



*Environment Protection and Biodiversity Conservation Act 1999.*

## General Permit Application for:

- **Threatened species and ecological communities (section 201)**
- **Migratory species (section 216)**
- **Whales and dolphins (section 238)**
- **Listed marine species (section 258)**



If the person completing this form is representing a small business (i.e. a business having less than 20 employees), please provide an estimate of the time taken to complete this form.

Please include:

- the time taken spent reading the instructions, working on the questions and obtaining the information; and
- the time spent by all employees in collecting and providing this information.

Hours

Minutes

### Purpose of this form

This form is for an activity which will affect any species or ecological community listed under the EPBC Act in the above categories where that activity is within a Commonwealth Area, and for whale/dolphins where the activity is within the waters of the Australian Whale Sanctuary, or internationally.

Complete this form in addition to either Supplementary Form A, B or C described in question 1 on the next page. Please return it, along with the relevant Supplementary Form to the Department of the Environment and Water Resources.

Note that it is a requirement of the *Environment Protection and Biodiversity Conservation Act 1999* that details of this application (which may include the applicant's name) and any supplementary forms (A, B or C) be provided to persons or bodies registered with the Department of the Environment and Water Resources under section 266A of the Act, for the purposes inviting submissions from those persons or bodies regarding permit applications.

### Do not use this form for permits in:

- The Great Barrier Reef Marine Park. These permits are available at: [www.gbrmpa.gov.au/corp\\_site/permits/](http://www.gbrmpa.gov.au/corp_site/permits/)
- A Commonwealth park or reserve (e.g. Kakadu National Park). These permits are available at: [www.environment.gov.au/epbc/permits/parks/](http://www.environment.gov.au/epbc/permits/parks/)

### Additional information

Please ensure that you have read the following information sheet:

*Permits required for activities affecting EPBC Act listed species in Commonwealth Areas including the Australian Whale Sanctuary*

This information sheet is available at <http://www.environment.gov.au/epbc/permits/index.html>. Further information is also available by contacting the Department on phone: (02) 6274 1111 or email: [epbcwild@environment.gov.au](mailto:epbcwild@environment.gov.au).

### Incomplete information

Applications that are incomplete or contain insufficient information cannot be assessed. Delays will occur whilst further information is sought from the applicant.

### If you need more space

If there is insufficient space on this form to fully address any of the questions please attach additional pages and list these attachments at question 10.

1 Which of the following best describes the purpose of this application?

**Research on whales/dolphins**  ⇒ You will also need to complete Supplementary Form A for Whales and Dolphins (cetaceans).  
**Now go to 2**

**To conduct an activity that will have an incidental impact on whales/dolphins**  ⇒ You will also need to complete Supplementary Form A for Whales and Dolphins (cetaceans).  
E.g. whales and dolphins are not the purpose of the activity but they will be indirectly affected  
**Now go to 2**

**Whale and Dolphin watching**  ⇒ You will also need to complete Supplementary Form B for Whale and Dolphin Watching.  
**Now go to 2**

**To kill, injure, take, trade, keep or move a listed species or ecological community in Commonwealth areas**  ⇒ You will also need to complete Supplementary Form C **Listed species / ecological community, listed migratory species or listed marine species.**  
**Now go to 2**

**Import/export of whale/dolphin parts or products**  ⇒ Please contact the Cetacean Research and Policy Section on 02) 6274 1111.

2 Period of permit requested  
*Permits are usually not issued for more than 5 years.*

|                              |                               |
|------------------------------|-------------------------------|
| Start date: <b>June 2007</b> | End date: <b>October 2009</b> |
|------------------------------|-------------------------------|

3 The permit holder can be a group such as a business, company, or corporation?

Is the proposed permit holder a group?

No  ⇒ *Go to next question*

Yes  ⇒ *Give details below*

|                 |
|-----------------|
| Group Name      |
| Street address: |
| Postal address: |
| Telephone No.:  |
| Fax No.:        |
| Email address:  |

**Now go to 5**

4 Is the proposed permit holder an individual?

No  ⇒ *Go to next question*

Yes  ⇒ *Give details below of each individual to whom the permit would be issued.* If insufficient space, attach a separate list.

|          |   |
|----------|---|
| <b>1</b> | Name: Dr. Susan M. Bengtson Nash  |
|          | Residential address: <b>223 Agnew St. Seven Hills QLD 4108</b>          |
|          | Postal address: <b>ENTOX, 39 Kessels Road, Coopers Plains, QLD 4108</b> |
|          | Telephone No.: <b>(07) 3274 9004</b>                                    |
|          | Fax No.: <b>(07) 3274 9003</b>  |
|          | Email address: <b>s.nash@uq.edu.au</b>                                  |

|          |   |
|----------|---|
| <b>2</b> | Name: Dr. Michael Noad  |
|          | Residential address: <b>42 Delville Ave, Moorooka, QLD 4105</b> |
|          | Postal address: <b>As above</b>                                 |
|          | Telephone No.: <b>(07) 3411 7515</b>                            |
|          | Fax No.:  |
|          | Email address: <b>m.noad@uq.edu.au</b>                          |

|          |   |
|----------|---|
| <b>3</b> | Name: Courtney Waugh  |
|          | Residential address: <b>18 Chaplin St. Stafford Heights. QLD 4053</b>   |
|          | Postal address: <b>ENTOX, 39 Kessels Road, Coopers Plains, QLD 4108</b> |
|          | Telephone No.: <b>(07) 3274 9147</b>                                    |
|          | Fax No.: <b>(07) 3274 9003</b>  |
|          | Email address: <b>cwaugh@entox.uq.edu.au</b>                            |

5 Applicant details (if different from proposed permit holder(s))

|   |
|---|
| Name: Dr. Susan M. Bengtson Nash  |
| Residential address: <b>223 Agnew Street, Seven Hills, QLD 4170</b>     |
| Postal address: <b>EnTOX, 39 Kessels Road, Coopers Plains, QLD 4108</b> |
| Telephone No.: <b>(07) 3274 9004</b>                                    |
| Fax No.: <b>(07) 3274 9003</b>  |
| Email address: <b>s.nash@uq.edu.au</b>                                  |

Qualifications and experience: **B. Sc. (hon)**. CW has no prior practical experience of the proposed sampling technique or individuals of the named species. As the PhD student on this research program she will conduct extensive literature reviews and, under the supervision of experienced whale researchers and the CI, will drive the development of sampling devices according to the literature and best practice, and will collect samples from the whales. Under supervision of the CI, she will conduct tissue POP analyses.

6 Give the relevant qualifications and experience of all people who will carry out the activities. If insufficient space, attach a list.

|          |  |
|----------|--|
| <b>1</b> | Name: Dr. Susan M. Bengston Nash   |
|          | Qualifications and experience: <b>B.Sc. (hon); PhD</b> . SBN represents the research leader and chemical analytical expertise on the project. Whilst SBN has no prior practical experience of the proposed sampling technique or living humpback whales, she has conducted extensive literature reviews on existing cetacean sampling techniques and will actively participate in the development and validation of the proposed methodology. SBN will undertake training in proposed sampling techniques together with CW under the supervision of Dr. Nick Gales and Dr Mike Noad. |

|          |   |
|----------|---|
| <b>2</b> | Name: Dr. Michael Noad  |
|          | Qualifications and experience: <b>BVSc.; PhD</b> . MN has extensive previous experience working with migrating humpback whales (12 years) including collecting biopsy samples using biopsy darts. |

|          |                      |
|----------|----------------------|
| <b>3</b> | Name: Courtney Waugh |
|----------|----------------------|

7 Have you applied for or obtained any other approvals, permits or licences relating to this activity under Commonwealth, State or Territory legislation?

No  ⇒ *Go to next question*

Yes  ⇒ *Attach copies*



8 Have you previously held a permit from the Australian Government to conduct this activity?

No  ⇒ *Go to next question*

Yes  ⇒ *Give details below*

| Permit number | Date permit expired |
|---------------|---------------------|
|               |                     |
|               |                     |
|               |                     |
|               |                     |
|               |                     |

9 Offences

A proposed permit holder is taken to have been convicted of an offence if, within 5 years before the application is made, the proposed permit holder:

- has been charged with, and found guilty of, the offence but discharged without conviction; or
- has not been found guilty of the offence, but a court has taken the offence into account in passing sentence on the proposed permit holder for another offence.

Section 6 of the *Crimes Act 1914* deals with being an accessory after the fact. Sections 7 and 7A and subsection 86(1) of the *Crimes Act 1914* and sections 11.1, 11.4 and 11.5 of the *Criminal Code* deal with attempts to commit offences, inciting to or urging the commission of offences by other people and, conspiracy to commit offences.

Part VIIC of the *Crimes Act 1914* includes provisions that, in certain circumstances, relieve persons from the requirement to disclose spent convictions and require persons aware of such convictions to disregard them.

Has the proposed permit holder been **convicted** of, **or subject to proceedings** for an offence under any of the following?

- offences under the *EBPC Act* or *Regulations*
- a law of the Commonwealth or a State or Territory about the protection, conservation or management of native species or ecological communities;
- section 6, 7 or 7A, or subsection 86(1), of the *Crimes Act 1914* (Commonwealth) or sections 11.1, 11.4 or 11.5 of the *Criminal Code Act 1995* (Commonwealth) in relation to an offence under a law mentioned in (a) or (b) above; or
- a provision of a law of a State or Territory that is equivalent to a provision mentioned in (c) above.

No

Yes  ⇒ *Attach details*



10 Attachments

Indicate below which documents are attached.

Additional permit holders  
*See question 4*

Additional qualifications details  
*See question 6*

Copies of other approvals/permits  
*See question 7*

Details of offences  
*See question 9*

Other supporting documentation  
*List all additional documents below*

Titles of all attached documents (*include the document title, the specific section(s) and the page number(s) on which the information appears*)

11 Declaration

I declare that the information contained in this application is correct to the best of my knowledge.

Signature of applicant

Name of person signing

Date



# Supplementary Form A — Whales and Dolphins (cetaceans)

Application under section 238 of the Environment Protection and Biodiversity Conservation Act 1999.



If the person completing this form is representing a small business (i.e. a business having less than 20 employees), please provide an estimate of the time taken to complete this form.

Please include:

- the time taken spent reading the instructions, working on the questions and obtaining the information; and
- the time spent by all employees in collecting and providing this information.

Hours

Minutes

## This form has two purposes:

1. To apply for a permit to undertake an action which will contribute significantly to the conservation of whales and dolphins such as research on whales and dolphins.
2. To apply for a permit to interfere with whales and dolphins, where that interference is incidental to and not the purpose of the action, for example, building an underwater structure where you may come into contact with whales or dolphins.

Please supply the following information if you will interfere with, injure, take, keep, move, possess or treat (cut up/divide) a cetacean or part of a cetacean in the Australian Whale Sanctuary or waters beyond the Australian Whale Sanctuary (overseas). If you are proposing to send specimens out of Australia you will need an export permit. Import permits will be necessary for bringing parts or products of cetaceans into Australia. For more information on imports and exports contact International Wildlife Trade Section on 02 6274 1900.

This form should be completed in conjunction with The General Permit Application form.

## If you need more space

If there is insufficient space on this form to fully address any of the questions please attach additional pages and list these attachments at question 16.

When using additional documentation to answer individual questions in this application, please refer to the document title, the specific section(s) and the page number(s) on which the information appears.

## Application fee

There is a \$25 fee for permits where the action will contribute significantly to the conservation of cetaceans. There are some fee exemptions in certain circumstances, details of which can be obtained from the Department at the below address.

## Where to send the forms and the application fee

Please send the completed General Permit Application and this form and any accompanying attachments to:

Director  
Cetacean Policy and Recovery Section  
Department of the Environment and Water Resources  
GPO Box 787  
CANBERRA ACT 2601  
Fax: 02 6274 1542

1 Details of species that will be affected by the action. Use the following codes to enter details in columns 3 and 5.

| <b>Column 1</b><br>Common name of species.<br>Common and scientific names are available at the Departmental website:<br><a href="http://www.environment.gov.au/erin/applications/biodiversity/sprat/">http://www.environment.gov.au/erin/applications/biodiversity/sprat/</a> | <b>Column 2</b><br>Scientific name of species | <b>Column 3</b><br>Conservation status of threatened species under EPBC Act (e.g. the blue whale is endangered EN)<br><br><b>Codes for Column 3</b><br>EW Extinct in the wild<br>EX Extinct<br>CE Critically endangered<br>EN Endangered<br>VU Vulnerable<br>CD Conservation dependent | <b>Column 4</b><br>Estimated number that will be affected. | <b>Column 5</b><br>Type of effect<br><br><b>Codes for Column 5</b><br>IC Interfering with a cetacean<br>IN Injuring<br>TA Taking<br>KE Keeping<br>MO Moving<br>TC Treating<br>PO Possessing |
|---|---|--|--|---|
| Humpback Whale  | Megaptera novaeangliae                        | VU   | 120  | IC  |
|   |   |  |  |   |
|   |   |  |  |   |
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|   |   |  |  |   |

2 Provide the latitude and longitude of where the action will be conducted. Latitude and longitude references should be used instead of AMG and/or digital coordinates.

Where the project area is less than 1 square km, provide a single pair of latitude and longitude references.

Where the project area is greater than 1 square km or any dimension is greater than 1 km, attach a list of coordinates to enable accurate identification of the location of the project area.

Latitude

Degrees Minutes Seconds

|  |  |  |
|--|--|--|
|  |  |  |
|  |  |  |
|  |  |  |
|  |  |  |

Longitude

Degrees Minutes Seconds

|  |  |  |
|--|--|--|
|  |  |  |
|  |  |  |
|  |  |  |
|  |  |  |

Locality

LIST OF COORDINATES ATTACHED

3 *Attach an A4 sized map to show the boundaries of the area in which the action will be conducted.*



4 Provide an attachment describing the action addressing the following points.




- A. The equipment and methods used to comply with the EPBC Act Regulations.
- B. What steps will be taken to minimise impacts on cetaceans.
- C. The objectives and purposes of the action.

5 Attach a description of any research relevant to the affected species or community that will be carried out in the course of or in conjunction with the proposed action, including:



- A. A copy of the research proposal.
- B. The names of the researchers and institutions involved in or supporting the research.
- C. Relationship of the researchers to the permit applicant, including any funding being provided by, or to, the permit applicant

- 6 Will the action involve invasive techniques?
- No  ⇒ *Go to next question*
- Yes  ⇒ Attach application and approval from an Animal Ethics Committee. 

- 7 Are you applying on the basis that the action will contribute significantly to conservation of cetaceans? (Please note, a fee of \$25 is required for this type of permit — see Question 21)
- No  ⇒ *Go to 9*
- Yes  ⇒ *Go to next question*

- 8 Why do you believe that the action will contribute significantly to the conservation of cetaceans?

In the immediate term the project will contribute a valuable new non-destructive sampling technique for studying a vulnerable species. The study will also contribute the first data on biopsy validation efforts for organochlorine (OC) studies on the species.

The study represents the first attempt at quantifying toxicological endpoints associated with mobilised OC burdens during extreme life-history adaptations such as migration and fasting. The timely need for this research is augmented by the 2008 inclusion of humpback whales in the annual harvest of whales in the Southern Ocean whale sanctuary under the Japanese Scientific purposes permit. Progressive scientific research on the species based on non-lethal sampling methodologies represents a resilient long-term strategy against scientific whaling programs.

- 9 Are you applying on the basis that the effect on cetaceans will be incidental to, and not the purpose of, the action?  
You must also answer questions 10, 11 & 12 to apply for this type of permit.

No  ⇒ You are not able to apply for a permit using this form, please contact the Cetacean Policy and Recovery Section at [epbcwild@environment.gov.au](mailto:epbcwild@environment.gov.au), or call (02) 6274 1111.

Yes  ⇒ Why do you believe that the impact of the action will be incidental to and not the purpose of the action?

The purpose of biopsy sampling will be the collection of high quality (healthy, free ranging animals) analytical samples that can not be obtained by any less intrusive means. The level of interference with the animals will be kept to a minimum throughout the process. Any reaction by the whales to our presence or sampling will be noted but the purpose of our study is not behavioural observations, rather tissue sample collection for chemical analyses.

*Now go to 13*

- 10 Why do you believe that the proposed action will not adversely affect the conservation status of a species of cetacean or population of that species?

Many studies have shown (e.g. Best et al., 2005) that whales either do not react at all to a shallow biopsy or display a single tail-lob or similar behaviour indicating that they have felt the biopsy dart but there is no lasting sign of irritation or pain. Documented reactions to deeper biopsies (Lambertsen et al., 1994; Gauthier & Sears, 1999; Hooker et al., 2001; Reeb & Best, 2006) show no evidence to suggest that the reaction is any greater than for shallow biopsies. This is probably due to the skin, which is rich in nerves, being penetrated regardless of the biopsy depth while the blubber itself has comparatively few nerves.

- 11 Describe how the proposed action will be consistent with any *recovery plans* or *wildlife conservation plans* in force for the species of cetaceans that may be affected by the action.

*Commonwealth recovery and wildlife conservation plans that are in force are available from the Department of the Environment and Water Resources web site:*

[www.environment.gov.au/biodiversity/threatened/recovery/index.html](http://www.environment.gov.au/biodiversity/threatened/recovery/index.html)

*State and territory recovery plans will be available from state and territory environmental agencies.*

The recovering populations of humpback whales face a number of threats as outlined in the Department of Environment and Heritage humpback whale recovery plan (DEH 2005). The proposed action will be consistent with this recovery plan by advancing the discipline of wildlife toxicology and thus providing important indication for an area of research where information is completely lacking via non-lethal means. Further, the importance and timely need for the proposed research is augmented by the imminent introduction (2008) of Antarctic humpback whale populations into the annual harvest of whales from the Southern Ocean whale sanctuary.


12 *The applicant is required to take all reasonable steps to minimise interference with cetaceans.*

How will this be carried out?

Sampling events will be conducted by Australia's leading humpback whale experts who are experienced in approaching and sampling the whales. Their expertise will enable us to minimise the time spent tracking the whales and obtaining suitable samples.

Two team members are qualified veterinary scientists. All sampling equipment will be flame sterilised and annointed with broad spectrum antibiotic crème between sampling.

The sampling technique will be re-evaluated following the first season's results. If there is no evidence to suggest blubber stratification differences in contamination burdens the biopsy depth will be reduced.

13 Attach details of any proceedings against the proposed permit holder under a Commonwealth, State or Territory law for the protection of the environment or the conservation and sustainable use of natural resources. 

#### 14 Fees

The following fees apply:

- If you answered **yes** at question 7, for an action which will contribute significantly to the conservation of cetaceans - \$25 (there are fee exemptions in some circumstances).
- An incidental action relating to cetaceans - nil

15 Are you paying by credit card?

No  ⇒ Attach a cheque, go to next question

Yes  ⇒ Complete the following details

Card: Visa  Bankcard  MasterCard

Card number

Expiry date (month/year)

Card holder's name as shown on card

Amount

Cardholder's signature

## 16 Attachments

Indicate below which documents are attached.

- Attach a map. *See question 3*
- The equipment and methods used to comply with the EPBC Act Regulations. *See question 4*
- What steps will be taken to minimise impacts on cetaceans. *See question 4*
- The objectives and purposes of the action. *See question 4*
- Copy of research proposal. *See question 5*
- Names of researchers and institutions. *See question 5*
- Relationship of researcher to permit applicant. *See question 5*
- Ethics committee approval. *See question 6*
- Details of any proceedings against the permit holder under a Commonwealth, State or Territory law. *See question 13*
- Cheque for payment of fee. *See question 14*
- List all additional documents below

Titles of all attached documents (*include the document title, the specific section(s) and the page number(s) on which the information appears*)

Question 2. List of coordinates attached.

## 17 Declaration

I declare that the information contained in this application is correct to the best of my knowledge.

Signature of applicant

Name of person signing

Date



**QUESTION 2 Attachment of latitude and longitude of where the action will be conducted**

In Australia humpback whales are distributed throughout the Australian Antarctic, Commonwealth offshore, State and Territory waters. Australia had two migratory populations of humpback whales, a west coast and an east coast (known as Group D and Group E1 respectively in international fora). We will be sampling both populations during northward and the southward migrations. The project area will vary according to the geographical and temporal movements of the humpback whales as described below.

**WESTER AUSTRALIA (Group D population)**

The northern and southern migratory paths of the humpback whale stock that winters off Western Australia (Group D population) covers some 3600 nautical miles (Jenner et al., 2001) from the calving grounds in the Kimberley to the feeding grounds south of 56°S and between 70°E and 110°E.

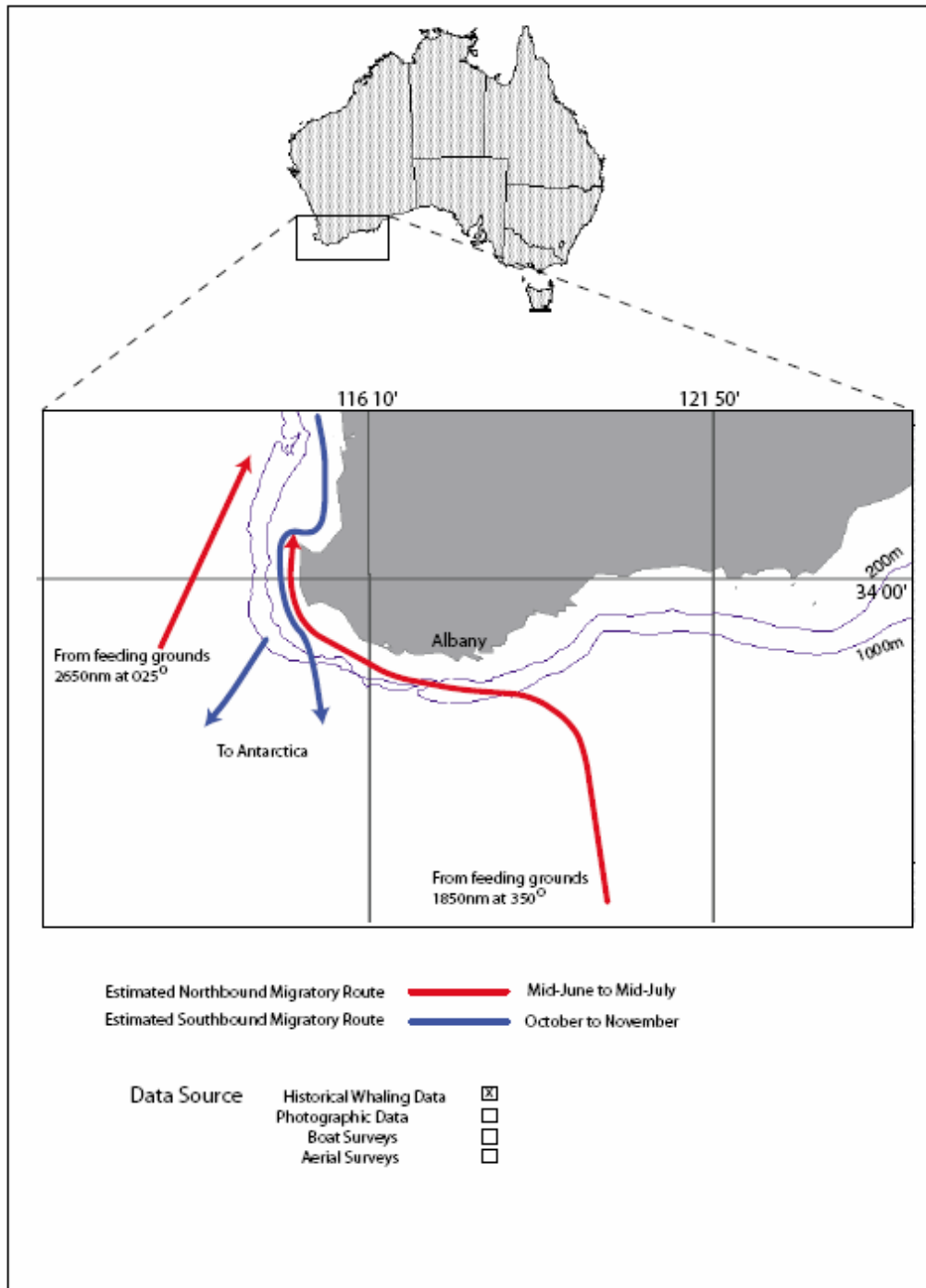


Figure 1. Estimated migratory routes for South-west Australian coast (35°S) (Jenner et al., 2001)

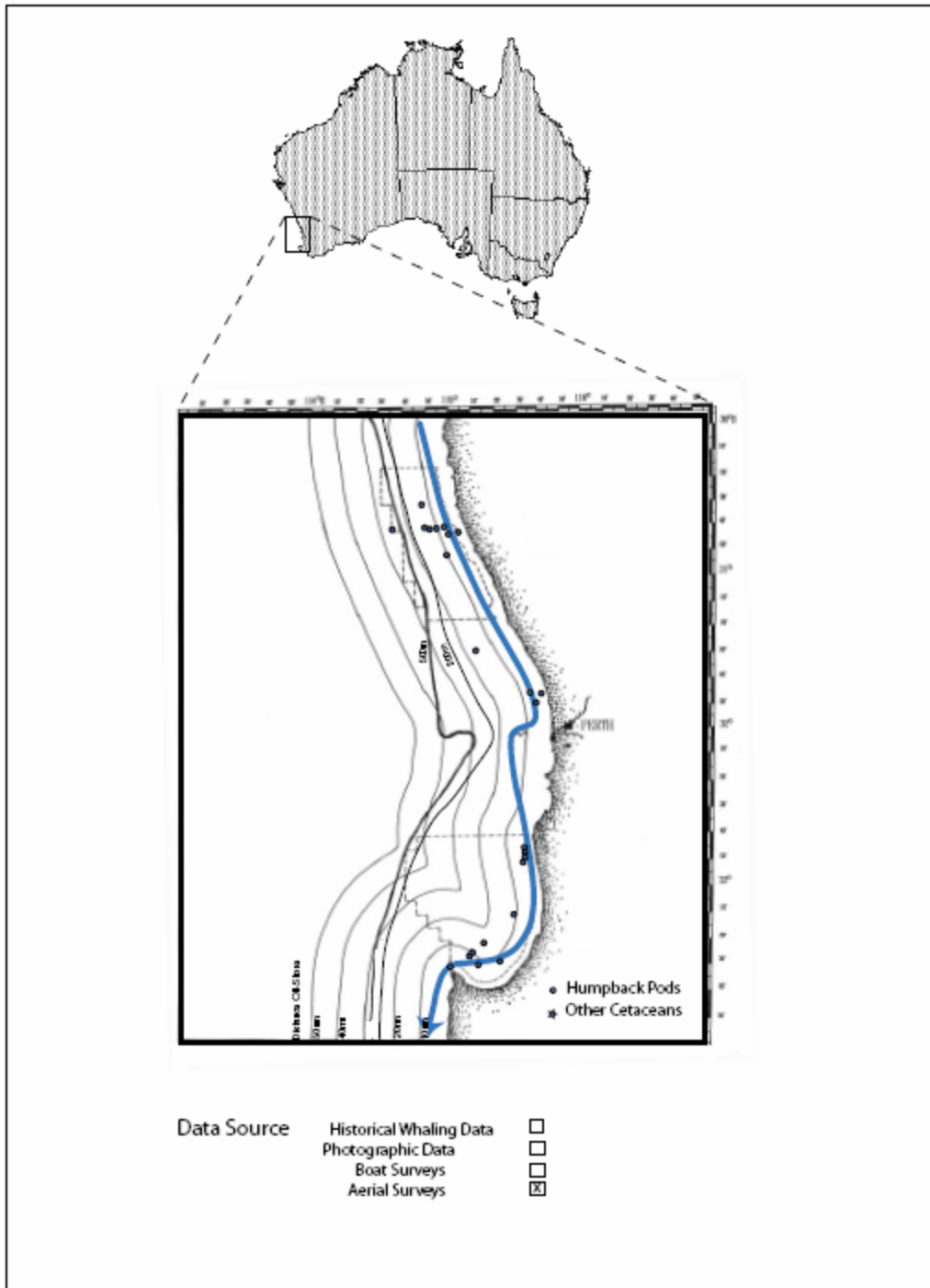


Figure 2. Estimated migratory routes from Perth Basin to Jurien Bay (33°40'S to 30°15'S)(Jenner et al., 2001).

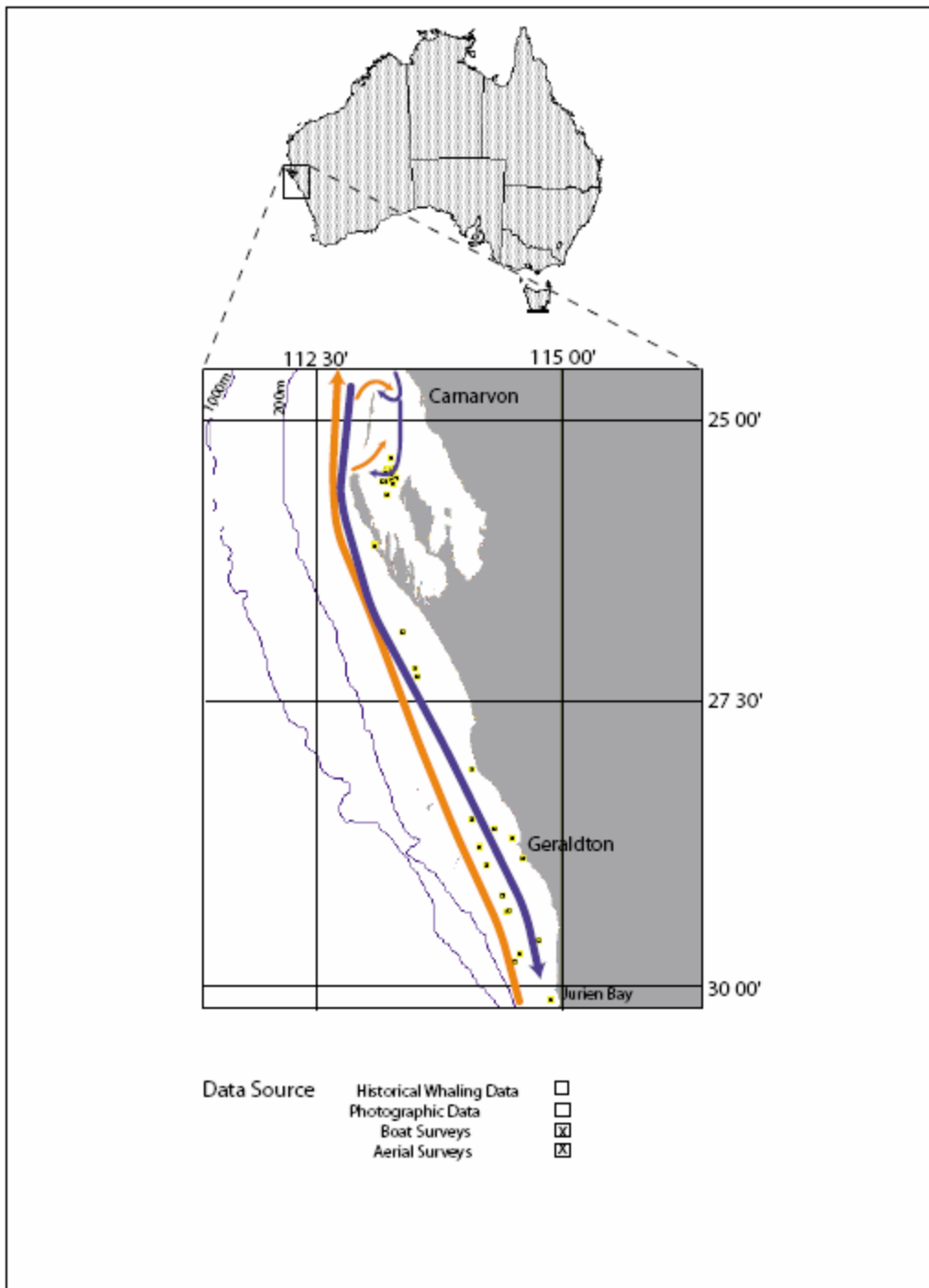


Figure 3. Estimated migratory routes from Jurien Bay to Carnarvon (30°15'S to 24°38'S(Jenner et al., 2001).

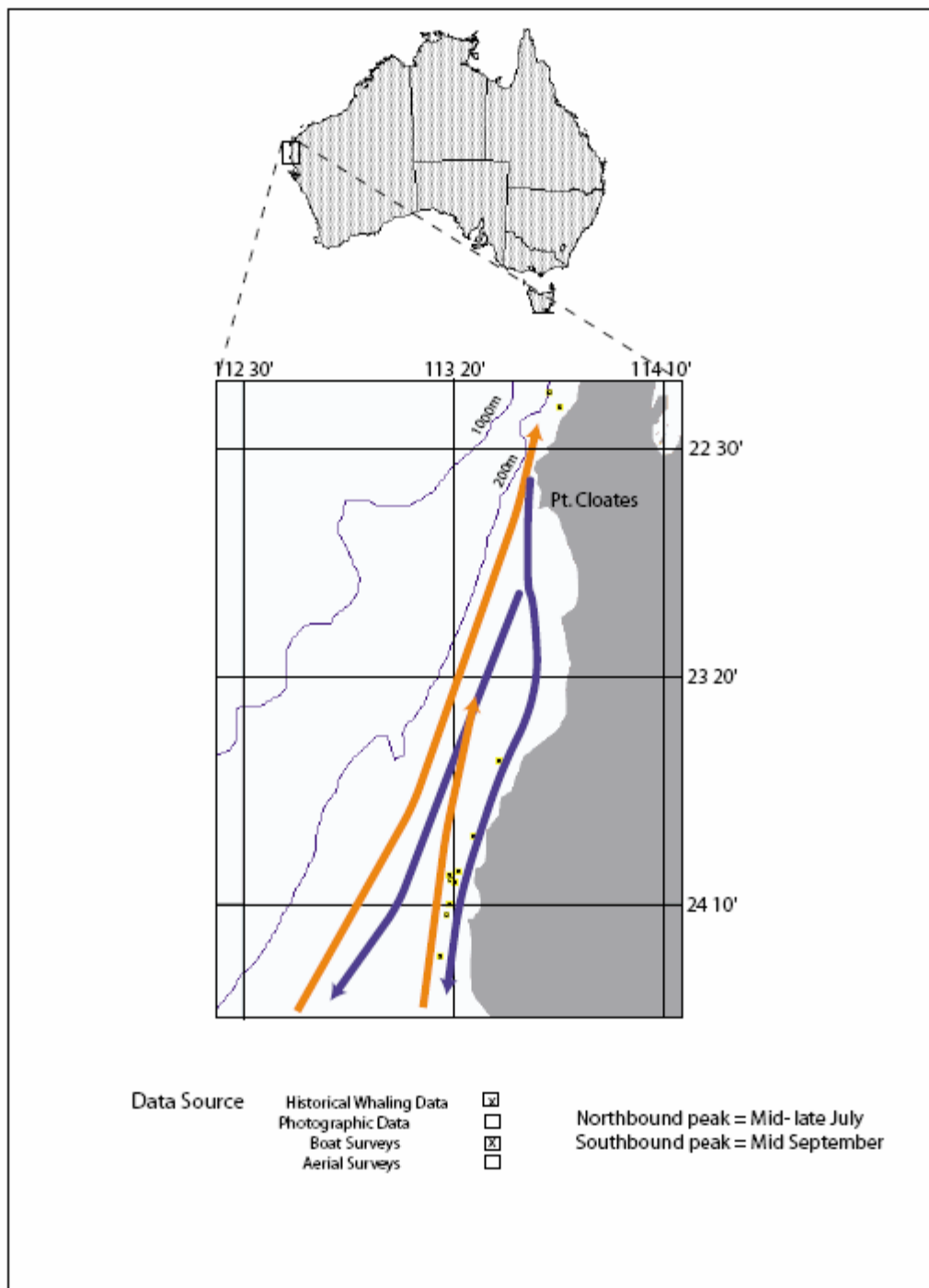


Figure 4. Estimated migratory routes from Carnarvon to Point Cloates (24°38'S to 22°S) (Jenner et al., 2001).

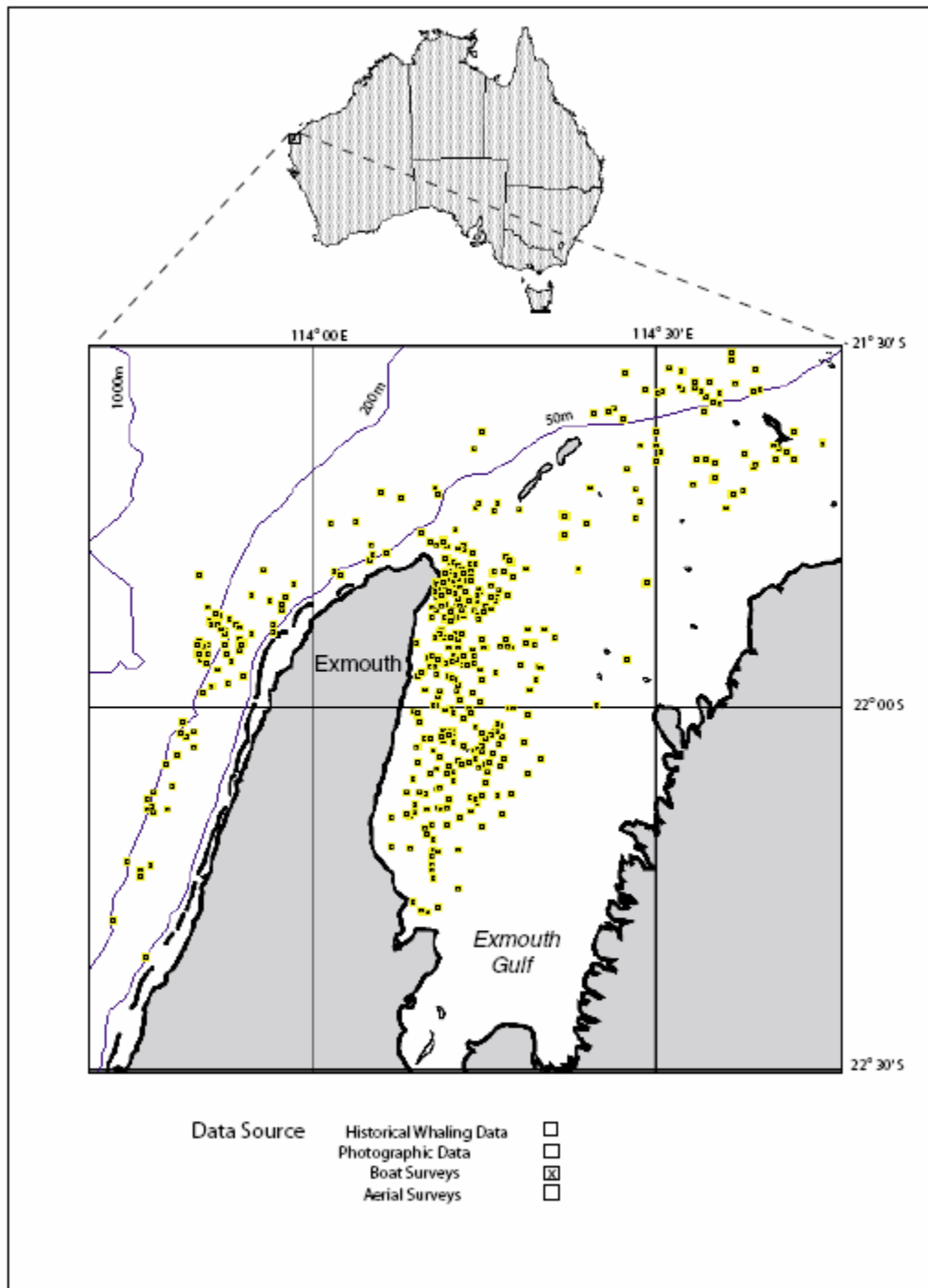


Figure 5. GPS positions of pods of humpback whales in Exmouth Gulf (21°S)(Jenner et al., 2001).

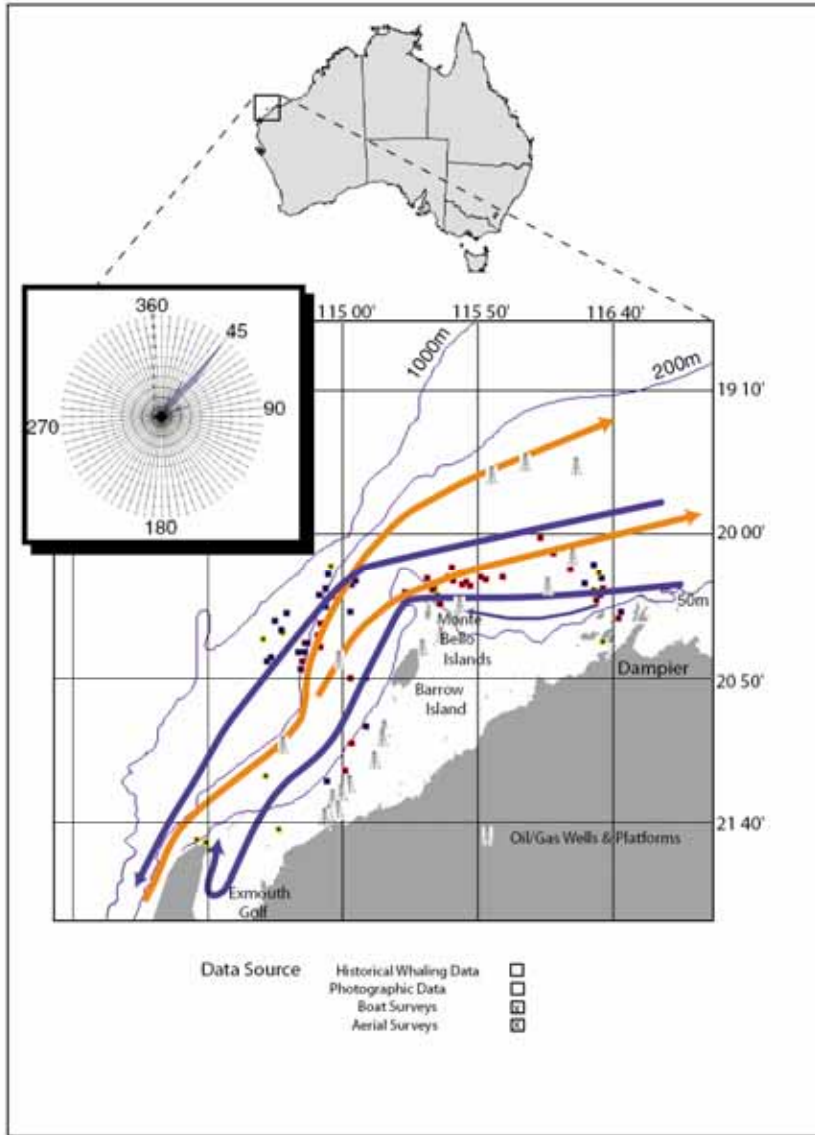


Figure 6. Estimated migratory routes for the Dampier Archipelago (20°S)(Jenner et al., 2001).

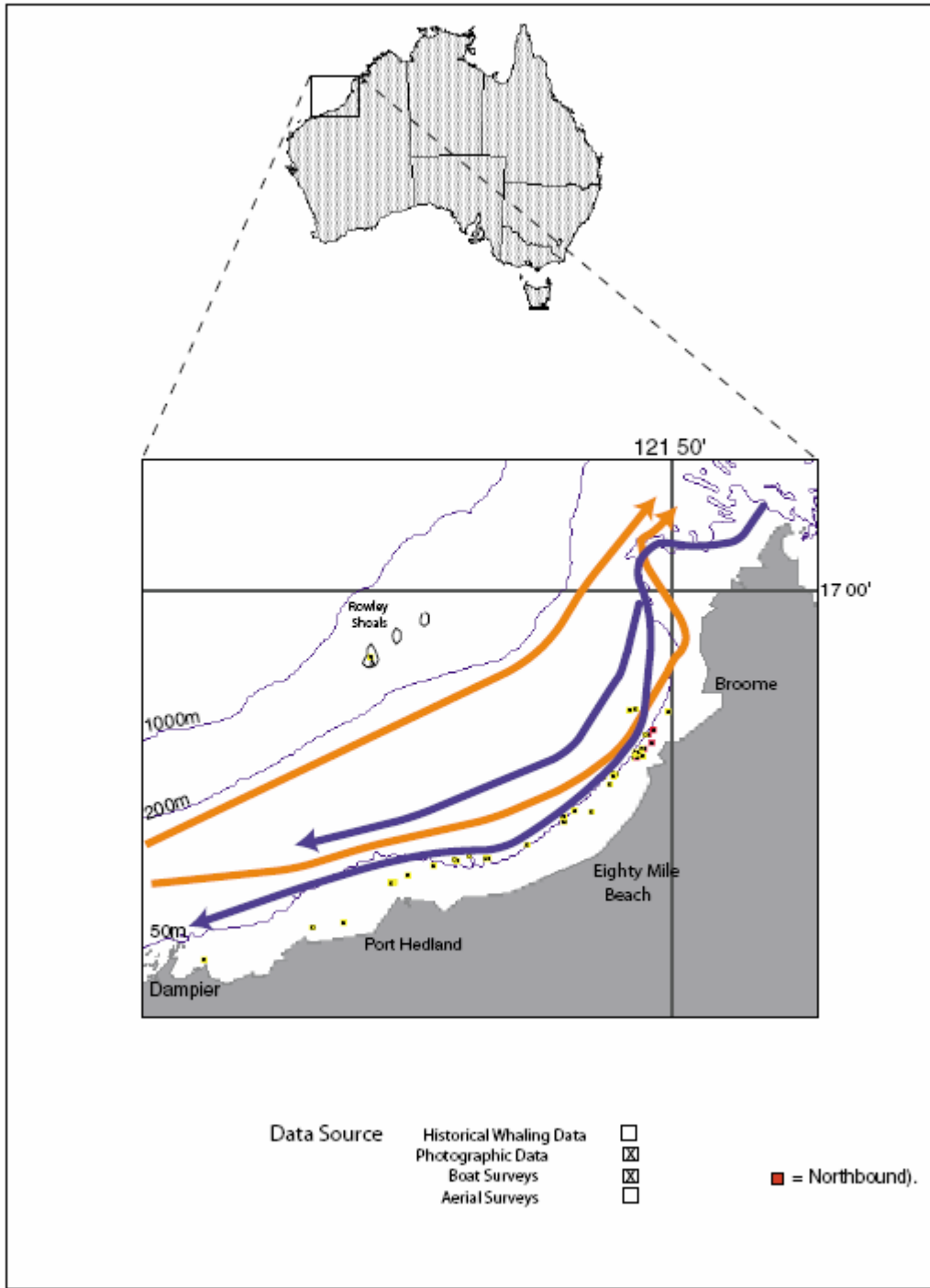


Figure 7. Estimated migratory routes from the Dampier Archipelago to Broome (20°S to 18°S)(Jenner et al., 2001)

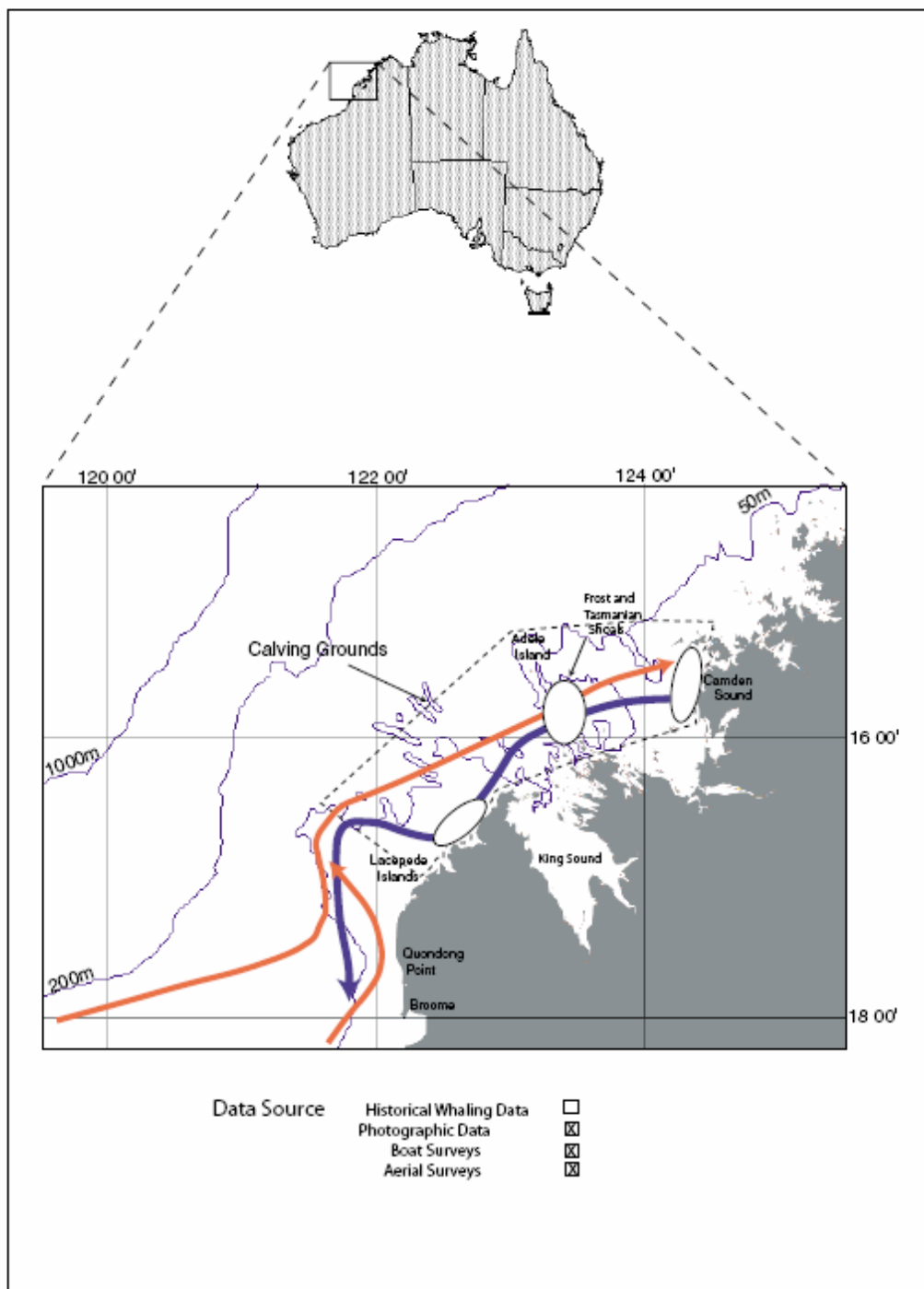
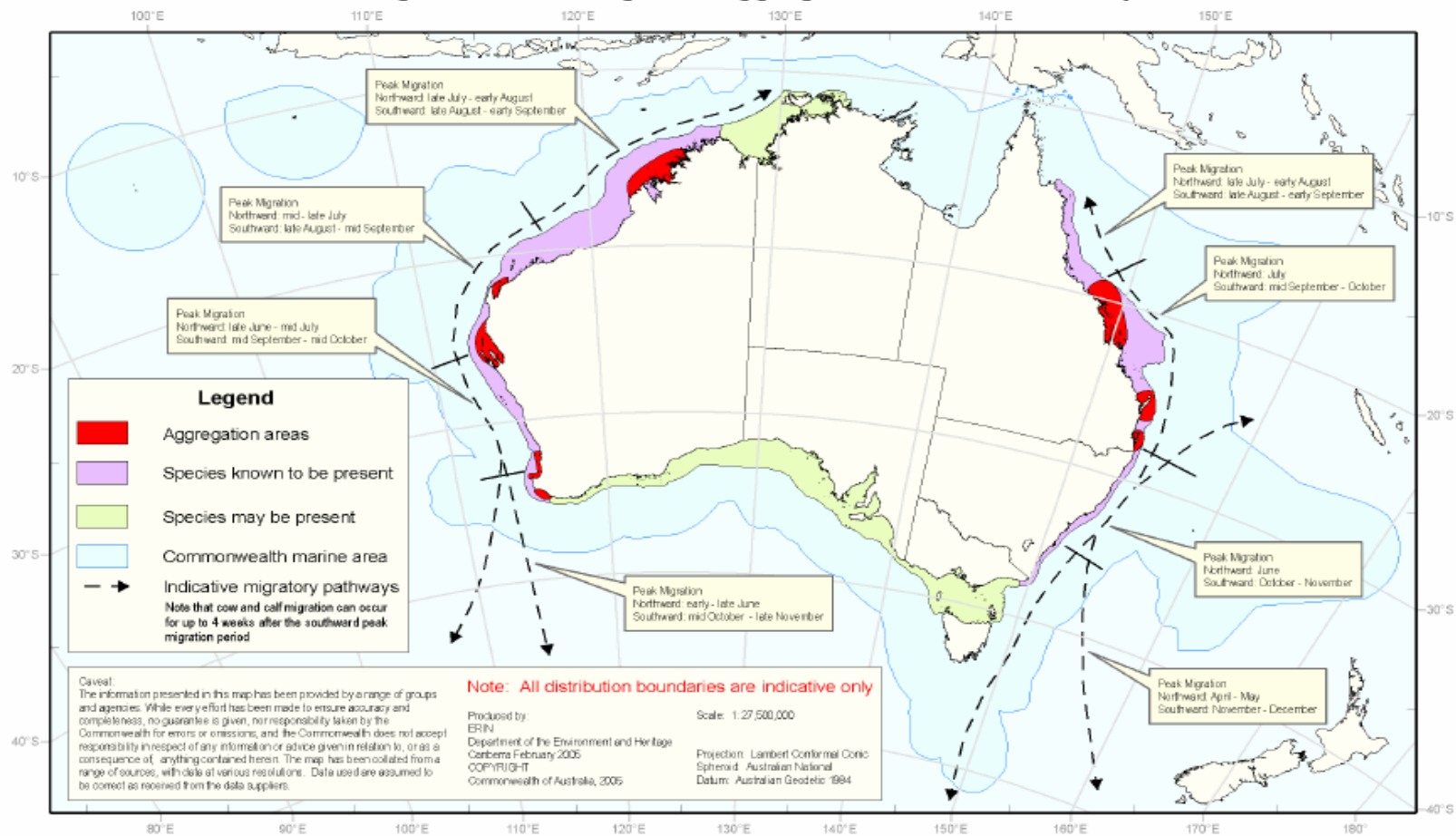


Figure 8. Estimated migratory routes from Broome to Camden Sound (18°S to 15°S) (Jenner et al., 2001)

Jenner, K. C. S., Jenner, M. N. & McCabe, K. A. 2001. Geographical and temporal movements of humpback whales in Western Australian waters. *APPEA Journal*, **38**, 692-707.

## EASTERN AUSTRALIA (Group E population)

The east Australian humpback whale population migrates along the coast between 130°E and 170°W. The southern latitudinal boundary is defined as the Great Barrier reef in an area between Port Douglas (16°S) and Whitsunday islands (21°S). The project area will be undertaken mostly in the Moreton Bay Region (26 to 28°S and 153 to 154°E).



**QUESTION 3. Boundaries of the project area will be confined to the Aggregation areas (red) and the areas in which the species are known to be present (purple) along the west and east coast of Australia. Map referenced from (DEH, 2005)**

DEH. 2005. Humpback Whale Recovery Plan 2005 -2010. pp. 10. Canberra: Australian Government Department of Environment and Heritage.

**QUESTION 4 Provide an attachment describing the action addressing the following points**

*A. The equipment and methods used to comply with the EPBC Act Regulations*

Recently our research framework (Persistent Organic Pollutant in the Southern Ocean Ecosystem), commenced a new project aimed at evaluating species specific toxicological effects of OC compounds in migrating humpback whales. The first stage of the project will be to investigate the relationship between blubber stratification and OC contaminant burdens in the species. To achieve this aim we propose full-thickness blubber biopsy sampling conducted within a team of humpback whale experts.

Sampling will be conducted onboard a small boat (open, aluminium or rigid-hulled inflatable, approx 6 or 7m in length, with outboard motor) which will approach the whales to within 10m to allow accurate aim of the sampling device. The biopsy dart is fired from a paxarms biopsy rifle at the flank of the whale immediately below the dorsal fin as the whale surfaces. The biopsy dart has a custom built head that consists of a stainless steel tube externally beveled at the tip to form a punch which is screwed into the front of the dart. The dart, containing the sample, bounces off the flank of the whale immediately and floats until retrieved. Between biopsies, the darts will be cleaned with ethanol, flamed, and anointed with a broad spectrum antibiotic cream to reduce the risk of infection at the biopsy site.

Previous studies for full-thickness sampling of whales have used depths varying between 90-200mm (Lambertsen, Baker et al. 1994; Reeb and Best 2006) and tags used have been to a depth of 200mm (Hooker, Baird et al. 2001). In large whales the blubber can be up to 400mm thick (Aguilar and Borrell 1994) For the current study two types of coring heads, with different depths (10-20cm), each with a diameter of 0.6-0.8cm, will be manufactured to account for the changing nutritive condition of the whales during the northward and southward migrations.

In 2007 the sampling periods of study will occur between June and November. Each sampling event will aim to capture biopsies from 30 male humpback whales (*Megaptera novaeangliae*) to avoid confounding factors of parturition and lactation. Actual sampling will be adjusted to ensure that the probability of capturing sufficient male samples is realistic. The main sampling period of this study will occur in 2008 between June and October. Samples will be taken during peak northward and southward migrations in QLD and WA. Each sampling event will again capture biopsies from a maximum of 60 animals to ensure confidence that 30 male samples have been captured. Sexing of samples will be later performed by PCR.

*B. What steps will be taken to minimise impacts on cetaceans*

The discomfort or stress presented by standard biopsy sampling on cetaceans is generally considered to be minimal. If properly done, collection of a biopsy is probably painless and reaction of the animal to darting is minimal and essentially short-term. Many field workers have observed to their surprise that cetaceans react more strongly to the noisy splash of a missing dart breaking the water surface near them than a hit to their own body (Aguilar and Borrell 1994). Despite its unusual thickness (1-mm) cetacean skin has a large germinative layer which permits high epidermal cell proliferation rates and healing is considered to be as fast as in man or other mammals (Aguilar and Borrell 1994).

Although deeper penetration (15-20 cm) of the proposed full-thickness biopsy core will present a higher level of invasiveness, studies have shown that the depth of the biopsy does not significantly affect the behaviour of the animal. Many studies

have shown that the overall reactions of large whales, including humpback whales, to deep-core biopsy sampling did not differ significantly from the reaction of the whales to the small biopsy system by (Lambertsen, Baker et al. 1994; Gauthier and Sears 1999; Hooker, Baird et al. 2001; Best, Reeb et al. 2005; Reeb and Best 2006). The average reaction to all studies was that of being startled, and the biopsy site was hardly visible after biopsying took place, with no sign of integumentary or other trauma.

Possibly the animals may have a set reaction to any impact, such that the same response will be elicited unless some threshold is exceeded, perhaps through excessive or repeated impact. This is probably due to the skin, which is rich in nerves, being penetrated regardless of the biopsy depth while the blubber itself has comparatively few nerves.

In our study, biopsies will be restricted to one per animal and as little time as possible will be spent obtaining biopsy samples from the whales to minimise disturbance.

### *C. The objectives and purposes of the action*

Currently non-invasive biopsy techniques are employed widely among marine mammal researchers for sex determination. The technique is simple and non-invasive and recently researchers have also adapted biopsy heads for capturing blubber samples for toxicological analyses. Traditionally toxicological sampling has relied upon samples from stranded or incidentally caught animals. The biopsy method, if validated, has the potential to provide a far superior sampling strategy for sampling living, healthy, free roaming individuals.

Whale blubber, particularly in large whales, is not homogenous tissue throughout its depth but is composed with different roles and biochemical composition (Aguilar and Borrell 1991). Since the first marine mammal contamination studies from biopsy samples, there has been much debate as to whether blubber stratification leads to variations in organochlorine (OC) contaminant levels and thereby results in skewed estimates of contaminant burdens. The most comprehensive study that has been performed to investigate these questions analysed for OC compounds in the inner and outer layers of dorsal blubber in commercially whaled fin (89) and sei (23) whales (Aguilar and Borrell 1991). Significant differences were found for some congeners in both species but differences varied significantly between sexes and between species. Other studies have been performed on a much smaller scale on mixed whale species (none on humpback whales) which have found differences to be negligible (e.g. (Gauthier, Metcalfe et al. 1997)). The discrepancy in findings between studies and whale species highlight the need for properly investigating the relationship before applying the contamination data for further investigations.

A new project under the Southern Ocean Ecosystem Persistent Organic Pollutant (POP) framework seeks to investigate species-specific toxicological parameters in migrating humpback whales. The first stage of the project will be to investigate the relationship between blubber stratification and OC contaminant burdens in the species. To achieve this aim we propose full-thickness blubber biopsy sampling conducted within a team of humpback whale experts.

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