identify domestic animals).

4.5.5 Dewatering Procedures in Giant Barred Frog areas

- a) The dewatering process will be supervised by the Project Ecologist with the aforementioned Giant Barred Frog experience (see Section 4.4.2 c), in accordance with an Environmental Work Method Statement (EWMS) and the DECC (2008) hygiene protocol for the control of disease in frogs. All waterways and dams within those areas identified as Giant Barred Frog habitat will be subject to this dewatering process if dewatering is required. .
- b) Where the water body is to be pumped dry the intake pipe must be positioned in the deepest section.
- c) Screening of the pump intake (5mm mesh size) will be installed to prevent tadpole entrainment.
- d) Once the remaining water body is shallow enough to be effectively waded through by field personnel intensive dip netting will be undertaken to remove as many aquatic fauna as practical.
- e) All tadpoles that can be clearly identified to a genus other than *Mixophyes* do not need sorting. Tadpoles to be placed into holding containers. The size of these containers will be left to the discretion of the Project Ecologist.
- f) All tadpoles will be released into permanent/semi-permanent pools in adjacent habitats. Tadpoles will be first acclimatised to the recipient sites water temperature by immersing bags or aquaria in the release pools to allow a gradual equilibrium of water temperature prior to release.
- g) In stances where there are numerous tadpoles from a wide range of species, preferential treatment will be given to Giant Barred Frog tadpoles due to their legislative status as an endangered species. The release of predatory species (i.e. eels) will not occur in areas where Giant Barred Frog tadpoles are being released. This will reduce the risk of predation and/or competition.

4.5.6 Permanent Frog Fencing

- a) Frog fencing must be installed in areas where the presence of Giant Barred Frogs has been confirmed and there is a 'high' risk of frogs accessing the carriageway. A high risk has been defined as earth embankments/batters within 200 m of the stream.
- b) The fence must provide the required protection for between 100-200 m either side of the stream. Based on the concept design frog fencing may be required at Ch. 41965-42515 (either side of Upper Warrell Creek as shown in Figure 1-2).

Design wise, the frog fencing can be a standalone fence positioned between the floppy top fauna fence or boundary fence and the carriageway (i.e. toe of the batter). From a design perspective, the fence is a larger version of the design used at a number of Green-thighed Frog locations. It will stand at least 900 mm in height and comprise neoprene rubber sheeting including a small rubber return of not less 100 mm on the ground. The fence hot dip galvanized pressed sheet metal or powder coated aluminum pressed sheet mounted on a galvanized star picket (Figure 4-1). This design has been installed on the Kempsey Bypass Project and was supported by the EPA (Lewis 2011). An alternative option may be to retrofit a similar design described above to any proposed floppy top fauna fencing.

The success of this design will be based on the absence of Giant Barred Frog fence breaches³. As part of the monitoring procedures for measuring the effectiveness of the frog fencing, some monitoring of fence breaches must be undertaken by a suitable qualified zoologist at certain times of the year (i.e. when population monitoring occurs). This monitoring program will involve surveys for Giant Barred Frog on both sides of the frog fence as this data will clearly show whether the frog fence is effective at excluding frogs.

³ This will also be detailed in the EMS required for the project.

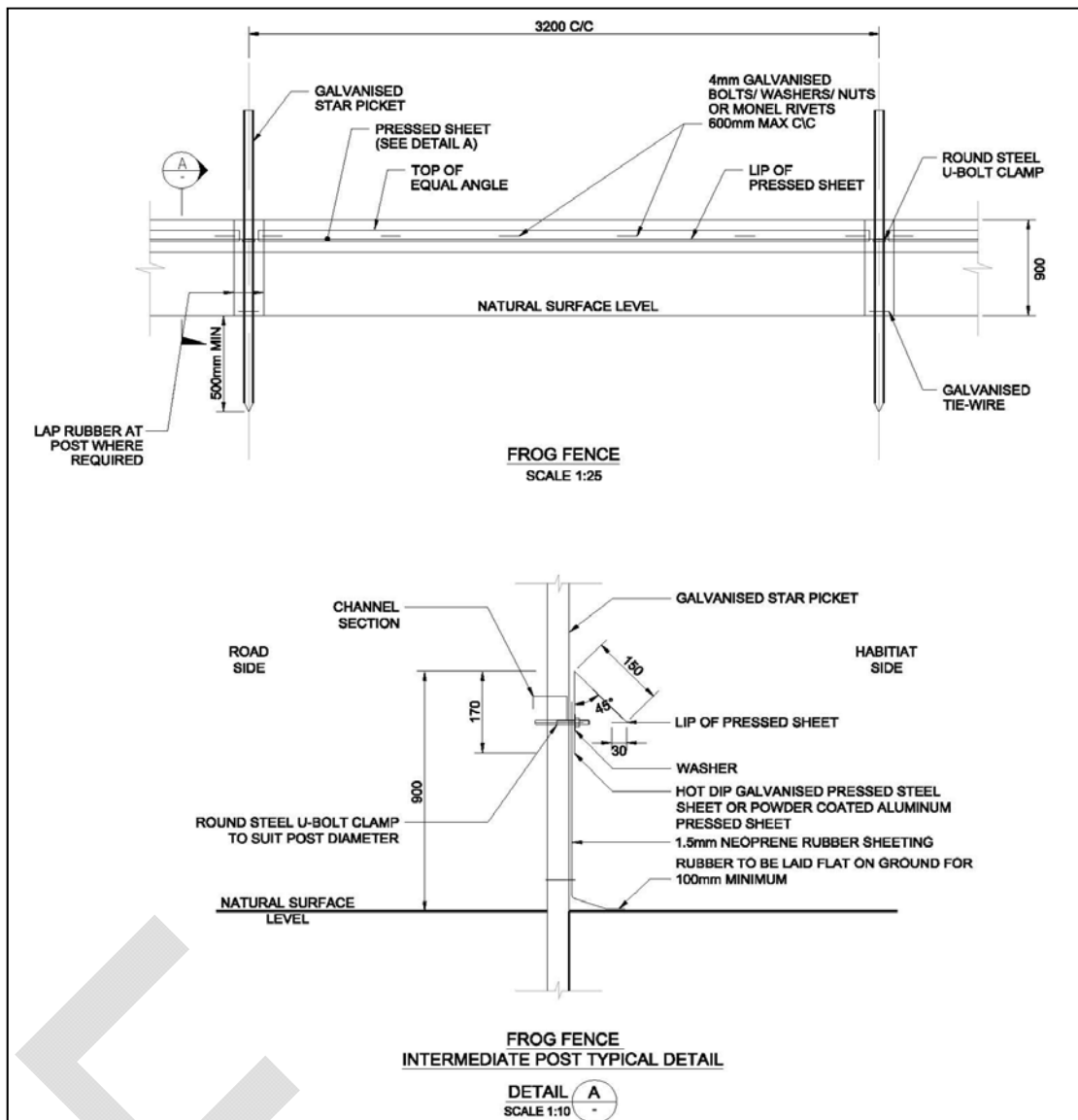


Figure 4-1. Example of a frog fence design for Warrell Creek to Urunga.

4.5.7 Unexpected Finds Process

An unexpected finds process has been developed to manage instances where Giant Barred Frog may be detected during pre-clearing surveys, clearing operations or dewatering works for the upgrade. This is in response to field surveys not being exhaustive (<3 surveys at any given site) and the ability of Giant Barred Frogs to move relatively large distances in short time periods. For example, many tens of metres when the clearing footprint will rarely extend beyond 100 m.

In an unexpected finds instance the management strategies outlined in this plan will be adopted and include:

1. Protection of Giant Barred Frog habitat including provisions for its protection from ancillary areas and their associated impacts consistent with MCoA C1 and C27;
2. Temporary and if required permanent frog fencing;
3. Additional pre-clearing surveys as deemed appropriate by the Project Ecologist or frog expert;
4. An examination and review of the adequacy of the proposed mitigation measures proposed at that site in consultation with the EPA, and
5. Implementation of the monitoring program in accordance with Section 7.0 and the performance measures outlined in this management strategy.

5.0 Construction Stage Monitoring

Monitoring during the construction phase of the Project will focus on the following:

- Ecological Assessments to be prepared for any additional areas to be cleared to verify minimal impacts to Giant Barred Frog habitat;
- Monitoring of temporary frog exclusion fencing and at a later stage in the project the permanent frog exclusion fencing once installed;
- Monitoring the stability of the Giant Barred Frog population and habitat condition in areas adjacent to the Project in the same manner as prescribed in the preconstruction baseline survey (see Appendix B);
- Road kill surveys performed daily during the clearing operations and weekly thereafter; and
- Monitoring and maintenance of plantings used in rehabilitated areas and monitoring on the extent of weeds (Table 5-1).

5.1 Performance Measures and Corrective Actions

The performance measures and corrective actions for the pre construction management of Giant Barred Frogs is summarised in Table 5-1. This table also describes how the identified mitigation measures are to be monitored, the timing and frequency of monitoring, who is responsible for implementing the measures, the performance thresholds that each goal is measured against and the corrective actions if the performance thresholds are triggered.

Table 5-1. Management goals, mitigation measures and their timing, performance thresholds and corrective actions during construction.

Management Goal	Mitigation/ Control Measure	Monitoring / Timing Frequency	Responsibility	Performance Threshold	Corrective Actions if Deviation from performance Criteria
Minimise habitat loss for the Giant Barred Frog from clearing.	Any design changes required during the construction stage would minimise clearing of Giant Barred Frog habitat where feasible and reasonable	Ecological Assessments to be prepared for additional areas to be cleared to verify minimal impacts to Giant Barred Frog habitat	AFJV (Environmental team, Design team)	Giant Barred Frog habitat to be cleared to not exceed approvals	Notification to DoE and EPA if the performance thresholds cannot be met
	The limits of clearing are to be clearly marked on all relevant work plans and protective fencing erected to mark these limits (i.e. 'no-go' areas)	Clearing limits to be checked prior to the commencement of clearing by survey and environmental team	AFJV (Environmental team, Survey team)	Final Sensitive Area Plans identify sensitive areas and 100% of clearing drawings identify clearing extents Clearing limit does not exceed approved limits (State and Commonwealth)	Additional habitat rehabilitation works to be undertaken on the Project to offset losses Consideration of additional offsets for habitat loss
No injury/ mortality to Giant Barred Frog from construction activities	Preparation of an EWMS would be undertaken for all construction activities to clearly communicate relevant measures within this plan to work crews Ongoing induction of all personnel involved with construction activities would be undertaken to advise of Giant Barred Frog management requirements Early Works – Establishing Site Controls (Temporary Frog Fencing) (4.4.2) Pre-clearing Survey for Giant Barred Frogs (4.4.3) Clearing Supervision in Giant Barred Frog areas (4.5.4) Dewatering Procedures in Giant Barred Frog areas (4.5.5) Permanent Frog Fencing (4.5.6) Unexpected Finds Procedure (4.5.7)	Pre-clearing permits/checklists to be completed by the Project Ecologist with Giant Barred Frog experience prior to the clearing of any vegetation Post-clearing inspections of recently cleared areas (<1 day) in known Giant Barred Frog habitat to identify any individuals injured or killed during clearing The detection of chytrid fungus 'sick and dying' frogs Dewatering permit/checklist to be completed by the Project Ecologist with Giant Barred Frog experience prior to any water bodies being dewatered in Giant Barred Frog habitat Daily inspections of temporary frog exclusion fencing following completion of pre-clearing survey until the installation of the permanent Giant Barred Frog fencing	AFJV (Environmental/ Construction team)/ suitably qualified Project Ecologist	No Giant Barred Frog injuries/ mortalities of adults or tadpoles as a consequence of construction activities.	Notification to DoE and EPA if Giant barred Frog mortality is recorded on the Project. Seek advice from DoE and EPA for current best practise for Chytrid fungus Reinstate site controls as relevant to this management strategy.

Management Goal	Mitigation/ Control Measure	Monitoring / Timing Frequency	Responsibility	Performance Threshold	Corrective Actions if Deviation from performance Criteria
To collect data to demonstrate that mitigation has maintained the population size and habitat of the Giant Barred Frog similar to results of the preconstruction baseline surveys	<p>Temporary frog exclusion fencing</p> <p>Maintenance of revegetation/ rehabilitation areas of Upper Warrell Creek</p> <p>All mitigation measures applied during construction as per Table 5-1</p>	Continuation of the pre construction field survey program on an annual basis and at appropriate times for sampling (i.e. >10 mm in past 7 days) in spring, summer and autumn in Years 1 & 3 (Construction stage of the Project)	Roads and Maritime / AFJV	<p>Giant Barred Frog recorded along the monitoring transect</p> <p>The detection of Chytrid fungus</p> <p>No breaches in fauna exclusion fencing.</p>	<p>Extend the monitoring transect by 500 m to determine presence of Giant Barred Frogs in adjacent areas</p> <p>Review/audit the performance of Weed and Pathogen Plan as (<i>see</i> Appendix D)</p> <p>Modify, if appropriate, design of existing measures where feasible and reasonable</p> <p>Advise DoE and EPA and discuss adaptive management actions including assisted plantings. Within two weeks of the change being identified with corrective action agreed by DoE, EPA and Roads and Maritime implemented within 3 months</p>
Minimise road kill of Giant Barred Frog during construction activities.	<p>Giant Barred Frog road kill to be reported to the Project Ecologist during daily/weekly monitoring</p> <p>An assessment of future road kill risks including adaptive management actions is to be provided by the Project Ecologist where:</p> <ul style="list-style-type: none"> A Giant Barred Frog is detected within/ near the site; or 	<p>Daily inspection of roads within likely Giant Barred Frog range (as assessed by Project Ecologist) during clearing operations</p> <p>Weekly inspection of roads within likely Giant Barred Frog range (as assessed by Project Ecologist) for duration of</p>	AFJV (Environmental team/ suitably qualified Project Ecologist)	No road kill of Giant Barred Frog resulting from the Project.	An assessment of future road kill risk will be undertaken by the Project Ecologist for areas where Giant Barred Frog road kill have been detected. This assessment will aim to provide actions to mitigate

Management Goal	Mitigation/ Control Measure	Monitoring / Timing Frequency	Responsibility	Performance Threshold	Corrective Actions if Deviation from performance Criteria
	<ul style="list-style-type: none"> Giant Barred Frog road kill is detected 	construction			<p>the risk of future Giant Barred Frog road kill in such areas</p> <p>Review the integrity of the fence, its design, its extent for either the temporary or permanent fencing.</p>
<p>Undertake habitat rehabilitation works within identified areas of the Project Site to create or improve existing Giant Barred Frog habitat</p>	<p>Progressive rehabilitation of identified areas (refer to Appendix C) Key rehabilitation measures will include planting of the northern bank of Upper Warrell Creek on either side of the bridge</p> <p>Progressive revegetation/ rehabilitation during construction</p> <p>Use of locally endemic native species representative of those currently growing along Upper Warrell Creek</p> <p>Monitoring and maintenance of plantings</p> <p>Managing and controlling weeds</p>	<p>Monitoring and maintenance of rehabilitation areas to be undertaken regularly as part of the Project landscaping contract.</p> <p>Weed monitoring would be undertaken on the site.</p>	<p>AFJV (Landscape Design/ Construction team)</p>	<p>Successful establishment of Giant Barred Frog habitat in the nominated areas</p>	<p>Consideration of additional landscaping/ habitat rehabilitation works.</p>

6.0 Operational Management Measures

6.1 Summary of Potential Impacts

The operational stage of the Project has the potential to have the following impacts on Giant Barred Frog:

- Fragmentation and loss of habitat; and
- Risk of vehicle strike associated with the upgrade.

6.2 Main Goals for Management

The main goals for management include:

- Maintain habitat connectivity for Giant Barred Frog as the population extends to both sides of the carriageway;
- Minimise vehicle strike of Giant Barred Frog during operational activities; and
- Maintain habitat rehabilitation areas.

6.3 Mitigation Measures

6.3.1 Habitat Offset Strategy

This Strategy would be prepared and implemented to offset the biodiversity impacts of the Project to address the *Environmental Protection and Biodiversity Conservation Act (1999)* offset requirements.

Table 6-1. Management goals, mitigation measures and their timing, performance thresholds and corrective actions during operational phase of the Project.

Main Goal	Mitigation/ Control Measure	Monitoring / Timing Frequency	Responsibility	Performance Threshold	Corrective Actions if Deviation from performance Criteria
Maintain habitat connectivity for Giant Barred Frog as the population extends to both sides of the carriageway	Permanent frog exclusion fencing Maintenance of revegetation/ rehabilitation areas of Upper Warrell Creek	Monitoring existing 1km transect at Upper Warrell Creek in spring, summer and autumn as per methods outlined in Appendix B for year 4, 6 and 8 Regular monitoring of the rehabilitation areas would be undertaken as part of the landscape maintenance works.	Roads and Maritime/ AFJV	Continued presence of Giant Barred Frog from any part of the 1km transect once Operational Monitoring commenced Presence of tadpoles, metamorphs or juvenile frogs during follow up surveys No greater than 30% change in foliage projection cover (fpc) for overstorey trees, shrubs and groundcover No greater than 30% reduction in litter cover No greater than 15% increase in soil cover No statistically significant differences (p<0.05 level) in declining water quality parameters	If no frogs are found, the search is repeated over an area extended by a further 500 m upstream and downstream and based on these results, the management actions and ongoing monitoring program for the Giant Barred Frog at Upper Warrell creek be redefined in consultation with the EPA and DoE Advise DoE and EPA and discuss adaptive management actions including assisted plantings. Within two weeks of the change being identified with corrective action agreed by DoE, EPA and Roads and Maritime implemented within 3 months
Minimise vehicle strike of Giant Barred Frog during operational activities	Permanent frog exclusion fencing	Initially during the monitoring existing 1km transect at Upper Warrell Creek in spring, summer and autumn as per methods outlined in Appendix B for year 4, 6 and 8 Post 5 years, the Roads and Maritime Roads Asset Division will undertake monitoring of fauna fencing on a regular basis	Roads and Maritime /AFJV	No road kill of Giant Barred Frog resulting from operation of highway	Review the integrity of the fence, its design, its extent of permanent fencing.

7.0 Monitoring Program

The monitoring methodology for the Giant Barred Frog on the WC2NH Project is included in **Appendix B**. This methodology has been peer reviewed by a Giant Barred Frog expert, Dr Frank Lemckert and is summarised in this section of the report. The objectives of the monitoring program are:

- To demonstrate through the life of the Project that mitigation has maintained or improved population sizes and habitat of the Giant Barred Frog. The use of preconstruction, during construction and post construction monitoring to measure both frog distribution, abundance and habitat quality with defined thresholds will be used to measure the overall performance of the mitigation; and
- To ensure that mitigation measures are effective in maintaining Giant Barred Frog connectivity near the Project.

7.1 Giant Barred Frog Population Monitoring

7.1.1 Objectives

To demonstrate through the life of the Project that mitigation has maintained or improved population sizes and habitat of the Giant Barred Frog. The use of preconstruction, during construction and post construction monitoring to measure frog distribution, abundance and habitat quality with defined thresholds will be used to measure the overall performance of the mitigation.

7.1.2 Methodology

See **Appendix B** for Giant Barred Frog monitoring procedure.

7.2 Sites Requiring Monitoring

The monitoring program will be limited to Upper Warrell Creek (ch. 42565) in the southern part of the project corridor. A reference or control site was not proposed because Giant Barred Frogs were found at only one site along the project and pairing this with a control site proved problematic for the following reasons:

- A nearby control site exhibiting the same habitat attributes, 'large slow moving stream in partly cleared farmland' unaffected by the Pacific Highway could not be located. The only site able to be located was Way Way State Forest which differed in its habitat, being a faster flowing stream, unfragmented forest and no agriculture in this part of the catchment (**Figure 1-1**).
- In consultation with EPA representatives, ongoing concern in Chytrid management during construction was considered a critical issue. The risk of managing Chytrid is considerable when workers, machinery and materials are transported from numerous locations. Therefore, increasing risk of chytrid transfer to a control site with very different habitat attributes, does not appear to be a good outcome for the Giant Barred Frog.

7.3 Construction and Post Construction Population and Habitat Monitoring Regime

During construction, the maintenance of existing Giant Barred Frog management actions and site controls will be performed on a regular basis as shown in **Table 5-1**. In addition, frog population and habitat monitoring surveys will be performed in accordance with the details described in **Appendix B**. This will include population monitoring along the existing 1 km transect in spring, summer and autumn of Year 1 and 3 of the construction phase of the project along with annual habitat monitoring. This program will include the monitoring of frog fence breaches during each monitoring event and involve surveys on both sides of the fence to clearly show whether the fence is effective at excluding frogs and thus mitigated a potential threat of road strike. Further details for construction and post construction monitoring are shown in **Table 7-1**.

Table 7-1. Summary of the monitoring schedule, goals, timing, responsibility, performance threshold and corrective actions during the construction and operation (i.e. post construction) phase of the Project.

Monitoring Component	Main Goal	Timing/ Frequency	Responsibility	Performance Threshold	Corrective Actions if Deviation from performance Criteria
Giant Barred Frog population and habitat monitoring	To collect data to demonstrate that mitigation has maintained or improve population sizes and habitat of the Giant Barred Frog	<p>Pre-construction baseline surveys completed between spring 2013 and autumn 2014 (see Appendix B)</p> <p>Continuation of the pre construction field survey program on an annual basis in spring, summer and autumn in Years 1 & 3 (Construction stage of the Project)</p> <p>Continuation of the pre construction field survey program on an annual basis in spring, summer and autumn in Years 4, 6 and 8 (operational stage of the Project)</p>	Roads and Maritime AFJV	<p>Giant Barred Frog recorded along the monitoring transect</p> <p>The detection of Chytrid fungus</p> <p>No breaches in fauna exclusion fencing.</p>	<p>Extend the monitoring transect by 500 m to determine presence in adjacent areas</p> <p>Modify, if appropriate, design of existing measures where feasible and reasonable</p> <p>Advise DoE and EPA and discuss adaptive management actions including assisted plantings. Within two weeks of the change being identified with corrective action agreed by DoE, EPA and Roads and Maritime implemented within 3 months</p> <p>Consider additional offset measures to provide additional compensation for animals and habitat lost due to the development</p>

8.0 REPORTING AND DOCUMENT REVIEW REQUIREMENTS

8.1 Pre-construction Baseline Monitoring

See Appendix B for the Preconstruction Baseline Monitoring report.

8.2. Monitoring During Construction

The contractor will submit twice yearly monitoring compliance tracking reports to Roads and Maritime for review. Roads and Maritime will then provide a final copy of the report for information purposes to the Environmental Protection Agency and Department of the Environment. This report will be subject to a review in relation to information provided on the mitigation of impacts for the Giant Barred Frog including the results of the pre-clearing surveys, any road kill related monitoring to date, the population and habitat monitoring surveys performed in Year 1 and 3, , integrity of the temporary frog exclusion fencing and any dewatering processes which have been performed within Giant Barred Frog habitat.

8.3 Post Construction (Operational) Monitoring

The contractor will submit an annual monitoring report to Roads and Maritimes Services for review. Roads and Maritime Services will then provide a final copy of the report for information purposes to the Environmental Protection Agency and Department of Environment. This report will be subject to a review in relation to information on the mitigation of impacts and include comparisons of frog numbers and habitat condition parameters between the preconstruction surveys and the subsequent surveys performed during construction (Year 1 and 3) and the post construction surveys performed in years 4, 6 and 8. This will be reported on bi annually (i.e. every second year).

9.0 CONCLUSION

Surveys for the Giant Barred Frog revealed the presence of a population at Upper Warrell Creek (ch. 42565). Although a reference site was located in nearby Way Way State Forest, the differing habitat at this site precluded it from being used as a control site in the pre and post construction monitoring program.

The strategy has two key objectives, firstly, to demonstrate through the life of the Project that mitigation has maintained or improved population sizes and habitat of the Giant Barred Frog. This is being delivered via a set of upfront management actions, centered on the identification and protection of Giant Barred Frog habitat, suitably experienced persons conducting pre-clearing surveys during early works when site controls are being established, clearing supervision with surveys being performed during suitable weather conditions and dewatering processes to capture tadpoles along with the installation of temporary and permanent frog fencing throughout the construction and operating phases of the Project. The use of pre and post construction monitoring to measure frog distribution, abundance and habitat quality with defined thresholds will be used to measure the overall performance of the mitigation.

In the event that significant negative changes are recorded, the second objective of this management strategy focuses on advising the Department of Environment (DoE) and Environmental Protection Agency (EPA) and agreeing on adaptive actions to be undertaken within three months to reverse the negative trend. Where the implementation of this management strategy identifies the mitigation as being unsuccessful, offsetting will be undertaken by the Roads and Maritime.

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11.0 APPENDIX A – LOCATION OF HABITAT SAMPLING POINTS

Table A-1. Location of habitat sampling sites and zones at the Upper Warrell Creek monitoring site and zones relevant to Figure 2-1.

Label/Zone	Easting GDA 94	Northing GDA 94
(Upstream) Zone 21	488905	6593837
Zone 21	488938	6593878
Zone 20	488978	6593903
Zone 19	489014	6593946
Zone 18	489046	6593992
Zone 17	489089	6594013
Zone 16	489133	6594030
Zone 15	489171	6594047
Zone 14	489206	6594072
Zone 13	489232	6594106
Zone 12	489243	6594152
Zone 11	489253	6594206
Zone 10	489274	6594256
Zone 9	489254	6594295
Zone 8	489261	6594342
Zone 7	489278	6594381
Zone 6	489293	6594422
Zone 5	489306	6594464
Zone 4	489319	6594520
Zone 3	489322	6594568
Zone 2	489313	6594618
(Downstream) Zone 1	489305	6594671

12.0 Appendix B – Pre-construction Baseline Monitoring

See next page.



WARRELL CREEK TO NAMBUCCA HEADS:

GIANT BARRED FROG PRE-CONSTRUCTION BASELINE MONITORING



AUGUST 2014

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1.0 INTRODUCTION

1.1 Project Overview and Background to this Monitoring

The Pacific Highway Upgrade Program is a joint commitment by the Australian and New South Wales (NSW) governments to improve the standard and safety of the Pacific Highway between Hexham and the Queensland border. The Pacific Highway Upgrade Program includes the upgrade of the Pacific Highway between Warrell Creek and Urunga (WC2U) comprised of approximately 42 kilometres of dual carriageway road that would bypass the towns of Warrell Creek, Macksville, Nambucca Heads and Urunga on the Mid North Coast of NSW. The WC2U Project has been divided into two stages and includes the following:

- Stage 1 consisting of the northern 22.5 kilometres of the Project between Nambucca Heads and Urunga (NH2U).
- Stage 2 consisting of the southern 19.5 kilometres of the Project between Warrell Creek and Nambucca Heads (WC2NH).

The Environmental Assessment (EA) prepared for the Warrell Creek to Urunga Pacific Highway Upgrade project identified potential habitat for the Giant Barred Frog at several creeks and drainage lines in the northern half of the study area, through Nambucca, Little Newry and Newry State Forests (SKM 2010). The EA identified the proposal as having the potential to impact on this species as it would directly traverse streams and rivers across the study area. Subsequent surveys of the project route and all freshwater streams between December 2011 and November 2013 (i.e. summer/spring) resulted in the discovery of a Giant Barred Frog population at Upper Warrell Creek at ch. 42565 (Lewis 2014). Consequently, a Giant Barred Frog Management Strategy was developed to ensure the management of this species during the construction and operation of the Upgrade which included a requirement to perform pre-construction monitoring prior to construction (Lewis 2014).

In the context of the above, Lewis Ecological Surveys (LES) was engaged by the NSW Roads and Maritime Services (RMS) to implement the pre-construction baseline monitoring for the Giant Barred Frog (*Mixophyes iteratus*) between spring 2013 and autumn 2014.

1.2 Purpose and Objectives

This pre-construction baseline monitoring fulfils the pre-construction monitoring commitments of the Giant Barred Frog management strategy (Lewis 2014). This includes the implementation of field survey techniques as specified in the management strategy and outlines key biological components of the frog population and the prevailing habitat and water quality attributes prior to construction commencing.

1.3 Subject Species – Giant Barred Frog

1.3.1 Description

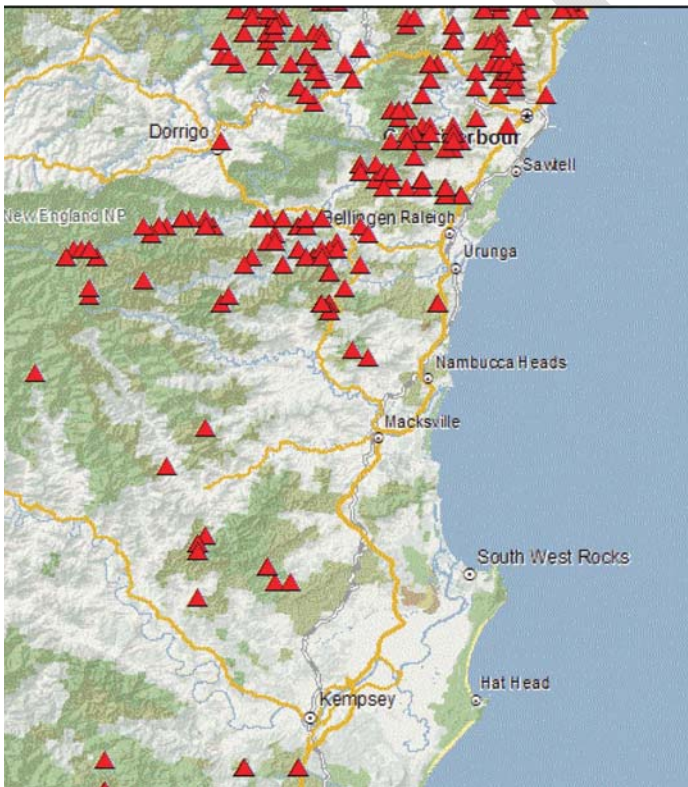
The Giant Barred Frog (*Mixophyes iteratus*) is a large, dark-olive green to black coloured frog that grows to 115 mm. It has a pointed snout and a broad lateral band of dark spots dividing the dark dorsal surface from the white or pale yellow, ventral surface (underside). The limbs have dark crossbars. The hind side of the thighs are black with large yellow spots. Two joints of the fourth toe are free of web (Cogger 2000). The skin is finely granular above but smooth below. The call of the male Giant Barred Frog is a deep guttural grunt (OEH 2014).

Giant Barred Frog tadpoles are large and grow to over 100 mm in length. They are deep-bodied and ovoid, with a tail length twice that of the body. The tadpole's eyes are dorsolateral. The tadpoles are coloured yellow-brown above with dark spots and a dark patch at the base of tail. The underside is silver-white. The intestinal mass is obscured but the heart and lungs are visible from below (except near metamorphosis). The tail is thick and muscular (Anstis 2002). Fins are low and opaque with dark flecking (except the anterior half of the ventral fin; Meyer *et al.* 2001).



Plate 1-1. Giant Barred Frog (*Mixophyes iteratus*).

1.3.2 Distribution



The Giant Barred Frog is currently known from mid to low altitudes below 610 m above sea level (Hines *et al.* 2004), along the Coast and ranges from south-eastern Queensland to the Hawkesbury River in NSW. North-eastern NSW, particularly the Coffs Harbour-Dorrigo area, is now a stronghold (Figure 1-1). Considered to have disappeared south of the Hawkesbury and there are no recent records from the Blue Mountains (Hines and SEQTFRT 2002). Between about Kempsey and Urunga the species appears to be patchily distributed with some confirmed recent locations from upper Warrell Creek and in smaller fast flowing streams in Way Way State Forest (Lewis 2014; Figure 1-1).

Figure 1-1. Localised distribution of Giant Barred Frog between Kempsey and Urunga.

Note - Triangles represent approximate location as sensitive 2 species.

1.3.3 Habitat and Ecology

The Giant Barred Frogs forage and live amongst deep, damp leaf litter in rainforests, moist eucalypt forest and nearby dry eucalypt forest, at elevations below 1000 m. Whilst it has been observed to prefer a closed forest canopy with a relatively light cover of vegetation at ground level (Aland and Wood 2013), they have been found in cleared or disturbed

areas, for example cattle farms with vegetated riparian strips and regenerated logged areas (Ingram and McDonald 1993; Hero and Shoo n.d., cited in Hines *et al.* 2004; Lemckert and Brassil 2000; Lewis and Rohweder 2005). Many sites where the Giant Barred Frog is known to occur are the lower reaches of streams which have been affected by major disturbances such as clearing, timber harvesting and urban development in their headwaters (Hines *et al.* 1999).

Giant Barred Frogs breed around shallow, flowing rocky streams from late spring to summer. Females lay eggs onto moist creek banks or rocks above water level, from where tadpoles drop into the water when hatched. Tadpoles grow to a length of 80–100 mm and take up to 14 months before changing into frogs. When not breeding, the frogs disperse hundreds of metres away from streams. They feed primarily on large insects and spiders.

1.3.4 Conservation Status

In NSW, the Giant Barred Frog is currently listed as Endangered pursuant to the NSW *Threatened Species Conservation Act* (1995) and Commonwealth *Environmental Protection and Biodiversity Conservation Act* (1999) (OEH 2014; SPRAT profile).

DRAFT

2.0 SURVEY METHODS

Field surveys were performed in accordance with the approved Giant Barred Frog management strategy for the Warrell Creek to Urunga Pacific Highway Upgrade (Lewis 2013). At this time of implementing the pre construction monitoring the strategy and survey requirements had been approved by the Department of Planning and Environment (DP&E) and the Environmental Protection Authority (EPA).

2.1 Timing of Surveys

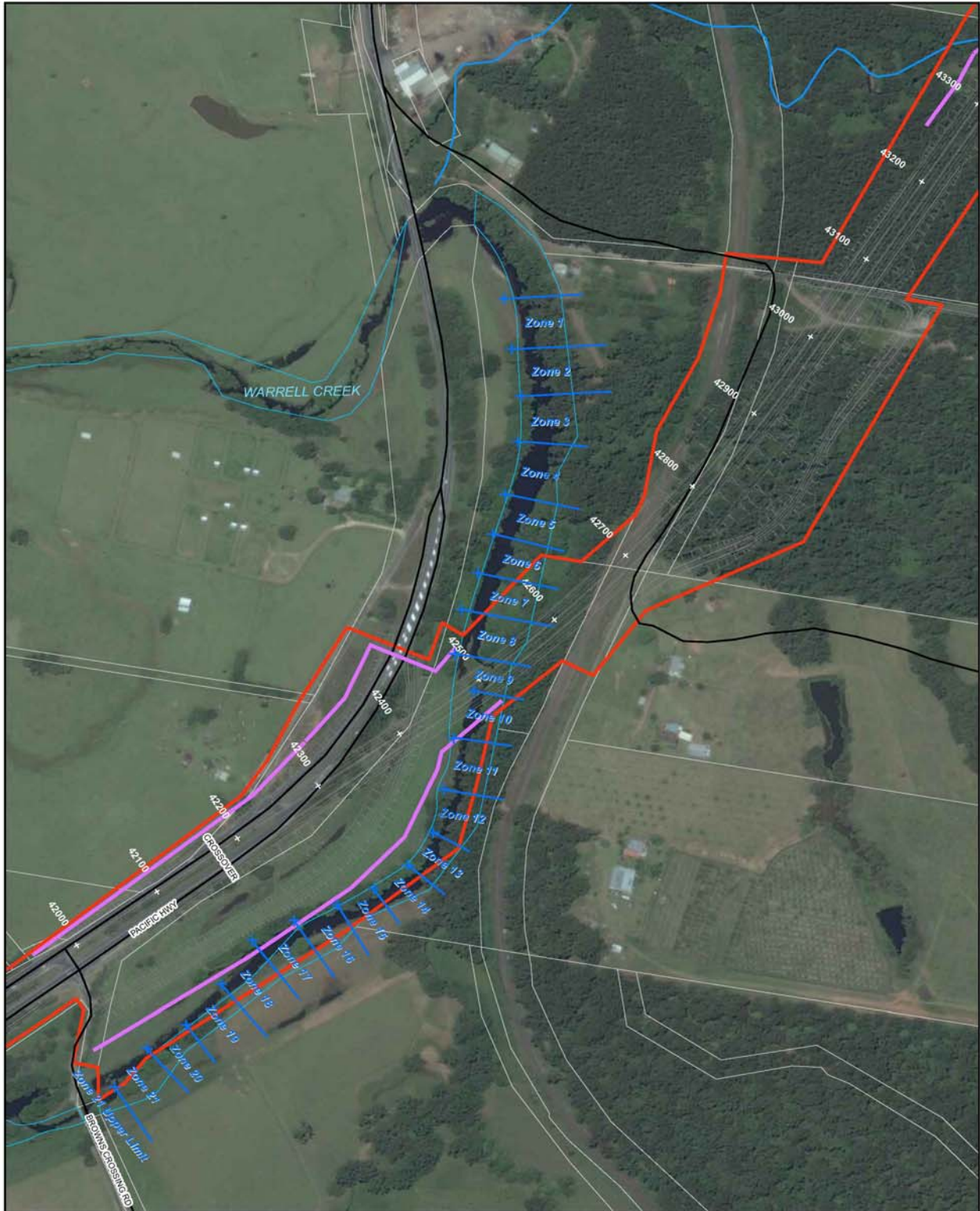
Field surveys were undertaken at the following times:

- Spring sampling was undertaken on the 20th September 2013 in response to a rainfall trigger event of 10.8 mm being recorded on the 17th September (Macksville Country Club Station No. 059018).
- Summer sampling was undertaken on the 29th January 2014 in response to a rainfall trigger event of 22.6 mm recorded on the 23rd January with an additional 13.2 mm being recorded in the 7 days leading up to the field survey.
- Autumn sampling was undertaken on the 2nd April 2014 in response to a rainfall trigger event of 20.6 mm recorded on the 28th March with an additional 10.4 mm being recorded in the 7 days leading up to the field survey.

2.2 Frog Surveys

Frog surveys were performed in the manner outlined in the Giant Barred Frog management strategy (Lewis 2013). This involved:

- Surveys being performed within 7 days of a rainfall event exceeding 10 mm in 24 hours using the Bureau of Meteorology (BOM) weather stations at Macksville Country Club (059018) and Fisherman's Reach (059143) when there was some missing data or some other discrepancy. For example, isolated thunderstorm activity that resulted in sporadic rainfall;
- 1 km transect with 450 m either side of the construction footprint (~100 m represents construction footprint) and divided into 20 x 50 m zones (Figure 2-1);
- Each field survey involved a meandering transect on both sides of the stream bank with all captured Giant Barred Frogs permanently marked using a PIT tag (i.e. micro-chipped) and specifically a Trovan Nanotransponder (000735#### series). Survey effort ranged from 3.25 – 6.25 hours per transect with variability in time length attributed to variations in habitat, accessibility and the number of frogs being processed;
- For each frog, the following information was collected:
 - Location according to demarcated survey zone (20 x 50 m zones);
 - Distance from the stream edge measured to the nearest 0.1 m;
 - Position within the microhabitat (i.e. under litter, above litter, exposed, on rock/log)
 - Sex (male, female, unknown);
 - Age class (adult = >60 mm; sub adult = 40-60 mm; juvenile = <40 mm)
 - Snout-vent length (mm);
 - Weight (grams); and
 - Breeding condition with:
 - males assessed on the colouration of their nuptial pads (i.e. no colour, light, moderate, dark) in accordance with a classification developed by Lewis Ecological Surveys (Table 2-1);
 - females based on whether they are gravid (i.e. typically adult weighing > 100 grams) or not gravid (egg bearing);
 - frogs with a snout vent length of <60 mm were classified as immature.



— Project Boundary — Detail Road Design (Construction Footprint) — Stream zone — Giant Barred Frog Fencing

	Scale: Calceolaria, Microlepis and Trichia Authority: 2017 Quail Shrike Frog Survey Survey: Zones 1-20 Date: 2018	Points: Geoscience Australia 2010 Name: Warrell Creek (Topographic elevation)	Figure 1: Giant Barred Frog Management Zones
	Disclaimer: This plan is based on or contains data provided by others. Lewis Ecological gives no warranty in relation to the data concerning accuracy, reliability, completeness, currency or suitability, and accepts no liability for any loss, damage or costs (including consequential damage) resulting from use of the data. Data must not be used for direct monitoring or be used in relation to any other data.	Coordinate System: MGA94 Projection: Transverse Mercator	

Figure 2-1. Location of the monitoring transect and recording zones 1-20.

2.3 Swabbing for Chytrid Fungus

Swabbing for Chytridiomycosis or Chytrid fungus was undertaken during the summer monitoring event on the 29th January 2014. The objective of this was to establish a pre-construction baseline indices as to the exposure of Chytrid fungus given the overall lack of information on the extent of this disease within Giant Barred Frog populations on the mid north coast given the disease is a highly contagious, highly virulent disease of frogs. Chytrid Fungus is currently listed as a key threatening process for frogs pursuant to the NSW *Threatened Species Conservation Act* (1995).

Half of all frogs captured during the summer monitoring event were swabbed for Chytrid testing. This involved the use of a sterile swab and wiping the outer skin with a sterile cotton-tipped swab. The swab is wiped over the body creases, such as under the arms and inside of the thighs and groin, to collect loose skin samples. Swabs were then placed into a sterile container and held in a refrigerator until they could be delivered to Newcastle University for testing.

All handling procedures were undertaken in accordance with the *Hygiene Protocols for the Control of Disease in Frogs* (DECW 2008).

2.4 Tadpole Surveys

Tadpole surveys were undertaken during the spring survey using the following procedure:

- The 1 km transect was divided up into 20 x 50 m zones with seven zones in the downstream corridor, five zones partially or totally within the construction corridor and eight zones upstream of the road corridor.
- Within each zone, one bait trap (~300 mm x 200 mm) were installed and left operating for 3 hours. This equated to 20 bait traps and 60 hours of survey effort.

Some dip-netting was undertaken to confirm the presence of Giant Barred Frog tadpoles during both the spring and autumn monitoring. During these surveys the presence of exotic fish was also recorded.

Table 2-1. A key developed for determining reproductive condition in male barred frogs (*Mixophyes*).

Nuptial Pad Colour	Comments
No Colour	<ul style="list-style-type: none"> • Males may be active or dormant but don't present as being sexually active to mate with females. • No colour can occur at any time throughout the year but pronounced periods include dry springs and late autumn with the onset of winter.
Light	<ul style="list-style-type: none"> • Some colouration indicating frogs are likely to become active (late winter) or have been active but generally not breeding. For example, prevailing weather conditions are unsuitable. • Frogs with light nuptials are generally on the shoulder periods of breeding events and a small percentage of the male population is likely to classify into this category at almost any time of the year apart from June and July.
Moderate	<ul style="list-style-type: none"> • Males are normally active, will often readily respond to calls. Ready to mate with gravid females if weather conditions are suitable. • These frogs may occasionally be involved in intraspecific aggression indicating their readiness to mate with females. • Colouring may be evident between August-May and is considered cyclic and surrounding breeding events.
Very Dark	<ul style="list-style-type: none"> • Males are normally active, ready to mate with gravid females if conditions are suitable. • Some observations of intraspecific aggression can occur between males at this stage. • Colouring may be evident between August-May and is considered cyclic with early season suspected of being driven through warming air temperature whilst prevailing rainfall conditions are considered the primary queue during summer and autumn.

2.5 Abiotic Data

The following abiotic variables were collected during the survey.

- Rainfall measured in four scales:
 - During the survey;
 - Within past 24 hours;
 - Within past 7 days; and
 - Within past 30 days.
- Relative humidity measured with wet/dry bulb thermometer at the start and finish of the frog survey and averaged;
- Air temperature measured with a thermometer at the start and finish of the frog survey and averaged;
- Wind speed measured in subjective scale (0= no wind, 1 = light rustles of leaves on trees, 2 = leaves and branches moving and 3 = whole canopy moving);
- Water level measured with a permanently installed water staff or an electronic device if available from the Bureau of Meteorology (BOM).

2.6 Habitat Data

The following habitat data were recorded at each of the 20 demarcated zones and for both the southern and northern riparian zones given they differed markedly:

- Landuse: Description of existing land uses of dairy cattle farming, beef cattle farming, private natural reserve;
- Broad vegetation type within the immediate riparian zone (primary stream bank): Riparian Rainforest, Dry Sclerophyll Forest, Woodland, Mallee; Heath/Shrub; Sedgeland, Grassland or Cleared Land;
- In stream physical characteristics including:
 - Stream width and depth (metres);
 - Presence of pools and/or riffles
 - bed composition (sand, clay, rock, organic or other to be specified);
 - type of emergent vegetation if present
- Stream bank characteristics including:
 - Bank profile expressed as steep, benched or a gradual incline from the water's edge
- Vegetation associated with the stream bank in terms of its foliage projection cover (fpc) for overstorey trees, shrubs and groundcover;
- Groundcover composition including a measure of vegetative ground cover, litter cover, soil cover and exposed rock expressed as a composition percentage of 100%.
- The depth of litter was also measured and assigned to one of the following categories:
 - Deep (>10 mm); Moderate (20-100 mm); Shallow (>0-20 mm); or Absent (0 mm).
- Water quality monitoring with water samples being taken on the day of the summer survey (29th January) and at the next pronounced wet weather period triggering runoff (18th February) following by another dry weather sampling event on the 25th February. The samples were measured for the following:
 - Heavy Metal including arsenic, cadmium, chromium, copper, lead, mercury, nickel and zinc.
- Hydrocarbons from the following groups:
 - Naphthalene group including TRH>C10-C16, TRH>C10-C16 less Naphthalene (F2), TRH>C16-C34, TRH>34-C40, TRH C6-C10 and TRH C6-C10 LESS BTEX (F1);
 - BTEX group including Benzene, Ethylbenzene, m&p-Xylenes, o-Xylene, Toluene and Xylenes – total
- Nutrients including Nitrogen (as N), Suspended Solids and Total Phosphorus
- Field Physicochemical data including Dissolved Oxygen, Conductivity, pH, Temperature and Turbidity

Water quality data was analysed by Coffey Geotechnics using a National Association of Testing Authorities (NATA) accredited laboratory.

2.7 Determining Population Size

The Lincoln–Petersen method (also known as the Petersen–Lincoln index) was used to calculate the population size. This method was used in preference to the triple catch calculation given the low numbers of frogs recorded during the spring survey would only inflate the population estimate. The Lincoln–Petersen method is used to estimate population size when only two visits are made to the study area and assumes the study population is "closed". In other words, the two visits to the study area are close enough in time so that no individuals die, are born, move into the study area or move out of the study area between visits. The model also assumes that no marks fall off animals between visits to the field site by the researcher, and that the researcher correctly records all marks.

The Lincoln–Peterson estimator is asymptotically unbiased as sample size approaches infinity, but is biased at small sample sizes. An alternative less biased estimator of population size is given by the Chapman estimator.

$$N = \frac{(M + 1)(C + 1)}{R + 1} - 1,$$

Where, as before:

N = Estimate of total population size

M = Total number of animals captured and marked on the first visit

C = Total number of animals captured on the second visit

R = Number of animals captured on the first visit that were then recaptured on the second visit

An approximately unbiased variance of N , or $\text{var}(N)$, can be estimated as:

$$\text{var}(N) = \frac{(M + 1)(C + 1)(M - R)(C - R)}{(R + 1)(R + 1)(R + 2)}.$$

As in all estimates, it is also useful to have some information about the uncertainty of the estimate (as measured by the standard error (SE), and/or by 95% confidence intervals). The standard error of the estimate of N is given by the following formula:

$$\text{SE} = \sqrt{\frac{[(M+1)(C+1)(M-R)(C-R)]}{(R+1)^2(R+2)}}$$

The standard error gives an idea of where the sample mean is likely to be found if the experiment were conducted repeatedly. From the standard error, we can also calculate the 95% confidence limits of the estimate (which defines the range of values within which the true population size is likely to lie with 95% certainty), using the following formula:

- 95% confidence interval = $N \pm (1.96)(\text{SE})$

3.0 RESULTS

3.1 Abiotic Data

The spring sampling was undertaken on the 20th September 2013 in response to a rainfall trigger event of 10.8 mm on the 17th September (Table 3-1). This was the most notable rainfall event since the 14th June when 14.8 mm was recorded and remained the most notable rainfall up until the 19th October when 22.4 mm was recorded (Appendix). Although the survey was performed during relatively mild temperatures of 15.1°C the dissipating cloud cover reduce the relative humidity further from 59% at 1850 hours to 40% at 2135 hours.

The summer sampling was undertaken on the 29th January 2014 in response to a rainfall trigger event of 22.6 mm on the 23rd January. This was the first suitable rainfall event for the summer period with the previous suitable event occurring on the 30th November (outside summer sampling period) when 32.6 mm was recorded (Appendix). The field survey was performed during mild summer temperatures of 20.9°C at 2100 hours which dropped to 14.5°C at the completion of the survey at 0245 hours. The humidity remained high throughout the sampling period ranging from 74-91% despite there being no cloud cover or rain being recorded in the past 5 days.

The autumn sampling was undertaken on the 2nd April 2014 in response to a rainfall trigger event of 20.6 mm on the 28th March. Around this time there were a number of sporadic rainfall events with 24 hours total approaching and often exceeding 10 mm (Appendix). The air temperature was warmer than expected with 21.1°C at 1948 hours and this declined to 18.4°C by 0030 hours making it warmer overall than the summer survey. The humidity remained high throughout the sampling period ranging from 77-91% and although there was very little cloud cover there had been some recent rainfall of 3.2 mm in the past 24 hours.

Table 3-1. Abiotic conditions during the pre-construction baseline monitoring at Upper Warrell Creek.

Date	Time	Time (24 hours)	Air Temp °C	Water Temp °C	Cloud Cover %	Humidity %	Wind ¹	Rain ²	Stream Depth (mm)
20.9.2013	Start Time	1850	15.5	13	15	59	0	0	nd
	Finish time	2135	14.6	13	0	40	1	0	nd
	Spring Summary	2 hours 45 minutes	15.05	13	7.5	49.5	0.5	0	nd
29.1.2014	Start Time	2100	20.9	24	0	74	0	0	nd
	Finish time	0245	14.5	24	0	91	0	0	nd
	Summer Summary	5 hours 45 minutes	17.7	24	0	82.5	0	0	nd
2.4.2014	Start Time	1948	21.1	19	15	77	0	1	nd
	Finish time	0030	18.4	19	0	91	0	0	nd
	Summer Summary	4 hours 48 minutes	19.75	19	7.5	84	0	0.5	nd
Rain	During (mm)		Past 24 Hours (mm)			Past 7 Days (mm)		Past 30 Days (mm)	
20.9.2013	0		0			18.3		18.3	
29.1.2014	0		0			31.2		56.8	
2.4.2014	0		3.2			40.4		77.4	

3.2 Giant Barred Frog - Demography

3.2.1 Captures and Age Classes

There was a total of 47 Giant Barred Frogs recorded during the pre-construction baseline survey (Table A3 in Appendix). This comprised:

- 38 individuals classified into the follow age classes:
 - 22 adults with 11 females and 11 males
 - 8 sub adults; and
 - 8 juveniles.

The remaining nine frogs included five recaptures and four adults identified as one female and three males that could not be captured to verify whether they had been previously PIT tagged. Consequently they could not be used in determining the population structure nor population estimate (see below).

The seasonal trend of frog captures is shown in Figure 3-1. Spring surveys recorded only one sub adult frog with this individual being captured from zone 10 on the northern bank which lies partially within the proposed construction footprint. The summer survey recorded 24 frogs comprising eight juveniles, three sub adults and 13 adults with this later group being comprised of five females and eight males. Three of the recorded male frogs could not be captured for micro-chipping. The autumn survey recorded 22 frogs comprising six sub adults and 16 adults with this later group being comprised of eight females and eight males (i.e. ratio of 1:1). One female frogs avoided capture during this monitoring period.

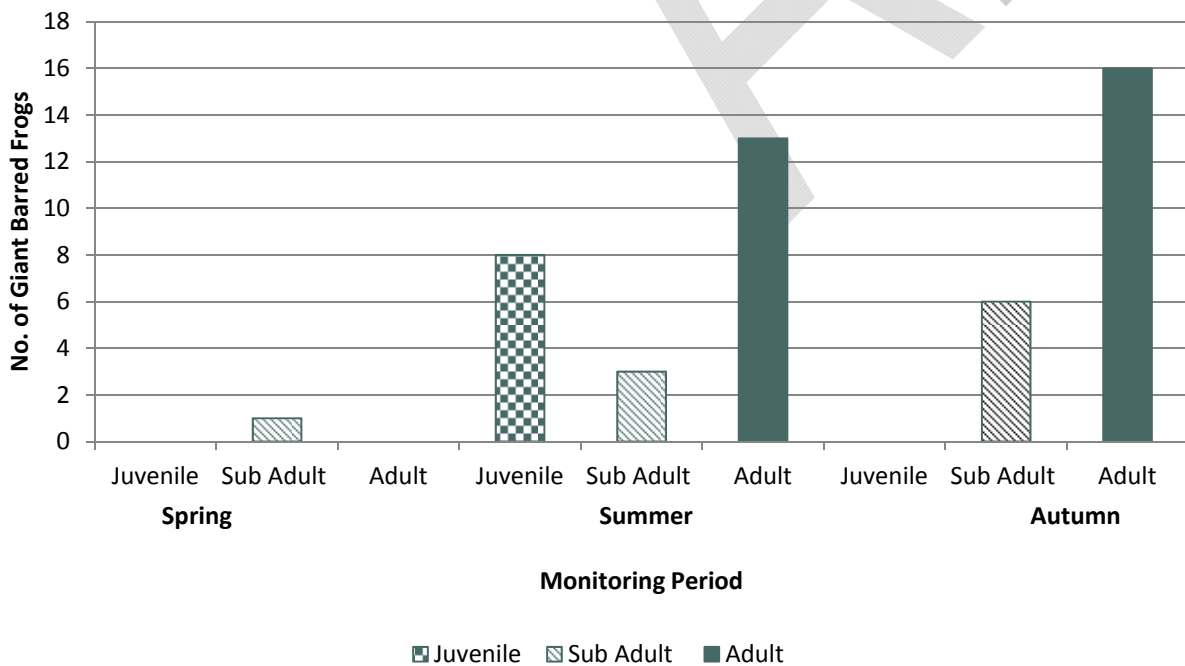


Figure 3-1. Age class structure of Giant Barred Frog across the three monitoring periods.

3.2.2 Calculating Population Size

The two samples collected during the summer and autumn monitoring have been used to calculate population size. The summer monitoring recorded 24 frogs, however, three of the male frogs could not be captured and were consequently removed from the population estimate (i.e. they weren't PIT tagged to confidently identify them as new or recaptured individuals). The autumn monitoring recorded 22 frogs with one frog avoiding capture and thus leaving 21 frogs. Five of the 21 captured frogs were recaptures from the summer sampling. Using the Lincoln–Peterson estimator for all frogs captured and PIT tagged the population has been calculated as follows:

- Population Estimate = 79.7 Giant Barred Frogs
- Standard Error = 22.17
- 95% confidence interval = 46.2

If this were to be divided proportionally between the recorded age classes then the population would be estimated to comprise:

- 16 Juveniles;
- 19 sub adults; and
- 45 adults with a male to female sex ratio of approximately 1:1.

The use of the adult population estimate may be more applicable given the subject animal is an *R* selected species which produces large numbers of offspring with a low probability of surviving to adulthood. Using the Lincoln–Peterson estimator for all adult frogs captured and PIT tagged the population has been calculated as follows:

- Population Estimate = 43 adult Giant Barred Frogs
- Standard Error = 13.59
- 95% confidence interval = 43 ± 26.6

3.3 Presence of Chytrid Fungus

One of the 17 frogs returned a positive test for the presence of chytrid fungus (*Batrachochytrium dendrobatidis*). The test was not conclusive across all three replicates but rather just one of the three (Table A4 in Appendix). This frog (ID:07359051) was recorded from the northern bank in Zone 8 which forms part of the proposed construction zone (Figure 2-1).

3.4. Habitat Use

3.4.1 Frog Distribution Along the Transect

Giant Barred Frogs were recorded between survey zones 2 through to 20 and occupied 14 (70%) of the 20 zones (Figures 2-1 and 3-2). Ten frogs were recorded below the construction footprint whilst 11 were recorded above it (Figure 3-3). Twenty-one frogs were recorded within the construction footprint with another six recorded from zones that may occur partially within the footprint depending on the final clearing footprint (Figure 3-3).

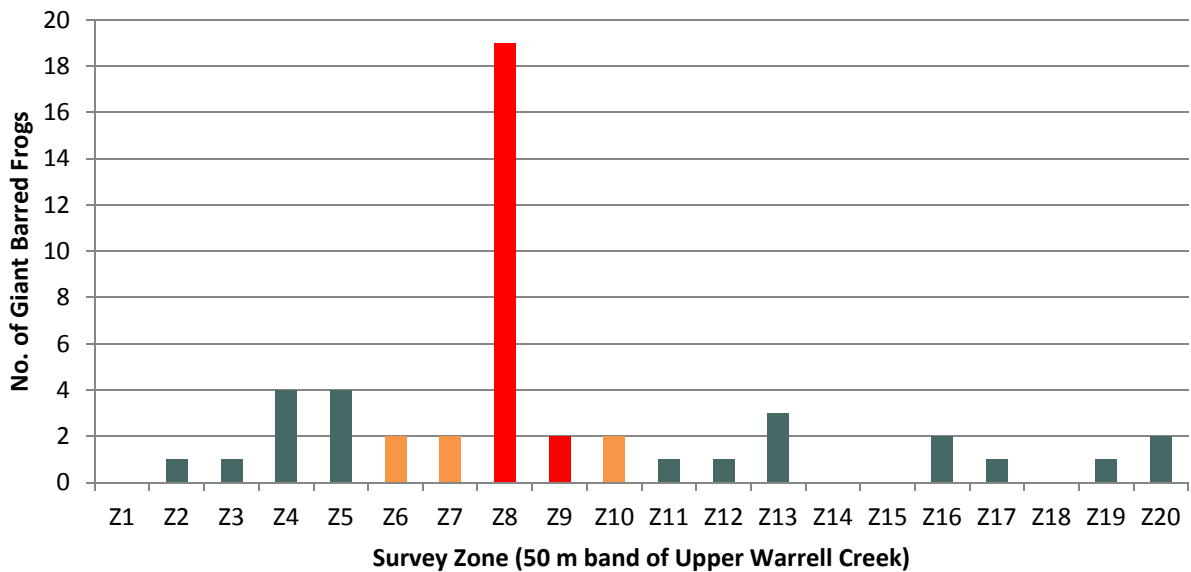


Figure 3-2. Cumulative number of frogs recorded in each of the monitoring zones.

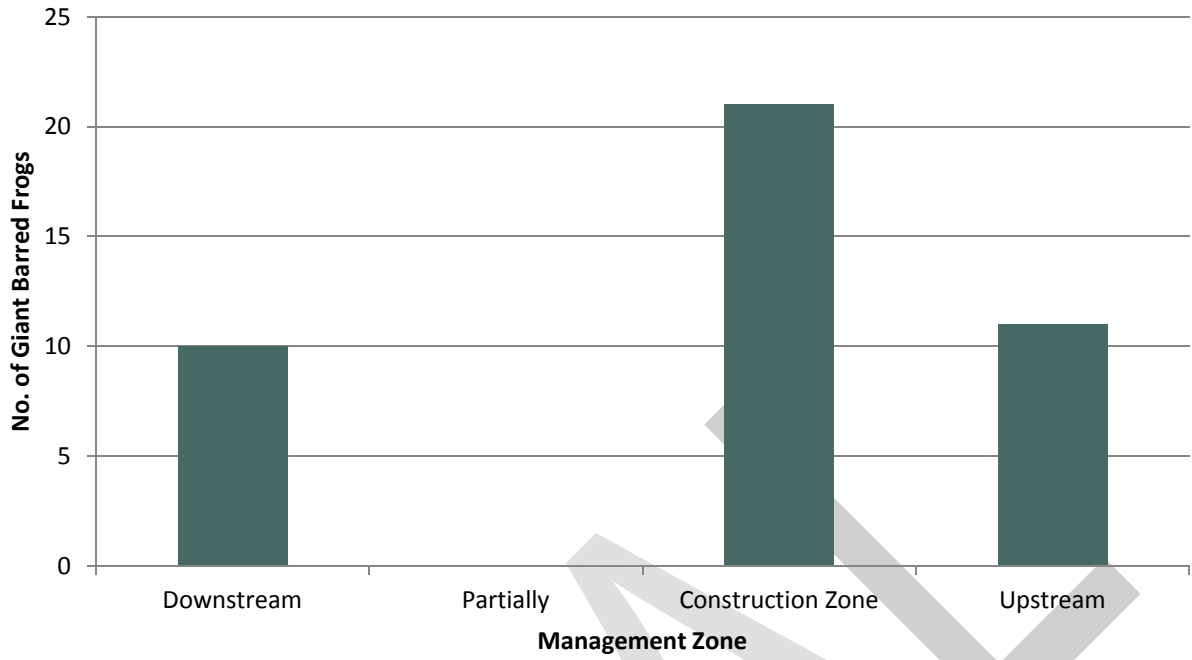


Figure 3-3. Cumulative number of frogs recorded in each of the monitoring zones.

3.4.2 How the Frogs are using the Existing Habitat

The mean distance frogs were recorded from the stream edge ranged from 1.1 m in spring (n=1) to 5.9 m in autumn (n=22, SE = 3.51; Figure 3-4). Eight frogs were recorded at distances of 10-22 m from the stream edge with 75% of these being adults comprising two males and four females. The two remaining frogs were sub adults observed at 10 and 11.5 m from the stream edge.

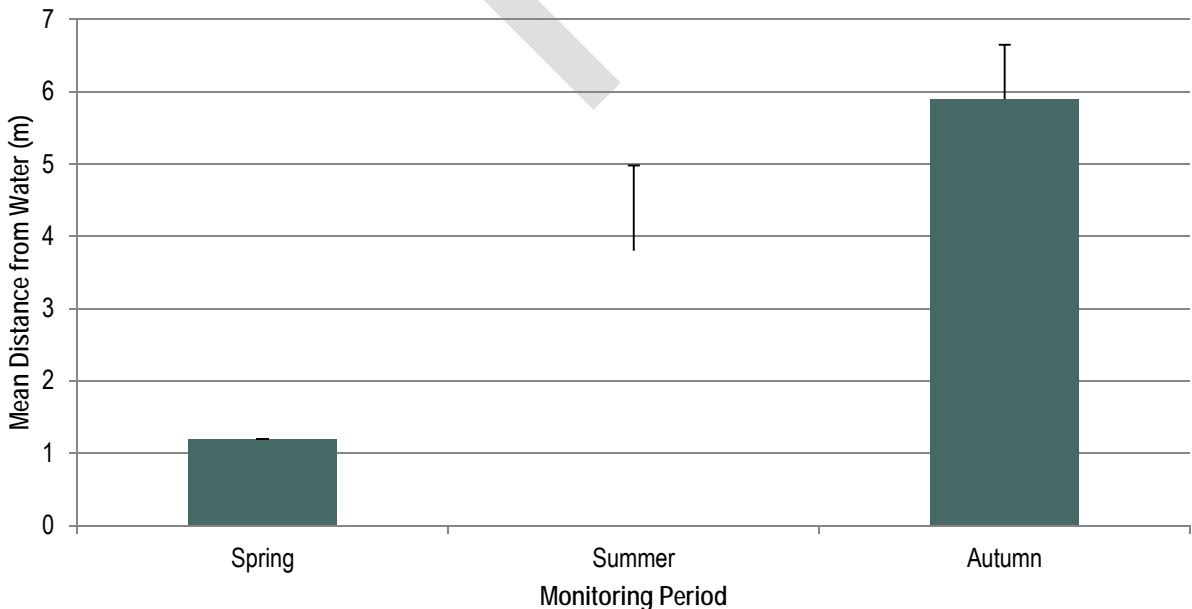


Figure 3-4. Mean distance (+SE) from water during each of the three monitoring periods.

3.4.3 Recaptured Frogs

Three of the five recaptured frogs remained in the same zone between the summer and autumn sampling (Table A3 in Appendix). The two remaining frogs, an adult male with moderate and dark nuptials moved between Zone 5 and 8 or about 120 m whilst a sub adult frog had moved from Zone 10 down to Zone 8 or about 75 m (Figure 2-1). During the two monitoring periods this particular individual (ID:7356782) had grown around 7 mm (14% increase in length) and attained an extra 5.75 grams (38% increase).

3.4.4 Breeding Cues

None of the captured female frogs were noted as being in a gravid condition (egg bearing condition) during each of the surveys. In contrast, all of the captured male frogs displayed some reproductive scoring with two assigned as having 'light' coloured nuptials, seven with 'moderate' coloured nuptials and four with 'dark' coloured nuptials. Individuals from each of the reproductive categories were present during both the summer and autumn monitoring events. No adults were captured during the spring monitoring.

No tadpoles were captured during the survey. All three monitoring events recorded data that suggests the Giant barred Frog population is breeding within the monitoring transect. This includes:

- One sub adult during the spring survey and represents the first time the population is producing offspring;
- Eight juveniles and two sub adults during the summer survey; and
- Six sub adults during the autumn survey.

This data would suggest that metamorphosis for most of the juvenile frogs occurred sometime between November and December with some extending through into early January. There was some clustering of juvenile captures and these were associated with the back channels bordering the southern bank of Zone 9 and Zone 18.

3.5 Habitat Condition

3.5.1 General Land Use and Broad Classification Type

The habitat data collected has characterised the site as being located predominantly within degraded agricultural land with the southern bank being entirely used as a beef cattle farming enterprise using set stocking principals (i.e. no rest period and constant herbivory pressure). The northern bank contains a mosaic of land uses with dairy cattle farming extending between Zone 14 upstream to Zone 21. This type of farming differs from that on the southern bank whereby the existing pastures are periodically intensely grazed and then left to recover leading to a dense sward of introduced grasses with native herbs and annuals growing in those periodically inundated areas. Below this point and up until Zone 11 the area is undisturbed closed forest classified here as riparian rainforest but also consistent with other vegetation classifications that may describe it as wet sclerophyll forest. Zones 8-10 are also riparian rainforest but with some minor disturbances associated with an access track to an irrigation pump. Below this point in Zones 5-7 there is a disturbed area which may have historically been grazed by cattle and be left to passively regenerate. Some periodic maintenance of taller trees tends to occur in this area with a powerline easement running east-west. Below this point, Zones 1-4 tend to be remnant vegetation again describe here as riparian rainforest or wet sclerophyll forest.

Compositionally, the southern bank is comprised of 37% as cleared land and 63% as disturbed riparian rainforest whilst the northern bank is comprised of 25% cleared land, 25% as disturbed riparian rainforest and 50% as undisturbed riparian rainforest.

3.5.2 Characteristics of the Riparian Terrestrial Zone

The vegetation on top of the primary or main stream bank is patchy distributed along the transect with some notable gaps between Zones 14-18 on the northern bank and Zones 0, 10-11 and 18-21 on the southern bank. An estimate of overstorey cover across the entire site was calculated at 50.4% but with marked variation calculated here with a standard deviation of 33.8%. With regard to the four management zones, overstorey vegetation cover was highest below the construction footprint (mean=62%; SD=25.4%) and at its lowest above the construction footprint (mean=43%; SD=37.8%). This was the same for shrub cover with the range varying from 13.2% (SD=8.1%) below the construction footprint to 6% (SD=6.38) cover above the construction footprint. The mean groundcover across the entire

transect was calculated at 54.3% (SD=33.7%) with this large variation attributed to variable tree cover with exposed tree less areas supporting higher levels of groundcover comprised often as improved pasture grasses.

The extent of litter cover was calculated at 33% (SD=29.6%) across the site but this varied between 24% (SD=24.1%) above the construction footprint to 47.8% (SD=30.8%) or almost twice that below the construction footprint. Apart from the management zone partially within the construction footprint, bare dirt was similar across the transect with a range of 8.5-14% recorded. Typically higher levels were recorded where cattle had accessed areas beneath trees on the primary bank as cattle camps displacing the expected leaf litter. Litter depth itself was calculated as 30% of the site containing no litter at all, 30% containing on shallow areas (0-20mm) of litter, 22.5% as having moderate (20-100 mm) litter and 17.5% as having deep litter (>100 mm) present with a particular zone. Most of the management zones had a range of litter depths (Table 3-3).

The stream bank profile is characterised with 16% of the transect containing steep sided banks, approximately 55% having benched or stepped banks and the remaining 29% being gradual (Table 3-1). Areas upstream and downstream of the construction site exhibit steep sided banks but not within the construction limit nor the partial zones. Some gradual banks were recorded in each of the management zones. All of the stream banks are comprised of a sandy loam soil type typically on lower catchments in the Warrell Creek area.

3.5.3 Physical Stream Characteristics

Upper Warrell Creek was estimated at 8 m width in Zone 20-21 and it becomes gradually wider reaching 18 m before reducing to around 10 m within Zone 9. At this point there is a riffle where the stream reduces to approximately 4 m in width. There are no other riffle zones within the monitoring transect. Beyond this point it quickly reaches and maintains a 20-25 m width for more than 500 m.

Water depth ranges from 1.5 m at Zone 9 to around 3 m through most of the main pools. The upper reaches of the transect were estimated to be around 2 m in depth. The stream bed itself is made up almost exclusively of sandy silts often with a deep detritus layer. The exception is the riffle zone location in Zone 9 which contains some gravel.

Emergent or floating aquatic vegetation is present in virtually all of the zones with the main species being Water Lilly (*Nymphaea spp*), Knotweeds (*Persicaria spp*) and Common Spikerush (*Eleocharis spachelata*). This later species is limited to the upper reaches of the transect and was recorded in Zones 18-21.

The exotic Mosquito Fish (*Gambusia holbrooki*) was recorded throughout the site. Greater numbers were generally dip-netted around dense aquatic vegetation.

3.5.4 Water Quality Monitoring

The water quality monitoring collected on the 29th January, 18th and 25th February is summarised in Table 3-2. None of the concentrations for the eight heavy metals and 13 Hydrocarbons were recorded at levels exceeding the ANZECC Freshwater Trigger Value. Nitrogen exceeded the trigger value for a lowland rivers in south eastern Australia with mean value of 0.53 mg/L (SD=0.13) although this figure was below 0.5 mg/L during the wet monitoring period. Dissolved oxygen consistently exceed the trigger value for a lowland rivers in south eastern Australia with 2.26-2.55 mg/L recorded across the four management zones and an overall site value of 2.42 mg/L (SD=1.12). The remaining physico chemical data were within the recommended values.

Table 3-2. Summary of the measured habitat attributes across the entire site and at four different management zones.

Attributes/Site	Entire Site	Below	Partially	Construction	Above
Landuse					
Type of existing landuse present	<ul style="list-style-type: none"> • Natural – North Bank: 41% South Bank: 0% • Natural regeneration from past disturbance – North Bank: 18% South Bank: 0% • Farming (Dairy) – North Bank: 41% South Bank: 0% • Farming (Beef) – North Bank: 0% South Bank: 100% 	<ul style="list-style-type: none"> • Natural regeneration from past disturbance - Northern Bank: Zones 0-5 Southern Bank: Nil • Farming (beef) – Northern Bank: Nil Southern Bank: Zones 0-5 	<ul style="list-style-type: none"> • Natural Forest- Northern Bank: Zone 10 Southern Bank: Nil • Natural but regeneration from past disturbance Northern Bank: Zones 6-7 Southern Bank: Nil • Farming (beef) – Northern Bank: Nil Southern bank: Zones 6,7&10 	<ul style="list-style-type: none"> • Natural but regeneration from past disturbance Northern Bank: Zones 8-9 Southern Bank: Nil • Farming (beef) – Northern Bank: Nil Southern Bank: Zones 8-9 	<ul style="list-style-type: none"> • Natural – Northern Bank: Zones 11-13 Southern Bank: Nil • Farming (beef) – Northern Bank: Nil Southern Bank: Zones 11-21 • Farming (Dairy) – Northern Bank: Zones 14-21 Southern Bank: Nil
Broad Vegetation Type	<ul style="list-style-type: none"> • Riparian Rainforest (undisturbed) – Northern Bank: 50% Southern Bank: 0% • Riparian Rainforest (disturbed) – Northern Bank: 25% Southern Bank: 63% • Cleared Land – Northern Bank: 25% Southern Bank: 37% 	<ul style="list-style-type: none"> • Riparian Rainforest (undisturbed) – Northern Bank: Zones 1-5 Southern Bank: Nil • Riparian Rainforest (disturbed) – Northern Bank: Nil Southern Bank: Zones 1-5 • Cleared Land – Northern Bank: Nil Southern Bank: Zone 0 	<ul style="list-style-type: none"> • Riparian Rainforest (undisturbed) – Northern Bank: Zone 10 Southern Bank: Nil • Riparian Rainforest (disturbed) – Northern Bank: Zones 6-7 Southern Bank: Zones 6-7 • Cleared Land – Northern Bank: Nil Southern Bank: Zone 10 	<ul style="list-style-type: none"> • Riparian Rainforest (disturbed) – Northern Bank: Zone 8 Southern Bank: Zones 8-9 • Cleared Land – Northern Bank: Zone 9 Southern Bank: Nil 	<ul style="list-style-type: none"> • Riparian Rainforest (undisturbed) – Northern Bank: Zones 11-13 Southern Bank: Nil • Riparian Rainforest (disturbed) – Northern Bank: Zones 19-21 Southern Bank: Zones 12-18 • Cleared Land – Northern Bank: Zones 14-18 Southern Bank: Zones 11&19-21

Attributes/Site	Entire Site	Below	Partially	Construction	Above
Riparian Terrestrial Zone					
Vegetation					
Overstorey	50.4 (33.8)	62.1 (25.4)	55.2 (36.0)	48.8 (28.7)	43 (37.8)
Shrub Cover	8.7 (7.5)	13.2 (8.1)	11.2 (7.7)	7.5 (5.6)	6 (6.38)
Groundcover	54.3 (33.7)	39.3 (33.0)	48 (38.6)	64 (34.9)	62 (31.6)
Litter Cover	33 (29.6)	47.8 (30.8)	43.5 (38.4)	24.5 (26.7)	24 (24.1)
Soil Cover	12.8 (10.4)	12.8 (9.9)	8.5 (6.1)	11.5 (8.4)	14 (12.0)
Rock	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
Litter Depth	Absent = 30% Shallow =30% Moderate =22.5% Deep = 17.5% Sample Size = 40	Absent = 16.7% Shallow =25% Moderate =16.7% Deep = 41.6% Sample Size =12	Absent = 33.3% Shallow =0% Moderate =33.3% Deep =33.3% Sample Size =6	Absent = 25% Shallow =50% Moderate =25% Deep = % Sample Size = 4	Absent = 36.4% Shallow =31.8% Moderate =18.2% Deep = 13.6% Sample Size = 22
Stream Bank Profile	<ul style="list-style-type: none"> Steep: North: 32% South: 0% Benched: North: 27% South: 82% Gradual: North: 41% South: 18% 	<ul style="list-style-type: none"> Steep: North: Zones 0-3 South: Nil Benched: North: Zone 5 South: Zones 1-5 Gradual: North: Zones 4 South: Zone 0 	<ul style="list-style-type: none"> Steep: North: Nil South: Nil Benched: North: Zone 7 South: Zones 6-7 Gradual: North: Zones 6,10 South: Zone 10 	<ul style="list-style-type: none"> Steep: North: Nil South: Nil Benched: North: Zone 9 South: Zone 8 Gradual: North: Zone 8 South: Zone 9 	<ul style="list-style-type: none"> Steep: North: Zones 12,19&20 South: nil Benched: North: Zones 11,13&21 South: Zones 12-21 Gradual: North: Zones 14-18 South: Zone 11
Stream Bank Composition	Sandy soil - loam	Sandy soil - loam	Sandy soil - loam	Sandy soil - loam	Sandy soil - loam
Stream Characteristics					
Structure	Two long pools with one small rifle zone at Zone 9	One long pool	One long pool	Convergence of two pools with a small riffle	One long pool
Width (m)	8-25 (m=16.6; SD=5.8)	20-25 (m=24.1; SD=1.9)	16-20 (m=18.1; SD=1.8)	10-12 (m=11; SD=1.2)	8-18 (m=13.2; SD=3)
Depth (m)	1.5-3 (m=3; SD=0)	3 (m=3; SD=0)	3 (m=3; SD=0)	1.5-1.8 (m=1.7; SD=0.2)	1.5-3 (m=2.2; SD=0.5)
Substrate	Sandy soil loam with deep detritus layer. Gravel limited to rifle zone within construction footprint	Sandy soil loam with deep detritus layer.	Sandy soil loam with deep detritus layer.	Sandy soil loam with deep detritus layer. Riffle has some gravel.	Sandy soil loam with deep detritus layer.

Attributes/Site	Entire Site	Below	Partially	Construction	Above
Types of Emergent Aquatic Vegetation	Water Lilly (<i>Nymphaea spp</i>), Knotweed (<i>Persicaria spp</i>) and Common Spikerush (<i>Eleocharis sphacelata</i>)	water Lilly (<i>Nymphaea spp</i>)	Knotweed (<i>Persicaria spp</i>) and Water Lilly (<i>Nymphaea spp</i>) + <i>Cyperus spp</i>	Knotweed (<i>Persicaria spp</i>) and Water Lilly (<i>Nymphaea spp</i>) + <i>Cyperus spp</i>	Water Lilly (<i>Nymphaea spp</i>), Knotweed (<i>Persicaria spp</i>) and Common Spikerush (<i>Eleocharis sphacelata</i>)
Non-native Fish					
Mosquito Fish (<i>Gambusia holbrooki</i>)	Present	Present	Present	Present	Present
Water Quality					
Heavy Metals					
Arsenic	0.104 (0.0005)	0.002 (0)	0.0017 (0)	0.0017 (0)	0.0015 (0.0005)
Cadmium	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Chromium	<0.001	<0.001	<0.001	<0.001	<0.001
Copper	<0.001	<0.001	<0.001	<0.001	<0.001
Lead	<0.001	<0.001	<0.001	<0.001	<0.001
Mercury	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
Nickel	<0.001	<0.001	<0.001	<0.001	<0.001
Zinc	<0.005	<0.005	<0.005	<0.005	<0.005
Hydrocarbons					
Naphthalene	<0.02	<0.02	<0.02	<0.02	<0.02
TRH>C10-C16	<0.05	<0.05	<0.05	<0.05	<0.05
TRH>C10-C16 less Naphthalene (F2)	<0.05	<0.05	<0.05	<0.05	<0.05
TRH>C16-C34	0.82 (0.3)	0.15 (0.1)	0.15 (0.1)	0.15 (0.1)	0.15 (0.1)
TRH>34-C40	<0.1	<0.1	<0.1	<0.1	<0.1
TRH C6-C10	<0.02	<0.02	<0.02	<0.02	<0.02
TRH C6-C10 LESS BTEX (F1)	<0.02	<0.02	<0.02	<0.02	<0.02
BTEX group					
Benzene	<0.001	<0.001	<0.001	<0.001	<0.001
Ethylbenzene	<0.001	<0.001	<0.001	<0.001	<0.001
m&p-Xylenes	<0.002	<0.002	<0.002	<0.002	<0.002
o-Xylene	<0.001	<0.001	<0.001	<0.001	<0.001
Toluene	<0.001	<0.001	<0.001	<0.001	<0.001
Xylenes – total	<0.003	<0.003	<0.003	<0.003	<0.003
Nutrients					

Attributes/Site	Entire Site	Below	Partially	Construction	Above
Nitrogen (as N)	0.53 (0.13)	0.51 (0.14)	0.51 (0.14)	0.51 (0.15)	0.539 (0.11)
Suspended Solids	7.52 (5.57)	7.18 (0.78)	7.25 (0.75)	7.25 (0.87)	11.61 (8.45)
Total Phosphorus	0.029 (0.016)	0.03 (0.01)	0.03 (0.01)	0.03 (0.01)	0.041 (0.013)
Field Physio-chemical Data					
Dissolved Oxygen	2.42 (1.12)	2.26 (0.29)	2.28 (0.29)	2.28 (0.32)	2.55 (1.56)
Conductivity	228.23 (13.83)	227.38 (15.24)	226.57 (14.75)	226.57 (16.16)	229.90 (12.58)
pH	6.50 (0.29)	6.58 (0.14)	6.57 (0.14)	6.57 (0.15)	6.42 (0.37)
Temperature	24.38 (0.34)	24.32 (0.30)	24.30 (0.29)	24.30 (0.32)	24.46 (0.37)
Turbidity	8.80 (4.38)	9.20 (3.91)	9.13 (3.70)	9.13 (4.05)	8.46 (4.97)

DRAFT

4.0 DISCUSSION

Pre-construction baseline monitoring has shown that Giant Barred Frogs continue to inhabit Upper Warrell Creek within and adjacent to the proposed construction footprint. Monitoring surveys performed during spring 2013 confirmed the population breeds at this location and subsequent surveys confirm at least two cohorts of offspring were present during the summer survey with both juvenile and sub adult frogs present. The autumn sampling found only sub adult frogs present in the population and based on recorded growth rates of juvenile frogs it is likely that tadpoles began metamorphosis between late spring through to about late December.

The influence of environmental variables on the recording rate of frogs is thought to be more influenced by a combination of both rainfall leading up to the survey and the prevailing temperature at the time of the survey. Both the summer and autumn surveys were performed at a time when the site had received more than 30 mm of rainfall in the past 7 days and combined with mild temperatures this contributed to the capture of relatively high numbers of frogs. It is thought that temperature tends to have less of an influence provided an adequate amount of rainfall has fallen at the site within 7 days of performing the survey (B. Lewis unpub data). This is exemplified by the cold summer night in January when temperatures were just 14.5°C at 0245 hours but frogs were still active and above the leaf litter. In contrast, the spring survey was performed during a period of reduced rainfall which resulted in far fewer frogs being detected during similar mild temperatures. Surveys performed around the same time between Port Macquarie and Kempsey tended not to show this marked affect with the number of captured frogs being similar between spring and summer and individuals, particularly female adults being active at temperatures down to around 10°C (Lewis 2014).

The study performed by Koch and Hero (2007) on the Giant Barred Frog suggested greater survey efficiencies when temperatures were above 18°C, but individuals in different age-sex classes can have different responses and other factors also influence the activity of individual frogs on any given night. Similar studies have also found that environmental variables alone did not explain the majority of the variation in amphibian density (Salvador and Carrascal 1990; Ovaska 1991; Fukuyama *et al.* 1998; Brown and Shine 2002). These additional factors, which influence how easily they are detected, may include differences in the actual density and behaviour between the different age-sex classes. To address this, the Warrell Creek to Urunga Giant Barred Frog management strategy proposed monitoring to occur at three seasonal time scales within the recognised period of activity (September-May) and that monitoring must take into account other environmental variables of which we have identified rainfall (>10 mm in past 7 days).

The population estimate has shown there is likely to be around 43 (± 26.6) adults present along the 1 km transect. Given the recorded sex ratio, the population is likely to comprise an equal number of male and females. If we use this as the baseline data set for determining a decline in the population then we must remember that the results from these surveys are only a snap shot in time and reflects a sample from the population during a period of below average rainfall and where no breeding is likely to have taken place in the 2013/14 season given no flood events occurred. In contrast, the previous 2012/13 season produced several flood events which enabled frogs to breed at this site and as a result the population size estimated here (43 ± 26.6 adults) may be slightly higher than normal.

The capture data showed a relatively continuous distribution of frogs across the 20 zones with this peaking within the construction footprint of Zone 8. Although most of the frogs from this zone were recorded from a back channel area which lies adjacent to the bridge and associated earthworks for the service road, it highlights the importance of temporary frog fencing during the construction of the project. The fact that frogs are seldom more than 10 m from the edge of the stream indicates they are less likely to access the service road. This may increase in response to increased planting of vegetation to improve habitat quality in the area as frogs were up to 22 m from the water's edge in areas where the forest was undisturbed. The distance frogs tend to be from the water edge is often linked to the amount of prevailing rainfall with frogs moving further from the stream edge during flood events but this does not normally exceed 50 m (Streatfeild 1999; Lemckert and Brassil 2000). The recorded frog movements during this baseline survey show that frogs move within and out of the proposed construction zone with individuals moving up to three zones within a relatively short amount of time (i.e. ~70 days). These movements imply that any prescribed relocation of individuals over relatively small distances (i.e. <60 m) is likely to result in individuals still remaining within their maternal home range. This is consistent with the findings of Lemckert and Brassil (2000) who reported nightly movements from 0 m to over 100 m, but all were within a 20 m wide band either side of the stream.

No tadpoles were recorded during the field sampling of the survey zones (1-20) with sampling being conducted in spring and again in autumn. This has been attributed to the difficulties of sampling the site where the open areas are often deep (>1.5 m) and inaccessible or contain dense aquatic vegetation in the shallower reaches (i.e. <1 m). Therefore, tadpole sampling may be of limited value. Based on the presence of multiple juvenile frogs around the back channels which support dense aquatic vegetation these areas are likely to be important for tadpoles to avoid predation. Given the proximity of one of these areas on the southern bank of Zone 8 it will be important for site controls such as temporary frog fencing to be installed and rigorously maintained.

The detection of Chytrid fungus from one frog in only one of the replicates indicates that Chytrid may be present in the population. This finding is consistent with sampling from some other populations between Port Macquarie and Kempsey performed at the same time (Lewis 2014). The management of Chytrid at this location will help to prevent the inadvertent spread of it to other locations along the construction corridor.

The habitat data showed no consistent pattern with the capture of frogs with both adults, sub adults and juveniles being captured across the broad land use and forest types. What is clear from the habitat data is that the southern bank is comprised of either cleared land or disturbed riparian rainforest as a result of ongoing cattle grazing and any impacts associated with the new bridge could easily be offset with the exclusion of cattle to reduce disturbance of leaf litter fall and some assisted rehabilitation of the creek banks riparian vegetation. The effects of agriculture are also evident in the water quality data with total nitrogen being recorded at elevated levels during the sampling period.

5.0 RECOMMENDATIONS

Based on the results obtained during the survey the following recommendations have been proposed:

- 1) RMS explore opportunities to improve habitat condition during the early stages of the project. This could include:
 - a. The removal of livestock from those areas now owned by the RMS;
 - b. Following the removal of livestock some assisted planting of locally occurring native riparian trees (i.e. Overstorey and mid stratum type species including Water Gum *Tristaniopsis laurina*, Waterhousea *Waterhousea floribunda*, Lillypilly *Acmena smithii* and some sparse groundcover plants including Matrush *Lomandra longifolia*) be undertaken. Performing this task early on will allow some measurable gain of habitat condition during the Giant Barred Frog monitoring period.
- 2) Water quality data continue to be collected at SW1 (Browns Crossing Road Bridge) and SW2 (Zone 8) ensuring sampling is undertaken during both dry and wet weather events.
- 3) Temporary frog fencing is installed at least 5 days before construction works take place within 50 m of the creek and a series of pre-clearance survey performed in accordance with the approved Giant Barred Frog management strategy to ensure no frogs remain within the construction footprint.

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ENVIRONMENTAL

7.0 APPENDIX – FIELD SURVEY DATA

Table A1. Rainfall data between June 2013 and April 2014 from weather station 059018 (Macksville Country Club). Source: www.bom.nsw.gov.au. Green shading = rainfall trigger event; Red shading = field survey date; Blue shading is water quality monitoring data. Days refer to cumulative data collected over longer periods.

Date/Month	June	July	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr
1st	0	6.6	0	0	0	0	0	0	0	17.6	3.2
2nd	↓	8	0	0	0	0	4.4	0	0	17.2	0
3rd	3.02 days	1.2	0	0	0	0	0	0	0	6.2	0
4th	0	0	0	0	0	0	0	0	0	0	0
5th	0	0	0	0	0	0	2.2	0	15.2	0	0
6th	0	0	0	0	0	0	0	0	1.4	0	0
7th	0	0	0	0	0	0	0	0	0	0	33
8th	0	0	0	0	0	0	0	9.8	0	0.6	0
9th	0	0	0	0	0	0	0	0.6	0	0	0
10th	0	0	0	0	0	↓	2	7.6	0	0	0
11th	3.6	1	0	0	0	31.72 days	0	0.8	0	14.2	0
12th	1	1.6	0	0	0	34.2	0	0	0	0	0
13th	14.8	0	0	0	0	18.2	0	0	1.8	0	0
14th	0	0	0	4.4	0	5	↓	5	9.8	0	0
15th	0	0	0	0	0	0	↓	1.8	0	0	0
16th	0	0	0	1.1	0	↓	5.03 days	0	0	0	0
17th	0	0	0	10.8	0	↓	0.8	0	48	8.2	0
18th	0	0	0	2	1.8	10.03 days	0	0	0.4	0	0
19th	0	0	0	0	22.4	3.2	0	0	0	0	0
20th	0	↓	0	0	0	0	0	0	0.4	0	0
21st	0	↓	0	0	0	0	0	0	42.8	7.8	0
22nd	0	15.03 days	0	0	0	0	0	22.6	0	0	0
23rd	0	0	0	0	0	8.6	0	8.6	0	0	0
24th	0	0	0	0	0	13.6	0	0	0	0	0
25th	0	0	0	0	0	0	↓	0	6.6	9.4	0
26th	0	0	0	0	0	31.2	↓	0	0	0	↓
27th	↓	0	0	0	0	0	15.43 days	0	0	0	↓
28th	33.42 days	↓	0	0	0	0	0	0	0	20.6	22.03 days
29th	↓	1.22 days	0	0	0	0	0	0	0	0	0
30th	11.12 days	0	0	0	3.4	32.6	0	0	0	7.2	0
31st		0	0		0		0	0		0	
Highest Daily	14.8	8	0	10.8	22.4	34.2	4.4	22.6	48	20.6	33
Monthly Total	66.9	34.6	0	18.3	27.6	188.3	29.8	56.8	126.4	109	58.2

Table A2. Minimum air temperature data between June 2013 and April 2014 from weather station 059017 (Wide Street Kempsey 35 km to the south). Days refer to cumulative **data** collected over longer periods.

Source: www.bom.nsw.gov.au. Red shading = field survey date

Day/Month	June	July	Aug	Sep	Oct	Nov	Dec	Jan	Feb	Mar	Apr
1st	10.8	13	13	9.9	12.5	10.5	13.6	17.4	14.7	19.8	18.5
2nd	14	14	6.4	9.6	15.6	12.4	13.3	20.8	16.4	19.1	18.4
3rd	13.4	8.8	6.2	10.5	11.3	14.3	12.6	21.7	15.9	18.4	17.6
4th	7.7	7.5	4.5	8.9	11	14.2	12.8	22.1	16	17.8	16.8
5th	8.9	7	4.2	7.4	8	11.8	19	20.7		16.7	16.9
6th	11.1	5	3.4	6.9	7.6	10.5	12.3	18.4	16.4	16.3	19.3
7th	13.1	2.5	8		11	10.6	10.4	18.5	16.7	15.6	17.8
8th	12.5	4	6.2	9.62 days	13.2	13.3	11.2	16.9	14.2	16.5	15.7
9th	11.3	4.8	11.8	17.9	8.9	14.9	14.7	17.7	16	16.2	16.4
10th	10.8	8.7	7.4	12.5	9.4	19.4	21	17.6	15.6	17.9	17
11th	9.5	9.4	6.8	12.3	13	16.5	16.9	18	16.1	16.6	16.5
12th	12.7	7	8.9	9.6	17.4	17.4	19.2	16.5	17.4	16.1	17.4
13th	12.2	7.3	5.1	11	15.9	15.1	17.7	17.4	21.9	16.9	14.5
14th	11.2	7.8	5.9	11.5	17.8	16.2	18	17.6	20.2	17	16.3
15th	5.5	8	5	11.7	8.2	16.3	18.2	16.9	20.2	16	14.6
16th	6.6	10	3.5	15.2	8.6	15.3	19	17.5	21.5	17.6	13.8
17th	6.5	9.3	6.7	13.4	10.6	13.2	16	18.3	19.7	15.3	12.7
18th	5	9	4.6	9.6	14.1	13.2	14.9	17	20	15.2	12
19th	4.4	10.3	3	8.9	11.4	14.5	15.6	16.6	20.6	15.7	11.5
20th	8.9	13.9	8.2	8.5	13.2	14.8	14.2	17.7	23	19.3	13.5
21st	8.4	4.5	0.6	6.7	14	14.6	15.8	20.9	22.1	19.4	13.2
22nd	6.6	8.3	1.4	8	14.5	18	16.7	22.5	20.5	16.4	14.8
23rd	5.9	2.8	1.8	9.5	14.7	18.5	18	20.6	19.7	17.3	15.2
24th	7.5	4	6.7	12	19.9	15.9	20.2	18.9	16.2	16.9	13.6
25th	3	12	7.9	11.8	13	14.7	19.2	20.6	17.9	17.2	12.5
26th	5.3	7.6	6.4	11	11.4	14	20.6	18.6	16.2	17	12.9
27th	10.7	6.7	10	7.6	11.6	13.8	17.2	16.8	18.5	19.7	17.2
28th	12.5	8.2	7.9	10.7	13.5	11.9	19.9	17.9	20.2	18.5	16.3
29th	11.2	9.7	9	7.7	14.5	16.6	17.8	15.9		17	13
30th	11	11.5	11	9.2	16.1	16.4	15.2	15.4		18.5	12.8
31st		9.1	12		9.2		15.8	15.6		18.9	
Highest daily	14	14	13	17.9	19.9	19.4	21	22.5	23	19.8	19.3
Lowest daily	3	2.5	0.6	6.7	7.6	10.5	10.4	15.4	14.2	15.2	11.5

Table A3. Raw data for the frog surveys during each survey period.
 Bold type denote recaptured frog.

Survey Period	Date	Sex	Age Class	Reproductive Status	Length	Weight	Pit Tag Code	Zone	Amended Zone	Relevance to Construction Footprint	Side of Creek	Distance to water	Activity	Microhabitat	Chytrid Swab	Notes
Spring	20.9.2013	Unknown	Sub Adult		50.7	19.0	735ADA8	C3	10	Partially Within	North	1.2	Observed	Above Litter	No	Captured around 10 m from the original capture site in Lewis 2012.
Summer	29.1.2014	Unknown	Sub adult	-	41.4	9.5	7356782	C1	8	Construction Footprint	SOUTH	2.5	Observed	Above Litter	Yes	
Summer	29.1.2014	Female	Adult	Not Gravid	70.3	46	7352C35	C1	8	Construction Footprint	NORTH	10	Observed	Above Litter	Yes	
Summer	29.1.2014	Female	Adult	Not Gravid	67.9	45.5	7359051	C1	8	Construction Footprint	NORTH	0.1	Observed	Partially buried UL	Yes	
Summer	29.1.2014	Unknown	Sub adult	-	41.3	11	735D187	C2	12	Construction Footprint	NORTH	3.5	Observed	Above Litter	No	
Summer	29.1.2014	Unknown	Juvenile	-	31.8	5	7354569	D2	2	Downstream	SOUTH	0.1	Observed	On dirt	No	
Summer	29.1.2014	Male	Adult	no data	no data	no data	no data	D3	3	Downstream	SOUTH	1	Heard	Under Litter	No	Frog could not be captured
Summer	29.1.2014	Male	Adult	no data	no data	no data	no data	D4	4	Downstream	NORTH	2	Heard	Under Litter	No	Frog could not be captured
Summer	29.1.2014	Male	Adult	no data	no data	no data	no data	D4	4	Downstream	NORTH	4	Heard	Under Litter	No	Frog could not be captured
Summer	29.1.2014	Unknown	Juvenile	-	35.1	6.75	735ABA3	D8	8	Construction Footprint	SOUTH	3	Observed	Above Litter	Yes	
Summer	29.1.2014	Unknown	Juvenile	-	34.7	6.75	735C8FA	D8	8	Construction Footprint	SOUTH	0.3	Observed	Above Litter	Yes	
Summer	29.1.2014	Unknown	Juvenile	-	37.3	9.75	7358816	D8	8	Construction Footprint	SOUTH	0.1	Observed	Above Litter	Yes	
Summer	29.1.2014	Unknown	Juvenile	-	36.3	8	735B63D	D8	8	Construction Footprint	SOUTH	0.1	Observed	On dirt	Yes	
Summer	29.1.2014	Unknown	Juvenile	-	39.7	10	7358320	D8	8	Construction Footprint	NORTH	1.5	Observed	Above Litter	No	
Summer	29.1.2014	Male	Adult	Dark	69	44.5	7357C02	D8	8	Construction Footprint	SOUTH	1.2	Observed	Above Litter	yes	Associated with back channel where several juvenile frogs
Summer	29.1.2014	Male	Adult	Dark	71.8	51.75	7357E40	D8	8	Construction Footprint	SOUTH	1.2	Observed	On Grass	Yes	
Summer	29.1.2014	Female	Adult	Not Gravid	90.5	132	7358A4D	D8	8	Construction	SOUTH	1.4	Observed	On Grass	Yes	

Survey Period	Date	Sex	Age Class	Reproductive Status	Length	Weight	Pit Tag Code	Zone	Amended Zone	Relevance to Construction Footprint	Side of Creek	Distance to water	Activity	Microhabitat	Chytrid Swab	Notes
										Footprint						
Summer	29.1.2014	Female	Adult	Not Gravid	85.6	97.5	735AFF6	D8	8	Construction Footprint	SOUTH	1	Observed	Above Litter	Yes	
Summer	29.1.2014	Unknown	Juvenile	-	29.9	5.5	TOO SMALL	U1	13	Upstream	NORTH	2	Observed	On dirt	No	22 m from main channel but using side creek where 2 m from water
Summer	29.1.2014	Female	Adult	Not Gravid	79.3	64	73542F8	U1	13	Upstream	NORTH	22	Observed	Above Litter	Yes	
Summer	29.1.2014	Unknown	Juvenile	-	37	7.25	735339E	U5	16	Upstream	SOUTH	0.5	Observed	On Flood Debris	Yes	On bank associated with back channel
Summer	29.1.2014	Male	Adult	Dark	66.9	36.5	735B207	U5	16	Upstream	SOUTH	0.1	Observed	On dirt	Yes	On bare bank area at waters edge
Summer	29.1.2014	Male	Adult	Light	71.2	48.25	735BEA5	U6	17	Upstream	SOUTH	17	Observed	Above Litter	Yes	On Bank of back channel which is dry except for one small pond surrounded with Persicaria
Summer	29.1.2014	Male	Adult	Moderate	71.5	50.5	7352E8E	U7	19	Upstream	SOUTH	13	Observed	On Pasture Grass	Yes	On Bank of back channel which is dry except for one small pond surrounded with Persicaria
Summer	29.1.2014	Unknown	Sub adult	-	41	10.5	73542E6	U9	20	Upstream	NORTH	4	Observed	on dirt	No	
Autumn	2.4.2014	Male	Adult	Moderate	71	52	7352A6F	C1	8	Construction Footprint	NORTH	3	Observed	Above Litter	No	
Autumn	2.4.2014	Unknown	Sub Adult		49.8	17.5	735C8FA	C1	8	Construction Footprint	SOUTH	10	Observed	Above litter at base of tree	No	Recapture
Autumn	2.4.2014	Unknown	Sub Adult		47.7	16.25	7359E81	C2	9	Construction Footprint	NORTH	4	Observed	On Dirt	No	
Autumn	2.4.2014	Unknown	Sub Adult		48.5	15.25	7356782	C3	10	Partially Within	SOUTH	7.5	Observed	On dirt	No	Recapture
Autumn	2.4.2014	Female	Adult		67.5	78.8	7352C35	C3	13	Partially Within	NORTH	9	Observed	Above litter at base of tree	No	
Autumn	2.4.2014	Unknown	Sub Adult		44.7	13.25	735746C	D4	4	Downstream	SOUTH	3.5	Observed	Above litter	No	

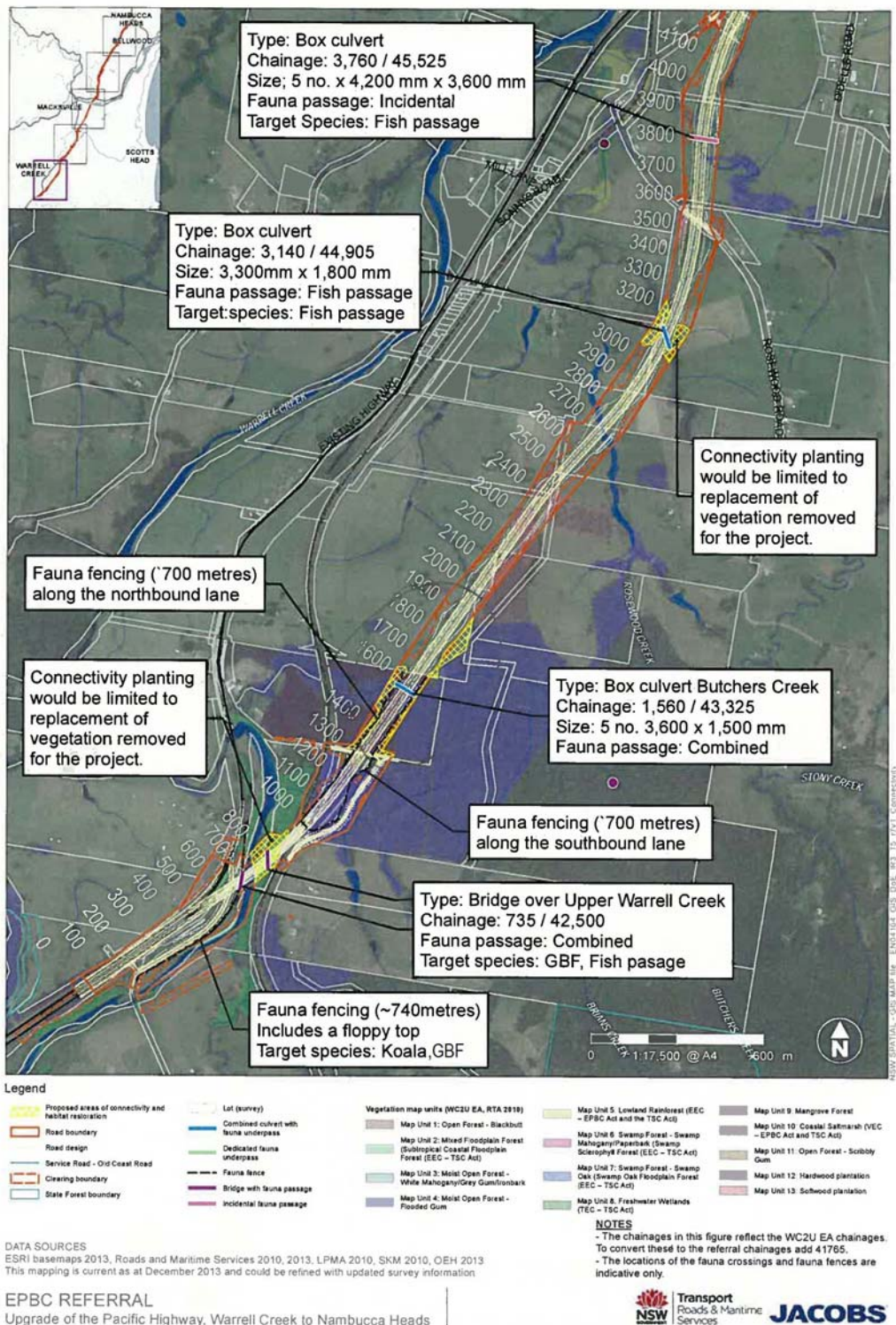
Survey Period	Date	Sex	Age Class	Reproductive Status	Length	Weight	Pit Tag Code	Zone	Amended Zone	Relevance to Construction Footprint	Side of Creek	Distance to water	Activity	Microhabitat	Chytrid Swab	Notes
Autumn	2.4.2014	Unknown	Sub Adult		49.6	17	7354CF6	D4	4	Downstream	NORTH	5	Observed	Above litter	No	
Autumn	2.4.2014	Male	Adult	Light	70.4	44	7353126	D5	5	Downstream	SOUTH	4	Observed	Above litter	No	Male wrestling with Frog 735746C
Autumn	2.4.2014	Male	Adult	Moderate	74.1	52.25	7357C02	D5	5	Downstream	SOUTH	4	Observed	Above litter	No	Recapture. Frog wrestling with Frog 7353126
Autumn	2.4.2014	Female	Adult		91.3	109	73535FD	D5	5	Downstream	SOUTH	8	Observed	Above litter	No	
Autumn	2.4.2014	Female	Adult		83.2	81	73586B8	D5	5	Downstream	SOUTH	5	Observed	Above litter at base of tree	No	
Autumn	2.4.2014	Male	Adult	Moderate	70.7	47.5	7355BE6	D6	6	Partially Within	NORTH	2.2	Observed	On Dirt	No	
Autumn	2.4.2014	Unknown	Sub Adult		50.2	17.5	735B10D	D6	6	Partially Within	SOUTH	11.5	Observed	Above litter	No	Possibly shed PIT tag
Autumn	2.4.2014	Male	Adult	Moderate	68.5	45	735A444	D7	7	Partially Within	SOUTH	1.5	Observed	Above litter	No	
Autumn	2.4.2014	Female	Adult		86.6	91	7359D06	D7	7	Partially Within	SOUTH	8	Observed	Above litter	No	
Autumn	2.4.2014	Male	Adult	Dark	73.5	54	7357E40	D8	8	Construction Footprint	SOUTH	3	Observed	Above litter	No	Recapture
Autumn	2.4.2014	Male	Adult	Moderate	73.4	57	735BE2B	D8	8	Construction Footprint	SOUTH	3	Observed	Above litter	No	
Autumn	2.4.2014	Female	Adult		84.8	90.5	735AFF6	D8	8	Construction Footprint	SOUTH	7.5	Observed	Above litter	No	Recapture
Autumn	2.4.2014	Female	Adult		85.5	87	735C651	D8	8	Construction Footprint	SOUTH	12	Observed	Above litter	No	
Autumn	2.4.2014	Female	Adult		81.7	69	73530F1	D8	8	Construction Footprint	NORTH	13	Observed	On Grass	No	
Autumn	2.4.2014	Male	Adult	Moderate	68.9	42.75	7353B68	U1	11	Upstream	NORTH	3	Observed	Above Litter	No	
Autumn	2.4.2014	Female	Adult		nd	na	ND	U9	20	Upstream	NORTH	2	Observed	On Grass	No	Escaped-avoided capture George!

Table A4. Results of Chytrid testing performed on a subset of individuals captured during the summer monitoring survey.

Frog Number	Date	Species	Animal number	Sex	Rep 1	Rep 2	Rep 3	Mean calculated concentration
1	29/01/2014	<i>Mixophyes iteratus</i>	07358A4D	Female	0	0	0	0
2	29/01/2014	<i>Mixophyes iteratus</i>	07359051	Female	0	0.058	0	0
3	29/01/2014	<i>Mixophyes iteratus</i>	0735AFF6	Female	0	0	0	0
4	29/01/2014	<i>Mixophyes iteratus</i>	073542F8	Female	0	0	0	0
5	29/01/2014	<i>Mixophyes iteratus</i>	0735ABA3	Juvenile	0	0	0	0
6	29/01/2014	<i>Mixophyes iteratus</i>	07358816	Juvenile	0	0	0	0
7	29/01/2014	<i>Mixophyes iteratus</i>	0735C8FA	Juvenile	0	0	0	0
8	29/01/2014	<i>Mixophyes iteratus</i>	0735339E	Juvenile	0	0	0	0
9	29/01/2014	<i>Mixophyes iteratus</i>	0735BEA5	Male	0	0	0	0
10	29/01/2014	<i>Mixophyes iteratus</i>	07352E8E	Male	0	0	0	0
11	29/01/2014	<i>Mixophyes iteratus</i>	07357E40	Male	0	0	0	0
12	29/01/2014	<i>Mixophyes iteratus</i>	07352C35	Male	0	0	0	0
13	29/01/2014	<i>Mixophyes iteratus</i>	07357C02	Male	0	0	0	0
14	29/01/2014	<i>Mixophyes iteratus</i>	07356782	Sub Adult	0	0	0	0
15	29/01/2014	<i>Mixophyes iteratus</i>	0735B63D	Juvenile	0	0	0	0
16	29/01/2014	<i>Mixophyes iteratus</i>	07354569	Juvenile	0	0	0	0
17	29/01/2014	<i>Mixophyes iteratus</i>	0735B207	Male	0	0	0	0

13.0 Appendix C – Fauna Connectivity and Habitat Restoration

Attachment B Figure 1 | Fauna connectivity and habitat restoration



14.0 Appendix D – Weed and Pathogen Plan (AFJV)



See next page.



1.1 Frog Hygiene Protocols

Frog hygiene protocols aim to prevent the spread of amphibian chytrid fungus during the Warrell Creek to Nambucca Heads Pacific Highway Upgrade Project. Indications of this pathogen have to date not been detected within the local frog population. As the pathogen typically exists within water bodies, topsoil and the upper soil profile, this protocol focuses on controlling the potential spread of this pathogen during the “high risk stage” which is defined as being when in contact with the existing natural ground surface within the Giant Barred Frog and Green Thighed Frog hygiene management areas (as defined in Map 1 to Map 3 of this protocol).

1.1.1 Wash Down

- Wash down procedures for vehicles, plant and footwear are to be implemented when entering / exiting the frog hygiene management area (refer to Map 1 to Map 3) at any time when these items have been in contact with the existing natural ground surface. Once topsoil and vegetative material has been removed from the designated frog hygiene management zone, new plant and equipment entering the zone would not require wash-down whereas plant and equipment leaving the zone and having had contact with the natural ground surface will still require wash-down.
- Wash down bays will be implemented at appropriate entry / exit points.
- Wash down bays will incorporate an area for site personnel to disinfect boots when entering / leaving sterile zones during clearing / grubbing and stripping of topsoil.
- Wash down bays will be situated at least 100 m from waterways.
- Wash down areas will be contained with wash-down material (liquid and sediment) to be removed off site to a licensed waste facility.
- All construction personnel must be made aware of the requirements for wash down with this procedure to be a hold point for works commencing.
- Disinfection will be via the use of proprietary available Chloramine and Chlorhexidine based fungicides, cleaning products containing benzalkonium chloride or bleach and alcohol (ethanol or methanol).
- 70% isopropyl wipes may be suitable for the disinfection of small equipment.

1.1.2 Excavated Topsoil

- Excavated topsoil from the frog hygiene management zone must be either reused within the same creek catchment or buried on site.
- If the material is to be stockpiled and reused at a later date, the origin of this material must be tracked and wash-down procedures implemented when reuse occurs.

1.1.3 Entry into GBF / GTF Habitat (outside the Project Site)

- A “permit to enter” system will be established to regulate entry of personnel into areas of GBF / GTF habitat occurring outside of the Project Site.
- Any entry into areas of GBF / GTF habitat (outside the Project Site) will require personnel to disinfect boots before / after entering such areas. Portable spray packs with appropriate disinfectant (refer to **Appendix A**) will be made available at wash down bays.
- All personnel will be made aware of their responsibilities relating to Chytrid management on the site.

1.1.4 Vehicle Movements

- Vehicle movements will be restricted to designated tracks, trails and parking areas by a specific Vehicle Movement Plan (VMP) which will apply at all times throughout the works.
- Vehicle movements within the frog hygiene management areas will be kept to a minimum during excessively wet or muddy conditions.
- Designated parking and turn-around points must be provided on hard well-drained surfaces within the frog hygiene management zone.

1.1.5 Frog Handling

The Project Ecologist and personnel licensed / authorised to handle GBF / GTF are to adhere to the following hygiene protocols in accordance with the *Hygiene Protocols for the Control of Disease in Frogs* (NPWS, 2008) (refer to **Appendix A**): -

- New gloves / bags will be used for each frog captured;
- Individual bags / containers will be used for each frog held and containers (if reusable will be washed) prior to reuse. Containers will be labelled with the date and location);
- When moving between separate sites during frog surveys, footwear / waders will be thoroughly cleaned and disinfected;
- When moving between separate sites during frog surveys, equipment used (such as callipers, scales etc) will be thoroughly cleaned and disinfected; and
- Vehicle tyres will be washed / disinfected before and after visiting frog sites.
- Vehicle tyres can be disinfected with the aforementioned disinfectants or cleaning products with active ingredient benzalkonium chloride (See **Appendix A**).
- Should a sick frog be identified the project environmental staff are to be notified to ensure that controls remain effective and that staff are reminded of their responsibilities. Manage the sick frog in accordance with the protocol.

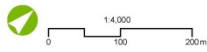
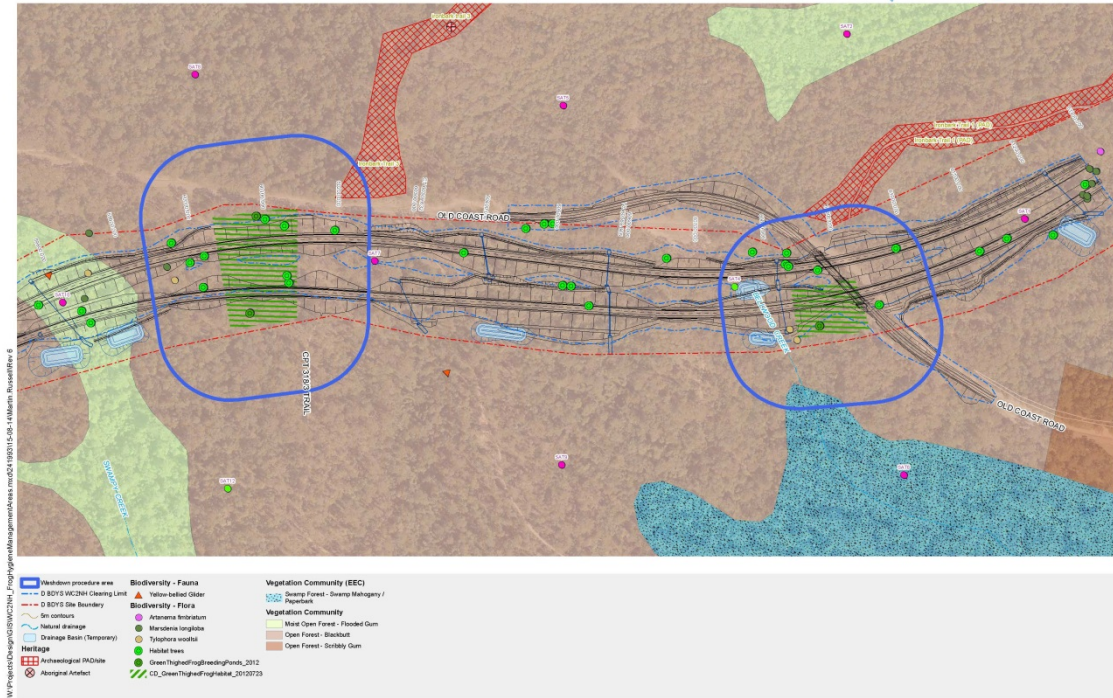
1.2 Frog Hygiene Management Areas

Frog hygiene management areas have been created based on previous ecological assessment and in locations that have been identified as one of the following:

- Green Thighed Frog habitat;
- Likely Green Thighed Frog habitat;
- Giant Barred Frog habitat; and
- Moderately likely Giant Barred Frog habitat.

The locations of the frog hygiene Management Areas are shown in the Frog Hygiene Management Area Maps (Figure 1 to Figure 3). The five locations are all between chainage 42400 and 61000, as identified below:

- Near Swampy Creek and CPT 318/3 Trail. Between chainage 59900 and 60300;
- Adjacent to Bellwood Creek. Between chainage 60700 and 61000;
- Between Teague Ridge Road and Belwood Road. Between chainage 57300 and 59500;
- On the eastern side of Warrell Creek. Between chainage 42400 and 42750; and
- Butchers Creek travels through the site. Between chainage 43200 and 43550.



PACIFIC HIGHWAY UPGRADE WARRELL CREEK TO NAMBUCCA HEADS
 Projection: GDA 1994 MGA Zone 56 Source: RMS, AAD/JV **FIGURE: Frog Hygiene Management Areas (Map 3 of 3)**

**APPENDIX A - HYGIENE PROTOCOLS FOR THE CONTROL OF DISEASE
IN FROGS (NPWS, 2008)**

Threatened Species Management Information Circular No. 6



hygiene protocol for the
control of disease in

frogs

April 2008

Department of **Environment & Climate Change** NSW



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This hygiene protocol is an adaptation of the Declining Amphibian Population Task Force (DAPTF) Fieldwork Code of Practice and the recommendations of Speare et al. (1999) and has drawn on recommendations from earlier guidelines prepared by Environment ACT.

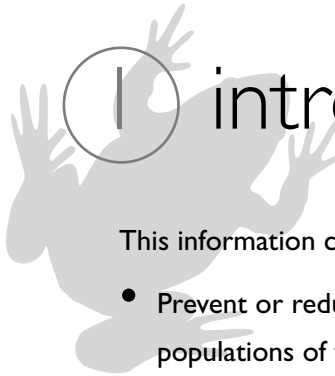
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hygiene protocol for the control of disease in

frogs

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I introduction

This information circular outlines measures to:

- Prevent or reduce disease causing pathogens being transferred within and between wild populations of frogs.
- Ensure captive frogs are not infected prior to release.
- Deal safely with unintentionally transported frogs.
- Assist with the proper identification and management of sick and dead frogs in the wild.

1.1 Who should read this document?

This protocol is intended for use by all researchers, wildlife consultants, fauna surveyors and students undertaking frog field-work. In addition, the protocol should be read by Department of Environment and Climate Change (DECC) personnel, frog keepers, wildlife rescue and carer organisations, herpetological/frog interest groups/societies, fauna park/zoo operators/workers and other individuals who regularly deal with or are likely to encounter frogs.

This protocol outlines the expectations of the DECC regarding precautionary procedures to be employed when working with frog populations. The intention is to promote implementation of hygiene procedures by all individuals working with frogs. New licences and licence renewals will be conditional upon incorporation of the protocol. The DECC recognises that some variation from the protocol may be appropriate for particular research and frog handling activities. Such variation proposals should accompany any licence application or renewal to the DECC.

1.2 Background

1.2.1 Amphibian Chytrid Fungus

The apparent decline of frogs, including extinctions of species and local populations, has attracted increased international and national concern. Many

potential causes for frog declines have been proposed (eg see Pechmann et al., 1991; Ferrero and Bergin, 1993; Pechmann and Wilbur, 1994; Pounds and Crump, 1994; Pounds et al., 1997). However, the patterns of decline at many locations suggest that epidemic disease maybe the cause (Richards et al., 1993; Laurance et al., 1996; Alford and Richards, 1997). Recent research has implicated a water-borne fungal pathogen *Batrachochytrium dendrobatidis* as the likely specific causative agent in many of these declines both in Australia and elsewhere (Berger et al., 1998; 1999). This agent is commonly known as the amphibian or frog chytrid fungus and is responsible for the disease Chytridiomycosis (Berger et al., 1999).

B. dendrobatidis is a form of fungus belonging to the phylum Chytridiomycota. Most species within this phylum occur as free-living saprophytic fungi in water and soil and have been found in almost every type of environment including deserts, arctic tundra and rainforest and are considered important primary biodegraders (Powell 1993). *B. dendrobatidis* is a unique parasitic form of Chytridiomycete fungi, in that it invades the skin of amphibians, including tadpoles, often causing sporadic deaths with up to 100% mortality in some populations. Chytridiomycosis has been detected in over 40 species of native amphibian in Australia (Mahony and Workman 2000). However, it is not currently known whether the fungus is endemic or exotic to Australia.



2 site hygiene management

A checklist of risk management procedures and recommended standard hygiene kit is provided in Appendix I. Please note Footnote I on page 4.

Individuals studying frogs often travel and collect samples of frogs from multiple sites. Some frog populations can be particularly sensitive to the introduction of infectious pathogens such as the frog chytrid fungus. Also, the arrangement of populations in the landscape may make frogs particularly vulnerable to transmission of infectious pathogens. Therefore, it is important that frog workers recognise the boundaries between sites and undertake measures which reduce the likelihood of spreading infection.

Where critically endangered species or populations of particular risk are known to occur, this protocol should be applied over very short distances ie a single site may need to be subdivided and treated as separate sites.

When planning to survey multiple sites, always start at a site where frog chytrid fungus is not known to be present before entering other infected areas.

2.1 Defining a site

Defining the boundary of a site maybe problematic. In some places, the boundary between sites will be obvious but in others, less so. Undertaking work at a number of sites or conducting routine monitoring at a series of sites within walking distance creates obvious difficulties with boundary definitions. It is likely that defining the boundary between sites will differ among localities. It may be that a natural or constructed feature forms a logical indicator of a site boundary eg a road/ track, a large body of water such as a river or the sea, a marked habitat change or a catchment boundary.

As a guiding principle, each individual waterbody should be considered a separate site.

When working along a river or stream or around a wetland or a series of interconnecting ponds it is reasonable, in most instances, to treat such examples as a single site for the purposes of this protocol. Such a case would occur in areas where frogs are known to have free interchange between ponds.

Where a stream consists of a series of distinctive tributaries or sub-catchments or where there is an obvious break or division then they should be treated as separate sites, particularly if there is no known interchange of frogs between sites.

2.2 On-site hygiene

When travelling from site to site it is recommended that the following hygiene precautions be undertaken to minimise the transfer of disease from footwear, equipment and/or vehicles.

Footwear

Footwear must be thoroughly cleaned and disinfected at the commencement of fieldwork and between each sampling site.

This can be achieved by initially scraping boots clear of mud and standing the soles in a disinfecting solution. The remainder of the boot should be rinsed or sprayed with a disinfecting solution that contains *benzalkonium chloride* as the active ingredient. Disinfecting solutions should be prevented from entering any water bodies.

Rubber boots such as 'gum boots' or 'Wellingtons' are recommended because of the ease with which they can be cleaned and disinfected.

Several changes of footwear bagged between sites might be a practical alternative to cleaning.

Equipment

Equipment such as nets, balances, callipers, bags, scalpels, headlamps, torches, wetsuits and waders etc that are used at one site must be cleaned and disinfected before re-use at another site.

Disposable items should be used where possible. Non-disposable equipment should be used only once during a particular field exercise and disinfected later or disinfected at the site between uses using procedures outlined in 2.4 below.

Vehicles

Where necessary, vehicle tyres should be sprayed/flushed with a disinfecting solution in high-risk areas.

Transmission of disease from vehicles is unlikely to be a problem. However, if a vehicle is used to traverse a known frog site, which could result in mud and water being transferred to other bodies of water or frog sites, then wheels and tyres should undergo cleaning and disinfection. This should be carried out at a safe distance from water bodies, so that the disinfecting solution can infiltrate soil rather than run-off into a nearby water body.

Spraying with 'toilet duck' (active ingredient *benzalkonium chloride*) is recommended to disinfect car wheels and tyres.

Cleaning of footwear before getting back into the car will prevent the transfer of pathogens from/to vehicle floor and control pedals.

2.3 Handling of frogs in the field

The spread of pathogenic organisms, such as the frog chytrid fungus, may occur as a result of handling frogs.

Frogs should only be handled when necessary.

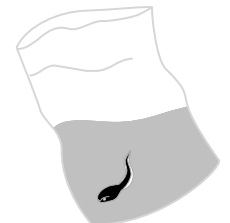
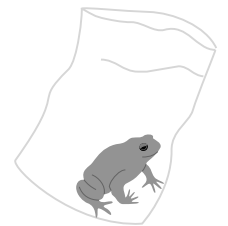
Where handling of frogs is necessary the risk of pathogen transfer should be minimised as follows:

- Hands should be either cleaned and disinfected between samples or a new pair of disposable gloves used for each sample¹. This may be achieved by commencing with a work area that has a dish containing a disinfecting solution and paper towels.
- A 'one bag – one frog' approach to frog handling should be used especially where several people are working together with one person processing frogs and others doing the collecting. Bags should not be reused.
- A 'one bag – one sample' approach to tadpole sampling should be used. Bags should not be reused.

Researchers who use toe clipping or Passive Integrated Transponder (PIT) tagging are likely to increase the risk of transmitting disease between frogs due to the possibility of directly introducing pathogens into the frogs' system. This can be minimised by using:

- Disposable sterile instruments
- Instruments disinfected previously and used once
- Instruments disinfected in between each frog

Disinfecting solutions containing *benzalkonium chloride* are readily available from local supermarkets. Some brands include Toilet Duck, Sanpic, New Clenz and Pine Clean.



¹ As a principle, this protocol assumes that not all frogs in an infected pond will be contaminated by the frog chytrid fungus. The infective load of a body of water may not be high enough to cause cross contamination of individual frogs in the same pond. Therefore care should be taken to use separate gloves and bags and clean hands for each sample, to avoid transmission of high infective loads between individuals.

Open wounds from toe clipping and PIT tagging should be sealed with a cyanoacrylate compound such as *Vetbond*® to reduce the likelihood of entry of pathogens. The DECC ACEC further recommends the application of topical anaesthetic *Xylocaine*® cream and *Betadine*® disinfectant (1% solution) before and after any surgical procedure. This should then be followed by the wound sealant.

All used disinfecting solutions, gloves and other disposable items should be stored in a sharps or other waste container and disposed or sterilised appropriately at the completion of fieldwork. Disinfecting solutions must not come into contact with frogs or be permitted to contaminate any water bodies

2.4 Disinfection Methods

Disinfecting agents for hands and equipment must be effective against bacteria and both the vegetative and spore stages of fungi. The following agents are recommended:

- Chloramine and Chlorhexidine based products such as *Halamid*®, *Halasept*® or *Hexifoam*® are effective against both bacteria and fungi. These products are suitable for use on hands, footwear, instruments and other equipment. The manufacturers instructions should be followed when preparing these solutions.
- Bleach and alcohol (ethanol or methanol), diluted to appropriate concentrations can be effective against bacteria and fungi. However, these substances may be less practical because of their corrosive and hazardous nature.

When using methanol either:

- immerse in 70% methanol for 30 minutes or
- dip in 100% methanol then flame for 10 seconds or boil in water for 10 minutes

Fresh bleach (5% concentration) may be also effective against other frog pathogens such as Rana Virus.

Some equipment not easily disinfected in these ways can be effectively cleaned using medical standard 70% isopropyl alcohol wipes – *Isowipes*®.



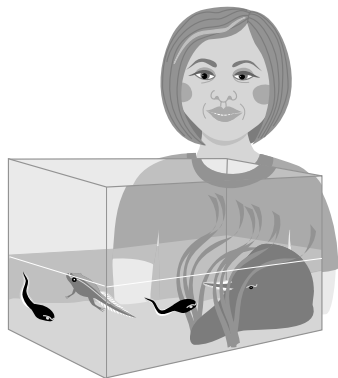
3 captive frog hygiene management

3.1 Housing frogs and tadpoles

Frogs and tadpoles should only be removed from a site when absolutely necessary.

When it is necessary for frogs or tadpoles to be collected and held for a period of time, the following measures should be undertaken:

- Animals obtained at different sites should be kept isolated from each other and from other captive animals.
- Aquaria set up to hold frogs should not share water, equipment or any filtration system. Splashes of water from adjacent enclosures or drops of water on nets may transfer pathogens between enclosures.
- Prior to housing frogs or tadpoles, ensure that tanks, aquaria and any associated equipment are disinfected.
- Tanks and equipment should be cleaned, disinfected and dried immediately after frogs/tadpoles are removed.



Careful maintenance of your enclosures will ensure a safe and hygienic environment for captive frogs and tadpoles.

3.2 Tadpole treatment

In most instances:

Release to the wild of tadpoles held or bred in captivity should be avoided.



When contemplating a release of captive bred tadpoles for conservation purposes a Translocation Proposal should be submitted to the DECC and pathological screening for disease should be undertaken (see also DECC Translocation Policy). Tadpoles can be tested by randomly removing 10 individuals at 6 weeks and again at 2 weeks before anticipated release. Testing could be undertaken by the pathology section at Taronga Zoo, Newcastle University, CSIRO Australian Animal Health Laboratories at Geelong and James Cook University at Townsville. Such an arrangement would need to be negotiated by contacting one of these institutions well before the anticipated release date. (see Appendix 2 for contact details)

DECC have licenced NSW Schools to allow students and/or teachers to remove tadpoles for classroom life cycle studies. They are authorised to remove individuals from only one location, each school also requires endorsement from Department of Education and Training Animal Care and Ethics Committee and comply with this protocol.

Tadpoles collected for these purposes are to be obtained from the local area of the school and are not to be obtained from DECC Reserves. As soon as tadpoles have transformed, froglets must be returned to the exact point of capture. Tadpoles from different locations are not to be mixed.

Antifungal cleansing treatments to clear tadpoles of the frog chytrid fungus are currently being trialed. In the future, such a treatment may be an added procedure required prior to froglet releases.

Detailed information on safely maintaining frogs in captivity is provided in Voigt (2001).

3.3 Frog treatment

The rigour with which frogs must be treated to ensure pathogens are not introduced to native populations means that any proposal for the removal of adult frogs (particularly threatened species) from wild populations should be given careful consideration.

When it is essential for frogs to be removed from the wild, the following should apply.

Individuals to be released should be quarantined for a period of 2 months and monitored for any signs of illness or disease.

Frogs must not be released if any evidence of illness or infection is detected. If illness is suspected, further advice must be sought from a designated frog recipient (Appendix 2) as soon as possible to determine the nature of the problem. Chytridiomycosis can be diagnosed in live frogs by microscopical examination of preserved toe clips or from shedding skin samples. Research is still in progress on the development of a simple technique for the detection of Chytridiomycosis and a treatment for infected frogs.

Current methods which may be used include:

- A technique for the treatment of potentially infected frogs is to place the frogs individually in a 1mg/L benzalkonium chloride solution for 1 hour on days 1, 3, 5, 9, 11 and 13 of the treatment period. Frogs are then isolated/quarantined for two months. This and other possible treatments are documented in Berger and Speare (1998)
- *Betadine*© and *Bactone*© treatments have also been used on adult frogs with some success (M. Mahony, Newcastle University pers. comm.)
- *Itraconazole*© is an expensive drug

which has been used successfully (Lee Berger CSIRO Australian Animal Health Laboratory pers. comm.). Information on this method is available on the Website <http://www.jcu.edu.au/school/PHTM/frogs/adms/attach6.pdf>.

Frogs undergoing treatment should be housed individually and kept separate from non-infected individuals.

3.4 Displaced frogs

Displaced frogs are those native frog species and introduced Cane Toads (*Bufo marinus*) which have been unintentionally transported around the country with fresh produce, transported produce and landscaping supplies. Procedures to be undertaken when encountering introduced/displaced native frog species (as well as Cane Toads) are as follows.

3.4.1 Banana box frogs

'Banana Box' frog is the term used to describe several native frog species (usually *Litoria gracilentata*, *L. infrafrenata*, *L. bicolor* and *L. caerulea*) commonly transported in fruit and vegetable shipments and landscaping supplies. In the past, well meaning individuals have attempted to return these frogs to their place of origin but this is usually impossible to do accurately. There is risk of spread of disease if these frogs are transferred from place to place.

It is strongly recommended that:

Displaced Banana Box frogs should be treated as if they are infected and should not be freighted anywhere for release to the wild unless specifically approved by DECC.

When encountering a displaced frog:

- Contact a licensed wildlife carer organisation to collect the animal. The frog should then undergo a quarantine period of 2 months along with an approved disinfection treatment.
- Post-quarantine, the frog (if one of the species identified above) may be transferred to a licensed frog keeper. All other species require the permission from DECC Wildlife Licensing and Management Unit (WLMU) prior to transfer. Licensed carer groups are to record and receipt frogs obtained and disposed of in this way.
- Licensed Frog Keepers are to list these frogs in their annual licence returns to DECC.

Frogs held by licensed frog keepers are not to be released to the wild except with specific DECC approval.

Displaced frogs may be made available to recognised institutions for research projects, display purposes or perhaps offered to the Australian Museum as scientific specimens once approval has been provided by the DECC WLMU.



Frogs are often unintentionally transported with fresh produce and landscaping supplies. They are collectively known as 'banana box' or displaced frogs.

3.4.2 Cane toads

Cane toads are known carriers of the Frog chytrid fungus and should not be knowingly transported or released to the wild.

If a cane toad is discovered outside of its normal range, it should be humanely euthanased in accordance with the recommended NSW Animal Welfare Advisory Council procedure (see Appendix 3). Care should be taken to avoid euthanasia of native species due to mistaken identity.

3.4.3 Local frog species

Frogs encountered on roads, around dwellings and gardens or in swimming pools should not be considered as displaced frogs.

Frogs encountered in these situations should be assisted off roads, away from dwellings, or out of swimming pools preferably to the nearest area of vegetation or suitable habitat.

Incidences of frogs spawning or tadpoles appearing in swimming pools should be referred to a wildlife carer/rescue organisation for assistance (see Appendix 4).

Contact the Frogwatch Helpline if you are unsure whether a frog is a local species or displaced.

An NPWS information brochure titled 'Cane Toads in NSW' provides further information on cane toads and assistance with identification of some of the commonly misidentified native species. This information is also available on the DECC website.

4 sick or dead frogs

Unless an obvious cause of illness or death is evident (eg predation or road mortality): Sick or dead frogs encountered in the wild should be collected and disposed of in accordance with the procedures described in section 4.2 below.

4.1 Symptoms of sick and dying frogs

Sick and dying frogs exhibit a range of symptoms characteristic of chytrid infection. Symptoms may be expressed in the external appearance or behaviour of the animal. A summary of these symptoms are described below. More detailed information can be found in Berger et al., (1999) or at the James Cook University Amphibian Disease website at: <http://www/jcu.edu.au/school/phtm/PHTM/frogs/ampdis.htm>.



Appearance (one or more symptoms)

- darker or blotchy upper (dorsal) surface
- reddish/pink-tinged lower (ventral) surface and/or legs and/or webbing or toes
- swollen hind limbs
- very thin or emaciated
- skin lesions (sores, lumps)
- infected eyes
- obvious asymmetric appearance

Behaviour (one or more symptoms)

- lethargic limb movements, especially hind limbs
- abnormal behaviour (eg a nocturnal, burrowing or arboreal frog sitting in the open during the day and making no vigorous attempt to escape when approached)
- little or no movement when touched

Great barred frog (*Mixophyes fasciolatus*) with severe Chytrid infection — note lethargic attitude and sloughing skin. Photo: L. Berger

Diagnostic behaviour tests

Sick frogs will fail one or more of the following tests:

test	healthy	sick
Gently touch with finger	Frog will blink	Frog will not blink above the eye
Turn frog on its back	Frog will flip back over	Frog will remain on its back
Hold frog gently by its mouth	Frog will use its forelimbs to try to remove grip	No response from frog

4.2 What to do with sick or dead frogs

A procedure for the preparation and transport of a sick or dead frog is given below². Adherence to this procedure will ensure the animal is maintained in a suitable condition for pathological examination and assist the DECC and researchers to determine the extent of the disease and the number of species affected.


- Disposable gloves should be worn when handling sick or dead frogs. Avoid handling food and touching your mouth or eyes as this could transfer pathogens and toxic skin secretions from some frog species.
- New gloves and a clean plastic bag should be used for each frog specimen to prevent cross-contamination. When gloves are unavailable, use an implement to transfer the frog to a container rather than using bare hands.
- If the frog is dead, keep the specimen cool and preserve as soon as possible (as frogs decompose quickly after death making examination difficult). Specimens can be fixed/preserved in 70% ethanol or 10% buffered formalin.

Cut open the belly and place the frog in about 10 times its own volume of preservative. Alternatively, specimens can be frozen (although this makes tissues unsuitable for some tests). If numerous frogs are collected, some should be preserved and some should be frozen. Portions of a dead frog can be sent for analysis eg a preserved foot, leg or a portion of abdominal skin.

- The container should be labelled showing at least the species, date and location. A standardised collection form is provided in Appendix 5.
- If the frog is alive but unlikely to survive transportation (death appears imminent), euthanase the frog (see Appendix 3) and place the specimen in a freezer. Once frozen, the specimen is ready for shipment to the address provided below.
- If the frog is alive and likely to survive transportation, place the frog into either a moistened cloth bag with some damp leaf litter or into a plastic bag with damp leaf litter and partially inflated before sealing. Remember to keep all frogs separated during transportation.
- Preserved samples can be sent in jars or wrapped in wet cloth, sealed in bags and placed inside a padded box.
- Send frozen samples in an esky with dry ice (available from BOC/CIG Gas outlets).
- Place live or frozen specimens into a small styrafoam esky (available from K-Mart/Big W for approximately \$2.50).
- Seal esky with packaging tape and address to one of the laboratories listed in Appendix 4.
- Send the package by courier.

Further information on sick and dying frogs is available on the Amphibian Disease Home Page at <http://www.jcu.edu.au/dept/PHTM/frogs/ampidis.htm> — in particular refer to 'What to do with dead or ill frogs'.

²The measures described below are standard procedures and may vary slightly depending on the distance and time required to reach the intended recipient. Contact the intended recipient of the sick or dead frog prior to sending to confirm the appropriate procedure.



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appendix I

hygiene protocol checklist and field kit

The following checklist and field kit are designed to assist with minimising the risk of transferring pathogens between frogs.

Have you considered the following questions before handling frogs in the field:

- Has your proposed field trip been sufficiently well planned to consider hygiene issues?
- Have you taken into account boundaries between sites (particularly where endangered species or populations at risk are known to occur)?
- Have footwear disinfection procedures been considered and a strategy adopted?
- Have you planned the equipment you will be using and developed a disinfection strategy?
- Are you are planning to visit sites where vehicle disinfection will be needed (consider both vehicle wheels/tyres and control pedals) and if so, do you have a plan to deal with vehicle disinfection?
- Have handling procedures been planned to minimise the risk of frog to frog pathogen transmission?
- Do you have a planned disinfection procedure to deal with equipment, apparel and direct contact with frogs?

If you answered NO to any of these questions please re-read the relevant section of the DECC Hygiene Protocol for the Control of Disease in Frogs and apply a suitable strategy.

Field hygiene kit

When planning to survey frogs in the field a portable field hygiene kit should be assembled to assist with implementing this protocol. Recommended contents of a field hygiene kit would include:

- Small styrofoam eski
- Disposable gloves
- Disinfectant spray bottle (atomiser spray) and/or wash bottle
- Disinfecting solutions
- Wash bottle
- Scraper or scrubbing brush
- Small bucket
- Plastic bags large and small
- Container for waste disposal
- Materials for dealing with sick and dead frogs (see section 4.2)



appendix 2

Always contact the relevant specialist prior to sending a sick or dead frog. In some cases, only wild frogs will be assessed for disease. Analysis may also attract a small fee per sample.

designated sick and dead frog recipients

Contact one of the following specialists to arrange receipt and analyse sick and dead frogs. Make contact prior to dispatching package:

Karrie Rose
Australian Registry of Wildlife Health
Taronga Conservation Society, Australia
PO Box 20
MOSMAN NSW 2088
Phone: 02 9978 4749
Fax: 02 9978 4516
Krose@zoo.nsw.gov.au

Diana Mendez or
Rick Speare
School of Public Health,
Tropical Medicine and
Rehabilitation Sciences
James Cook University
Douglas Campus
TOWNSVILLE QLD 4811

Phone: 07 4796 1735
Fax: 07 4796 1767
Diana.Mendez@jcu.edu.au
Richard.Speare@jcu.edu.au

Michael Mahony
School of Biological Sciences
University of Newcastle
CALLAGHAN NSW 2308

Phone: 02 4921 6014
Fax: 02 4921 6923
bimjm@cc.newcastle.edu.au

For information on frog keeping licences and approvals to move some species of displaced frog contact:

Co-ordinator, Wildlife Licensing
Wildlife Licensing and Management Unit
DECC
PO Box 1967
Hurstville NSW 1481
Ph 02 9585 6481
Fax 02 9585 6401
wildlife.licensing@environment.nsw.gov.au

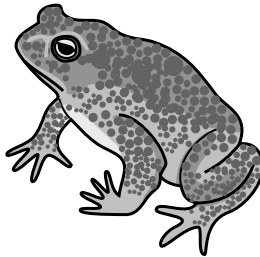
For information on the possible identity of displaced frogs contact:

Frog and Tadpole Society (FATS)
Frogwatch Helpline
Ph: 0419 249 728

appendix 3

NSW Animal Welfare Advisory Council methodology

The NSW Animal Welfare Advisory Council procedure for humanely euthanasing cane toads or terminally ill frogs is stated as follows:



- Using gloves, or some other implement, place cane toad or terminally ill frog into a plastic bag.
- Cool in the refrigerator to 4°C.
- Crush cranium with a swift blow using a blunt instrument.

Note: Before killing any frog presumed to be a cane toad, ensure that it has been correctly identified and if outside the normal range for cane toads in NSW (north coast) that local DECC regional office is informed.

appendix 4

licensed wildlife carer and rescue organisations

Following is a list of wildlife rehabilitation groups licensed by
Department of Environment and Climate Change (NSW):

Northern NSW

Australian Seabird Rescue
For Australian Wildlife Needing Aid
(FAWNA)
Friends of the Koala
Friends of Waterways (Gunnedah)
Great Lakes Wildlife Rescue
Koala Preservation Society of NSW
Northern Rivers Wildlife Carers
Northern Tablelands Wildlife Carers
Tweed Valley Wildlife Carers
Seaworld Australia
WIRES branches in Northern NSW

Southern NSW

Looking After Our Kosciuszko Orphans
(LAOKO)
Native Animal Network Association
Native Animal Rescue Group
Wildcare Queanbeyan
WIRES branches in Southern NSW

Sydney, Hunter and Illawarra

Hunter Koala Preservation Society

Ku-ring-gai Bat Colony Committee
Kangaroo Protection Co-operative
Native Animal Trust Fund
Organisation for the Rescue and Research of
Cetaceans (ORRCA)
Sydney Metropolitan Wildlife Services
Wildlife Aid
Wildlife Animal Rescue and Care (Wildlife
ARC)
Waterfall Springs Wildlife Park
Oceanworld
Wildlife Care Centre, John Moroney
Correctional Centre
Koalas in Care
WIRES branches around Sydney, Hunter and
Illawarra

Western NSW

Rescue and Rehabilitation of Australian
Native Animals (RRANA) Ê
RSPCA Australian Capital Territory Inc.
Wildlife Carers Network (Central West) Ê
WIRES branches in Western NSW Ê
Cudgegong Wildlife Carers Ê

appendix 5 — sick or dead frog collection form

Sender details:

name: _____ address: _____ postcode: _____
phone: (w) _____ (h) _____ fax: _____ email: _____

Collector details: (where different to sender)

name: _____ address: _____ postcode: _____
phone: (w) _____ (h) _____ fax: _____ email: _____

Specimen details:

record no: _____ no. of specimens: _____ species name: _____ date collected: _____
day/month/year

time collected: _____ sex: _____ status at time of collection: _____ date sent: _____
male/female healthy(H)/ sick(S)/ dead(D) day/month/year

location: _____ map grid reference: _____
(easting) (northing)

reason for collection: _____

Batch details for multiple species collection:

species	no.	locality	(AMG)	date	sex	status (H/S/D)

habitat type: _____ vegetation type: _____ micro habitat: _____
eg creek, swamp, forest eg rainforest, sedgeland eg creek bank, under log, amongst emergent vegetation,
on ground in the open

unusual behaviour of sick frogs: _____
eg lethargic, convulsions, sitting in the open during the day, showing little or no movement when touched.

dead frogs appearance: _____
eg thin, reddening of skin on belly and/or toes, red spots, sore, lumps or discolouration on skin

deformed frogs: _____ dead/sick tadpoles: _____
eg limb(s) missing, abnormal shape or length eg numbers/behaviour

unusual appearance of egg masses: _____ recent use of agricultural chemicals in area: _____
eg grey or white eggs eg pesticides, herbicides, fertilisers

other potential causes of sickness/mortality/comments/additional information: _____



**NSW
NATIONAL
PARKS AND
WILDLIFE
SERVICE**

General inquiries: PO Box A290 South Sydney 1232 Ê
Phone: 9995 5000 or 1300 361967 Ê
Fax: 02 9995 5999 **Web site:** www.environment.nsw.gov.au Ê

