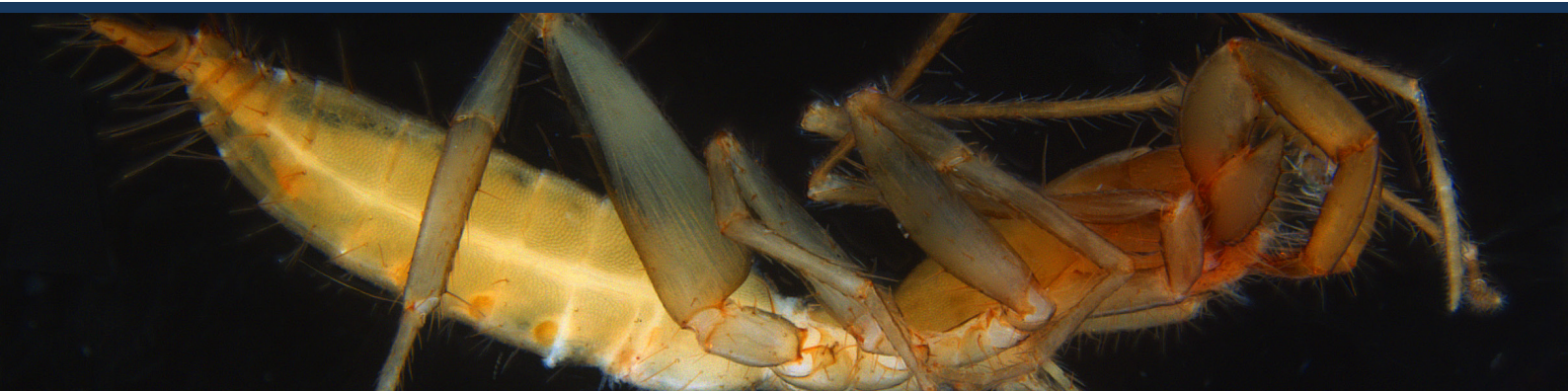


Technical guidance – Subterranean fauna surveys for environmental impact assessment



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1 Introduction

The purpose of this technical guidance is to ensure that subterranean fauna data of an appropriate standard are obtained and used for environmental impact assessment (EIA) and to determine whether the Environmental Protection Authority (EPA) objective for subterranean fauna can be met.

The EPA objective for the factor subterranean fauna is *to protect subterranean fauna so that biological diversity and ecological integrity are maintained.*

This technical guidance provides advice on:

- survey preparation and desktop study
- determining the type of survey required
- sampling techniques and survey design
- data analysis and reporting.

This guidance should be applied in conjunction with the *Environmental factor guideline – Subterranean fauna* (EPA 2016) under the 'land' theme in the EPA policy suite. Please refer to the environmental factor guideline for information on determining the impacts to subterranean fauna in the context of a proposal.

This guidance is applicable to subterranean fauna. For the purposes of EIA, subterranean fauna are defined as fauna that live their entire lives (obligate) below the surface of the earth (EPA 2016). Other fauna groups, including terrestrial short-range endemic (SRE) invertebrates, are addressed in the relevant EPA guidance documents.

This guidance should be applied when planning and undertaking subterranean fauna surveys for EIA under Part IV of the *Environmental Protection Act 1986* (WA).

In a state as large and diverse as Western Australia (WA), site-specific circumstances may require deviation from the methodology outlined in this document. In the case of any deviation, the appropriate agency or agencies should be consulted to discuss the adequacy of the survey design and techniques. Justification for the deviation from this guidance such as reduced sampling effort or use of alternative methods, and how best practice has been applied, must be presented in the limitations sections of survey reports.

2 Desktop study

A desktop study is a prerequisite for surveys and EIA. The purpose of a desktop study is to assess the likelihood of subterranean fauna being present in a proposal area based on a habitat assessment and a review of relevant fauna records. A desktop study is not a survey.

Much of WA remains poorly studied for subterranean fauna. Even in areas with high levels of survey, such as the Pilbara, site-specific information can often be limited or absent entirely. The desktop study gathers contextual information about an area from existing surveys, literature, database searches and spatial datasets. The outcomes of the desktop study are used to determine whether further survey is required and the appropriate level of survey.

The desktop study should place the proposal area into local and regional contexts and determine whether the proposal area is likely to provide habitat for subterranean fauna. This should include a summary of any fauna records from the proposal area, or relevant records from other areas with similar habitats in the region.

The basis for evaluating the likelihood of subterranean fauna habitat and species occurrence should be clearly stated with supporting evidence, particularly if concluding that no subterranean fauna habitat is present. Subjective, generic or poorly supported conclusions are unlikely to be sufficient justification for not undertaking survey.

A summary of previous surveys, including survey type and methods, survey effort, dates and timing, and the locations of previous sampling sites to the current proposal boundaries and predicted areas of impact should be collated to place the existing records into context, and to identify sampling and other knowledge gaps.

All information used in a desktop study should be evaluated for reliability and any limitations discussed. This should include consideration of the source and currency of the information, the adequacy of existing surveys and the consistency of the survey methods against current EPA guidance, data analysis used, changes in species status, nomenclature or taxonomy since reporting.

At the completion of a desktop study there should be sufficient information to determine the likelihood of fauna and habitats that may be present and place them in a regional context.

Background environmental information

A desktop study includes background environmental information and habitats likely to occur and a discussion of any significant species and habitats identified.

An accurate summary of background environmental information is required to place data into a local and regional context. This information should include discussion of relevant:

- Interim Biogeographic Regionalisation of Australia (IBRA) bioregions and subregions
- land use and tenure, e.g. land reserved for conservation purposes, pastoral leases, Indigenous Protected Areas, unallocated crown land and private freehold land
- recognised sensitive sites, e.g. priority ecological communities, Key Biodiversity Areas, Environmentally Sensitive Areas and Important Wetlands
- landscape characteristics, e.g. land systems, soil-landscapes, surface geology, topography, elevation, aquifers, surface water and drainage
- climate information, including rainfall and temperature, from a weather station with adequate long-term data representative of the study area.

2.1 Database searches

The desktop study includes a search of existing data on stygofauna or troglofauna records to provide an inventory of species or taxa groups that may occur in the area and the regional context. Examples of relevant data sources include:

- the relevant specimen databases of the [Western Australian Museum](#) (e.g. arachnids and myriapods, crustaceans and worms, molluscs, and entomology, where appropriate)
- specialist and private third-party datasets, where available
- regional subterranean fauna survey reports, available through the [Index of Biodiversity Surveys for Assessment \(IBSA\)](#)
- records of some groups may be available through [NatureMap](#) and the [Atlas of Living Australia](#).

IBSA may be useful for identifying where other surveys have been undertaken in the local area or region.

Records from all sources should be consolidated, identifying the diversity of subterranean taxa known from within the proposal area and surrounding locality, including any species formally recognised as conservation significant. Any issues regarding the taxonomy or identification of existing records should be considered and evaluated for accuracy.

2.2 Literature review

A literature review captures species records not available through databases, identifies the habitats that may be present and contributes to an overall understanding of the area and its regional context.

Literature reviews should include, but not be limited to, previous survey reports, unpublished survey datasets, locally held records (e.g. registers kept at mine sites) and any major regional survey reports. Scientific literature including peer-reviewed journal articles are useful for providing information on a species status, biology, and ecology.

Records incorporated into the literature review should represent observations from original sources, rather than records that were extrapolated or cited from derived sources. If information in a previous desktop study is relevant, the underlying original data should be included in the review rather than the previous desktop study itself.

2.3 Habitat assessment

The objective of the habitat assessment is to identify, characterise and document the extent of potential habitat in the proposal area, including any geological features that may limit the distributions of subterranean fauna.

The geological formations that support subterranean fauna contain suitable pores, voids, fractures and cavities that are necessary to provide interconnected spaces for air or water to be present.

Geological units known to provide stygofauna habitat are rock types or regolith deposits that have secondary porosity and are fully or partly saturated, usually occurring below the watertable, including, but not limited to: alluvial formations; calcretes - particularly when associated with paleochannel aquifers; channel iron deposits; fractured rock aquifers; karstic limestone and dolomite. Hyporheic zones, such as adjacent to streams and stream beds, marine interstitial zones and anchialine systems, such as those occurring in coastal limestone caves, may also host stygofauna from phreatic zones.

Troglofauna habitat is supported by geological units with rock types or regolith deposits occurring above the watertable that have secondary porosity including vuggy, weathered or cavernous rock formations, including but not limited to those present in karst, channel iron deposits, banded iron formations, calcretes, and weathered or fractured basalt and sandstone.

The key biophysical attributes that should be considered in identifying potential stygofauna and troglofauna habitats are presented in Table 1 and Table 2 respectively.

Table 1: Biophysical attributes and considerations in assessing habitat suitability for stygofauna

Biophysical attribute	Considerations
Surface hydrology	The location of drainage lines and catchments, and characterisation of surface flow in relation to groundwater recharge events and hyporheic zones.
Depth to watertable	Stygofauna may be more likely to occur when the watertable is relatively shallow and are generally less likely when it is deep. For example, there is a higher probability of stygofauna in the Pilbara bioregion where depth to watertable is less than 30 metres below ground level (Halse et al. 2014), but some stygofauna may occur at depths exceeding 100 metres (Hose et. al. 2015).
Surface geology	The extent of surface geology units may provide a foundation for stygofauna habitat mapping, particularly where groundwater is shallow and the superficial geology units that represent stygofauna habitat continue below the watertable. Consider whether the units representing stygofauna habitat mapped on the surface continue through the profile. If a different rock type occurs at depth, particularly if it hosts the aquifer present, then this may mean that surface geology mapping is of limited relevance.
Stratigraphy below watertable	The host geology types for the underlying aquifers, and their physical and hydraulic characteristics, should be considered and related to the geological units mapped on the surface. If the same units mapped at the surface occur through the profile to below watertable (e.g. as is often the case in alluvial valley fill settings) then this can provide a useful indication of habitat spatial distribution and extent. Drill logs and any other stratigraphic information available, e.g. geological / hydrological cross-sections and 3D modelling, should be considered jointly with geological mapping.
Spatial extent and vertical arrangement of aquifers and hydrated zones	The type of aquifers, depth and extent of aquifers, and inferred extent of host geology, can inform mapping of potential habitat and may be particularly relevant in setting context to assessments of stygofauna species distributions and dewatering impacts.
Cavities and fractures in below watertable geological units	Transmissivity of the host geology of the aquifers within the proposal area, and whether they contain fractures or cavities (faults, folds or shear zones), or gravel or grit layers, that could provide habitat space or zones for stygofauna.
Aquitards or aquicludes	The presence of impervious rock and shale, clay strata, dykes, sills or other impermeable layers may hydraulically separate aquifers, with the potential to restrict the distributions of stygofauna species and limit the extent of groundwater dewatering extents.
Groundwater physicochemical parameters	Potential habitat suitability can be informed by groundwater quality parameters (e.g. salinity, dissolved oxygen, pH) and how these parameters may vary spatially, at depth and temporally. How do aquifer conditions in the proposal area compare with documented ranges for key parameters from other areas of similar hydrology where stygofauna have previously been recorded?
Temporal variability in depth to watertable	The magnitude of watertable fluctuation may provide context to sampling results and characterise natural variability in stygofauna habitat conditions.

Table 2: Biophysical attributes and considerations in assessing habitat suitability for troglofauna

Biophysical attribute	Considerations
Surface geology	The extent of surface geology may provide a foundation for troglofauna habitat mapping, especially where superficially mapped geological units continue with depth through the vadose zone above the watertable. Cross-sections or drill core stratigraphy are important to understand continuation of subsurface geology.
Landforms	Troglofauna records are often associated with topographically-inverted landscapes, especially channel-iron deposits (CID), unconsolidated regolith, or hard rock ranges and hills, particularly when these are weathered and have been continuously emergent for long periods.
Depth to watertable	The area between the ground surface (base of the soil layer) and the watertable, often referred to as the vadose or unsaturated zone, defines the maximum potential troglofauna habitat available. However, some strata within this zone may not provide habitat space for troglofauna, and reviews of drill logs, geological cross-sections and other information should be considered when delineating potential habitat.
Stratigraphy above watertable	Identify the geological units containing fractures, cavities or interstices that could provide habitat for troglofauna. Consider whether suitable sub-surface habitats continue to the surface or are overlain by other geological units unlikely to be suitable habitat, and if the habitats identified occur as contiguous strata or as isolated patches.
Topographical and structural features	Deeply incised gorges or eroded river valleys, and faults and dykes may represent potential barriers to species dispersal, habitat connectivity or gene flow if intruded with solid rock. Conversely, deeply incised gorges or eroded river valleys, and faults and dykes may represent potential habitat connections if weathered, fractured or hydrated and containing habitat space that may be utilised by troglofauna and provide opportunities for movement and gene flow.

3 Determining survey type

This guidance outlines three survey types: basic, detailed and targeted. The type of survey required should be determined based on the survey objectives, existing available data, information required and the scale and nature of the potential impacts of the proposal. These aspects should be considered in the context of the information acquired by the desktop study.

Aspects to be considered when determining the type of survey required include:

- the level of existing local and regional knowledge to predict likely faunal assemblages
- comprehensiveness and currency of local surveys
- size and connectivity of habitats
- significance of species likely to be present
- the sensitivity of the environment to the proposed activities.

Figure 1 outlines the process for determining the survey type for subterranean fauna survey for EIA.

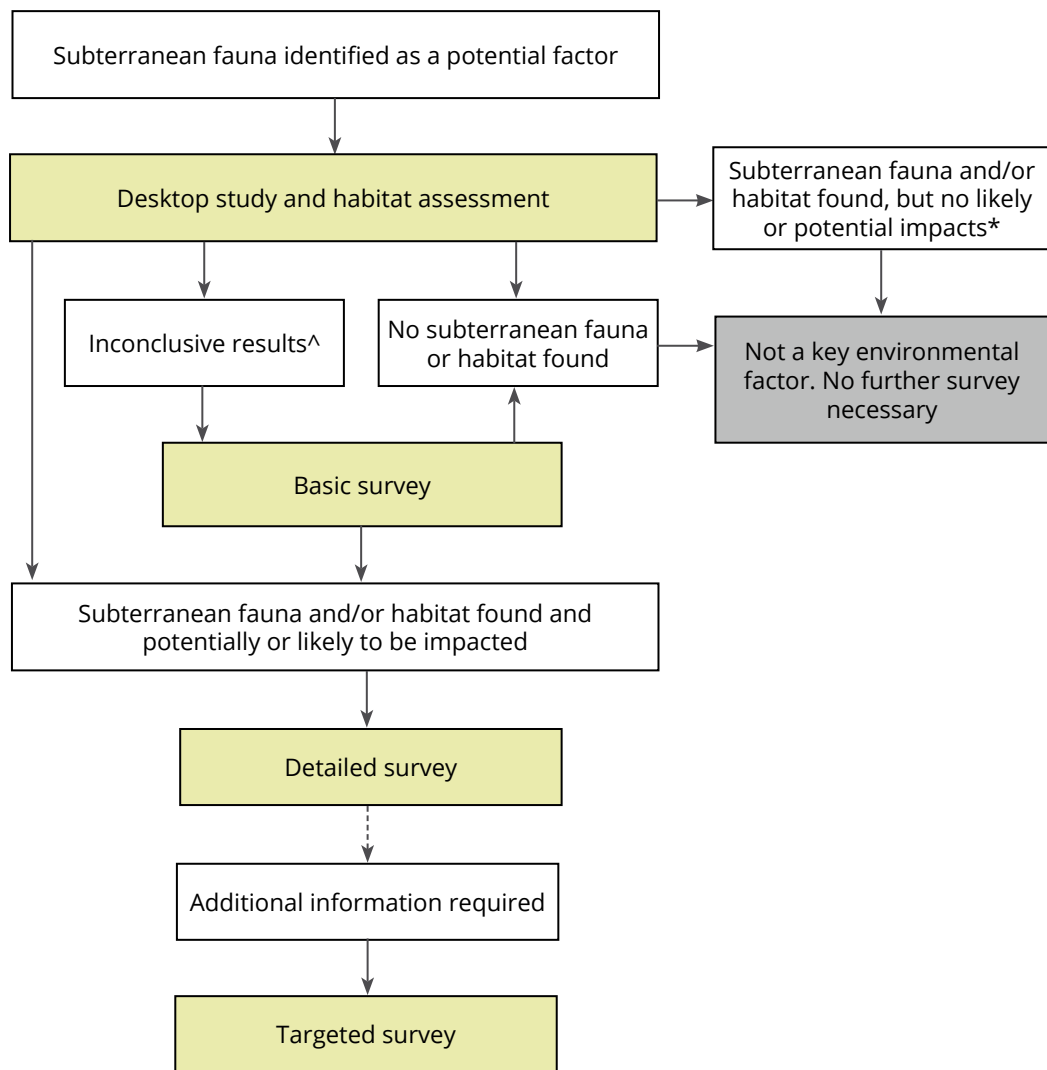
A basic survey is required where there is a lack of information or uncertainty in the available data examined through the desktop study, or when the outcome of the desktop study is inconclusive in determining whether subterranean fauna or habitat is likely. A paucity of records is often due to limited sampling effort and does not indicate a lack of subterranean fauna in an area, unless equivalent habitats in the surrounding area have been well sampled and were found to support low communities of fauna.

A detailed survey is required for proposals where subterranean fauna habitat is known or likely to occur, based on the desktop study. Where there is a high likelihood of subterranean fauna occurring, proponents should proceed directly to a detailed survey.

A targeted survey is undertaken when there is insufficient information, following a detailed survey, to inform the assessment and/or determine the significance of the proposal impacts e.g. targeting areas of the proposal or species.

An appropriate survey type should provide adequate information to determine impacts, conditions, offsets and an analysis of the cumulative impacts. Determining the type of survey requires consideration of the characteristics of the proposal and the scale and nature of the impact. However, small-scale impacts may not negate the need for a detailed or targeted survey. For example, given the high levels of short-range endemism of many subterranean fauna species, even small-scale impacts may be significant to locally restricted species.

Where a well-supported desktop study concludes that no subterranean fauna habitats are present and fauna are unlikely, and the results of the basic survey support this conclusion, no further survey would be necessary.



* Depending on the likely scale and nature of the proposal impacts

^ Go direct to Detailed survey if preferred

Figure 1: Process for determining the level of survey for subterranean fauna

4 Survey types

4.1 Basic

A basic survey involves low-intensity sampling of prospective habitats to verify the findings of the desktop study. The basic survey is usually completed in combination with or following the desktop study to provide a more confident evaluation of habitat occurrence or likelihood of subterranean fauna presence in the proposal area, particularly when the desktop information is inconclusive (see Figure 1). Standard sampling techniques are used, but the number of samples taken will vary according to the local area and information obtained during the desktop survey.

The results of a basic survey are used to inform and refine survey design for subsequent detailed or targeted surveys. A basic survey also provides the opportunity for reconnaissance of the proposal area, to confirm the landforms present and to identify logistical issues, such as borehole suitability and site access.

4.2 Detailed

A detailed survey aims to collect sufficient information to provide an understanding of the subterranean fauna values of a proposal area and to place these into an appropriate local and regional context.

Detailed surveys require repeat sampling, over multiple phases, and adequate survey effort to characterise the subterranean fauna of a proposal area and its habitats. The purpose of a detailed survey is to gather quantitative data on species, assemblages and habitats in an area. A detailed survey requires a comprehensive survey design, with sampling of impact and reference areas, and should include at least three survey phases.

A combination of primary sampling methods is appropriate for most detailed surveys.

4.3 Targeted

The objective of a targeted survey is to provide additional or specific data to address unresolved knowledge gaps. It can be used to supplement existing survey data by specific sampling of targeted areas to validate predictions of subterranean habitats; resolving knowledge gaps around habitat connectivity; or clarifying the wider distribution of a particular species, typically one known only from the proposal impact area.

Where the targeted survey does not clarify the knowledge gaps, then repeat sampling of the targeted areas, or sampling of additional areas, may be required.

Because impacts must be placed into context, targeted surveys are not necessarily confined to potential impact areas. For example, if the extent of habitat outside of the impact area is unknown, targeted sampling in the surrounding region may be required to obtain contextual data.

The sampling methods used should be selected based on the question to be resolved. Targeted sampling of specific subterranean taxa is usually only possible at the broadest ecological grouping (i.e. stygofauna or troglofauna). However, some sampling methods can prove more effective for certain taxonomic groups.

5 Preparation for survey

Surveys should be coordinated and led by zoologists with knowledge and experience in systematic subterranean fauna sampling, identification and analysis methods.

Survey leaders should have the skills to appropriately collect and voucher specimens, prepare and store genetic samples, accurately identify key morphological diagnostic characteristics and/or the ability to deal with taxonomic uncertainty. The survey leader should ideally oversee all aspects of the survey, including the analysis, reporting and data management, supervision of less experienced team members, and appointment of third-party specialists for taxonomic or genetic analysis.

Appropriate licences must be obtained to collect subterranean fauna specimens and permission must be obtained from landholders and managers to access or undertake surveys on their land. Proponents and consultants should liaise with the Department of Biodiversity, Conservation and Attractions (DBCA) to ensure that they comply with current statutory licensing obligations.

Surveys are often conducted in remote and difficult terrain, and health and safety issues must be planned for. Appropriate survey-specific safety procedures should be identified early in planning and complied with to ensure that work can be safely undertaken, e.g. cave work requirements.

Early engagement with the proponent's geotechnical and mineral exploration or other drilling programs (e.g. hydrogeological) is valuable as this may influence the location of bore or drill sites improving their suitability for subterranean fauna sampling. In addition, it offers the opportunity: for provision for additional sites to specifically target potential habitat (both within impact and reference areas); to request collection of specific geological information of value to habitat assessments; or supplementary sampling, including springs and hyporheic zones, and pump sampling of bore coordinated with groundwater or bore testing.

6 Survey techniques

A variety of sampling methods have been developed for subterranean fauna. Those considered most suitable for the purposes of EIA are reviewed below. This guidance is not prescriptive about the use of any particular technique, but highlights the benefits and limitations of the different techniques for stygofauna and troglifauna.

6.1 Stygofauna

Five stygofauna sampling methods have been identified as having applicability to EIA, of which haul net sampling is considered the primary method. The use of pumps, traps, plankton nets and environmental DNA techniques are all considered to be supplementary or only applicable in certain situations. When sampling for stygofauna is undertaken in conjunction with haul net scraping for troglifauna, the contents and outside of nets used for scrape samples should also be checked for stygofauna, particularly when sampling close to the watertable, and any bycatch added to the sample for that site.

1 Haul nets

Sampling groundwater using phreatic haul nets is the longest standing and most used sampling method for stygofauna surveys in WA.

The haul net used is a modified plankton haul net, sometimes using a modified scraping attachment above each net to dislodge specimens, with a metal (e.g. stainless steel or brass) catch bottle assembly at its lower end. Generally, a plankton mesh size of 63 μm is recommended, but other sizes i.e. 50 or 150 μm can be used to target specific sized taxa groups or to reduce net resistance from sediment during hauls.

The net is lowered ensuring the net reaches the bottom of the borehole, as stygal animals may often be in, or close to, the sediments at the base of the bore, and then hauled back to the surface, filtering animals out of the water column on the upward haul. Agitating the sediments during the latter hauls is also recommended to mobilise animals into the groundwater above the net, as this may increase the number of specimens collected.

Each sampling site is hauled a minimum of six times. After each haul, the groundwater sample collected is emptied from the catch jar and combined into an overall sample from each individual sampling site. The net and jar should be inspected for any animals after emptying and any organic matter rinsed into the combined sample to minimise the risk of specimen loss during subsequent hauls. To improve the likelihood of DNA sequencing, excess water can be removed by filtering the sample or flushing the sample with ethanol and the contents preserved immediately in chilled (less than 4°C) 100% ethanol, which should be changed in the first 12 to 24 hours. The sample should be kept chilled during transport and storage for later laboratory sorting.

On completion of sampling at each site, the haul nets should be thoroughly cleaned and inspected to remove all material and ensure no specimens are inadvertently transported between sites, as this may lead to incorrect conclusions regarding species distributions.

Groundwater samples can often be turbid, and if so, samples should be sieved and left to settle as part of the sorting process or clarified by other suitable methods. Combined samples from each bore should be sorted in the laboratory under a dissecting microscope by zoologists with training and experience in the identification and sorting of subterranean fauna specimens. All stygofauna specimens should be recovered and identified at a preliminary level to order or morphotype.

The established use of haul nets is due to multiple factors, including the minimal and relatively inexpensive equipment, the relatively short time required to sample each site, and suitability for almost all bore depths.

However, the method has some limitations. For example, haul nets are most effectively used in vertical boreholes, and animals may swim away from the net, or be trapped below it, potentially resulting in taxa being missed.

2 Pumping

Pump sampling of boreholes has traditionally been used infrequently in EIA and regional biodiversity surveys in WA (Eberhard et al. 2009), but the method is used widely in eastern Australia and Europe (Hancock and Boulton 2009). Pumping is considered a supplementary method but may be a useful method in some situations. For example, pump sampling can yield additional specimens or sometimes additional taxa compared with haul nets alone, and damaged specimens can still be used for DNA comparisons. Pumps may be able to sample groundwater in holes where haul net sampling cannot be undertaken or for sampling hyporheic zones. Whether the method can be used on inclined bores is dependent on slope and construction of the bore. When pump sampling is coordinated with routine hydrology studies (e.g. bore testing) it provides an opportunity to collect additional supplementary data, including data on stygofauna in the surrounding aquifer compared with inside the borehole itself.

The use of pump sampling should consider the depth of sampling, the inlet filter size, pumping rate and the use of pumps with impellers, which can result in damage to larger specimens. There may be additional cost, logistical overheads and sampling time per site inherent in pumping compared with the use of nets, particularly in multiple phase studies.

3 Trapping

Trapping and baiting is used infrequently for stygofauna in EIA surveys in WA. It is likely that trapping methods would yield only a subset of the stygal assemblage in comparison to repeated haul net sampling. Therefore, the use of traps is considered a supplementary sampling method for application where particular stygal taxa are being targeted, or where data are required on vertical stratification in fauna distribution.

Traps are typically constructed from PVC tubing and plankton mesh, and can be deployed within a borehole or well for a period of a few hours to a few days (baited traps) or approximately one to two weeks (unbaited traps) (Hahn 2005, Bork et al. 2008).

Baiting can also be an effective method of attracting stygofauna into a haul net when deployed for a short period at the base of a well or bore. It is possible to target specific groups with other trapping designs, such as the use of traps built from mop head fibers to attract stygal snails.

4 Plankton nets

Stygofauna can occur in superficial habitats, such as in areas where the groundwater is very shallow or discharged to the surface via spring flow, or in hyporheic zones such as along riverbanks or stream beds (Boulton 2001; Halse et al. 2002). It is possible to sample these habitats with plankton nets, either via digging into stream bed shingle and waiting for the depression to fill with water (Karaman-Chappuis method) or by the use of a Bou-Rouch pump, and filtering water through the net to capture specimens (Pospisil 1993; Sket 2018). Care should be taken when sorting samples as these habitats may include epigeal and hyporheic specimens.

Plankton nets can be used to directly sample spring outflows, but settings where this technique can be used are uncommon. Only a subset of the assemblage of an area is typically collected from outflow sampling, limiting the adequacy with which these methods characterise the fauna. Given these limitations, outflow sampling is considered a supplementary method.

5 Environmental DNA

Several studies have successfully used environmental DNA (eDNA) samples to detect the presence of aquatic fauna in marine, estuarine and freshwater surface systems (Lodge et al. 2012, Minamoto et al. 2012, Thomsen et al. 2012, White et al. 2020), and the techniques have potential for application in stygofauna surveys.

Evidence is emerging that the use of eDNA approaches may be an effective method for improved species detection in aquatic environments, particularly for elusive or hard to sample taxa that are difficult to detect by conventional sampling methods (Bohmann et al. 2014). Detection of species using eDNA from bulk environmental samples may improve general biodiversity assessments (e.g. Minamoto et al. 2012) provided sufficient reference sequences exist to positively identify all taxa detected in a sample (known as 'eDNA barcoding'). When used in conjunction with conventional sampling methods, it may also help with the detection of very low abundance taxa and improve estimates of total species diversity in a system.

Different eDNA techniques can be used in a targeted approach to collect data on the distribution and habitat of more poorly-known taxa or those of formally recognised conservation significance, and techniques are continually being improved and updated. Appropriate sampling control and storage, in the field and lab, is essential to avoid contamination and degradation of samples. For example, water samples must be kept chilled and should be filtered on-site, or within six to eight hours of collection, to maintain DNA quality.

While eDNA techniques have good potential for application to stygofauna surveys, these techniques still need proof of concept, and require suitable reference sequence databases to be truly effective, especially for monitoring. Given these considerations, the use of such molecular techniques is considered to be in development and qualified practitioners in eDNA sampling and analysis should be consulted prior to undertaking such work.

6.2 Troglifauna

Four troglifauna sampling methods have been identified as having applicability for use in EIA, with the two primary methods trapping and haul net sampling. It is recommended that both techniques are used at the same site during each sampling round as each method collects different communities of troglifauna (Halse and Pearson 2014; WABSI 2021). Manual collecting of troglifauna and the use of environmental DNA techniques, are considered to be supplementary methods or only applicable in certain situations. When sampling for troglifauna is undertaken in conjunction with haul net sampling for stygofauna, the nets should also be checked for troglifauna and any bycatch added to the sample for that site.

1 Trapping

The most widely used method of sampling troglifauna for EIA in WA is the use of custom-built PVC traps, suspended within sampling sites (drillholes), and baited to act as both an attractant and colonisation medium. The most common bait used within the traps is leaf litter and organic material collected from the surface, which is soaked in water and sterilised in a microwave prior to loading into the traps.

Trap effort per sampling site will vary depending on the depth of the drillhole, depth of suitable habitat and variation in habitat strata, taking into account the site-specific information available. At least one trap per site sampled should be used, noting that the use of multiple traps is likely to increase the number of specimen captures. Multiple traps should be deployed where haul net scraping method is unable to be used (e.g. in angled bores).

Traps should be placed between the lower limit of the surface soil layer and the watertable, recording the depth that traps are set in relation to drillhole depth or watertable level. In situations where adequate stratigraphic data exist, trap deployment can be designed to target specific habitat strata, which may result in improved habitat and species distribution information.

Maintaining a moist environment around the trap is likely to increase trapping success. Making sure the bore is capped and sealing around the cap with plastic will help maintain humidity.

No systematic research has been published to date that unequivocally demonstrates an optimal period for trap deployment, but past sampling suggests fauna are reliably collected following a minimum six to eight weeks deployment. Shorter sampling periods tend to result in lower recording rates and a higher frequency of juvenile animals, and longer trap deployment may result in reduced specimen collections from predation within the trap microhabitat and reduced bait effectiveness.

Traps should be examined on removal from the sampling site as specimens can be opportunistically collected from the exterior of the traps or from plant root matter that may be brought to the surface. Traps should be wrapped in absorbent material, such as paper towel, to prevent condensation and individually stored in sealed and appropriately labelled zip-lock bags or similar packaging for transport to a laboratory for sorting. This maintains humidity within the leaf litter and prevents the movement of animals between samples collected from different sites during transit.

Samples should be kept cool, but not chilled, to minimise the risk of degradation or loss of specimens due to heat or desiccation.

Tullgren, or Berlese, funnel arrays are typically used in the initial recovery of animals from trap samples. A heat gradient is generated by these devices that results in the downward migration of invertebrates present in the samples into a catch bottle filled with 100% ethanol. The resultant bulked samples are then sorted under a dissecting microscope to recover any troglomorphic specimens and allocated to orders and morphotypes. When using Tullgren funnels, it is recommended that leaf litter samples are checked for live and dead specimens both prior to and post sorting.

Manual sorting of leaf litter samples by hand is not recommended as there is a risk of missing small or cryptic animals and it is labour intensive.

2 Haul net scraping (scrape sampling)

Haul net scraping is an effective primary sampling method for troglofauna (Halse and Pearson 2014). When sampling troglofauna using haul nets, a reinforced stygofauna sampling net, sometimes using a modified scraping attachment above each net to dislodge specimens, is lowered to the bottom of the sampling site and gradually drawn to the surface while dragging the net along the interior surface of the drillhole (Subterranean Ecology 2011; Halse and Pearson 2014). This is repeated a minimum of four times to provide coverage around the circumference of the drillhole and improve the likelihood of dislodging any animals present.

Live or obvious animals in the sample can be recovered and preserved on-site into 100% ethanol. Nets should be examined on removal from the sampling site for animals opportunistically collected from the exterior of the device. The remainder of the collected sample, including any plant root material, sediment or organic matter, should be preserved with chilled 100% ethanol, which should be periodically replaced, and kept cool for transport to a laboratory. Samples are then examined under a dissecting microscope to recover any troglomorphic specimens and sorted by order or morphotype. Samples may also be sieved to improve sorting efficiency (Halse and Pearson 2014).

Troglofauna yields from haul net samples may be reduced by site-specific factors such as drilling method and casing specifications, and the resultant nature of the interior surface of the sampling site. However, in most cases using haul nets in combination with trapping will yield additional specimens and taxa compared with trapping alone. Haul nets yield samples immediately, compared with the deployment period required for trapping, and can be a time-efficient method, particularly when used

as a component of basic surveys.

3 Direct collecting

Troglobitic animals can be collected manually, including the use of hand-nets, baits and pooters. This method can be used in surveys of cave systems (Humphreys 1993, 2001, Culver and Sket 2000). However, direct collecting of troglofauna in this way is only possible in cave settings that are of sufficient size for human entry, so is unlikely to be appropriate in the majority of EIA surveys.

4 Environmental DNA

There is the potential for DNA from troglofauna to be amplified from environmental samples collected from suitable habitat, such as bulk trap and scrape samples. Currently, the use of eDNA techniques for troglofauna has not been experimentally tested and the method requires considerable refinement to be applied as a reliable method for EIA.

6.3 Sample tracking

Knowing the collection location for records of subterranean fauna is critical to inform assessment. Sample and specimen tracking must be accurately maintained through all stages of field survey, sorting, preservation, identification and lodgment into collections. It is the responsibility of the survey leader to ensure that an appropriate tracking system is in place including chain of custody records, such as WA Museum lodgment forms or where specimens are transferred to specialists or third parties e.g. for sequencing.

6.4 Data collection

Data collected during the survey should be adequate to inform assessment and to enable analysis and comparison of results between surveys and study areas. Collecting data consistently during EIA surveys will increase the understanding of subterranean fauna ecology and distributions (WABSI 2021). Raw data should capture the parameters for each unique sample.

Examples of the survey parameters that should be recorded include:

- company, personnel, project or proposal name or ore body
- sampling phase, survey start and finish dates, and sampling date
- site number or code, site location (e.g. latitude/longitude; datum) and description (e.g. impact or reference)
- sampling method used, number of traps and/or scrapes per site/sample, deployment duration, sample field number, bore description, bore depth, depth to watertable, trap or sample depth
- preservation method, and environmental variables recorded at the time of sampling such as temperature, humidity, and water chemistry
- limitations (access, bore construction, trap loss).

Examples of the parameters that should be recorded during sample examination include:

- specimen number life stage, lowest taxon identifier, number of individuals
- whether the specimen is determined to be stygofauna or troglofauna (or other)
- likelihood of short-range endemism
- name of the person making the identification, identification method used and relevant genetic reference, and confidence or reliability of the identification.

These parameters should be consolidated with general survey and site data such as weather and climate, geological and hydrological information, bore construction information, and baseline environmental variables (WABSI 2021).

Information from surveys should be made publicly available, including via IBSA, to improve the efficiency and timeliness of the assessment and approval process (EPA 2020).

7 Survey design

Appropriate survey design is critical in EIA and is determined by factors including the survey type, the objectives of the survey, the scale of the proposal, the local environment and the faunal assemblages expected.

Adequate effort must be applied during surveys to enable the assessment of impacts of the proposal on fauna and habitats. Survey adequacy is a function of the techniques, number of samples, site selection, survey duration, number of phases, and seasonality. The rationale for the chosen survey design should be clear.

7.1 Site selection

Sampling sites usually comprise of boreholes for stygofauna and drillholes for troglifauna. The selection of sites will depend on the habitat characteristics of the study area and its surrounds, such as the type and variety of substrates; topography and stratigraphy, geographic extent; and the degree of similarity between habitats within an area or any changes in habitat that may represent barriers to dispersal or isolated areas that may contain different species or assemblages.

Selection of sampling sites should consider the most accurate information available on: the local geological and hydrological setting, such as extent of relevant aquifers; and the scale and level of the direct impacts predicted as a result of the proposal, such as groundwater drawdown from dewatering, extent of proposed mine pits; and any other characteristics of the proposal relevant to subterranean fauna habitat.

For detailed surveys, sampling sites should be selected across the spatial extent of the proposed areas of impact and should include sampling in relevant reference (non-impact) areas, with the primary objective to adequately sample subterranean fauna habitats. Sampling in relevant reference areas provides a better understanding of the wider distribution of the taxa present in impact areas and as a means of validating theoretical habitat models. Sampling of reference sites includes habitats equivalent to those within the impact area, particularly where there are no obvious major geological or landform barriers separating them.

Site selection for targeted surveys will depend on the knowledge gaps in question and will usually be focused on sampling sites within specific habitats or that may host a particular taxon, or sites within unsampled areas of a proposal to resolve sampling gaps. For example, targeted survey sites may include repeated sampling of the same sites (e.g. for confirmation or monitoring) or increased sampling of reference sites to support inferences around habitat connectivity.

A range of factors can affect sampling efficiency from boreholes, including construction specification, time since last recharge event, bore location, depth to watertable, site accessibility in the field, and ecological variability inherent in the sampling methods themselves. These factors mean that some proportion of sites sampled do not yield fauna, even in relatively species-rich areas, and are not useful for EIA.

The selection of individual sampling sites should take account of the installation method, and the specifications of any casing, screen and gravel packs installed. Boreholes or drillholes have, in most cases, been installed for other reasons (e.g. mineral exploration, groundwater abstraction or watertable monitoring). This can mean that the drilling method or construction specification may limit their effectiveness as a sampling site for subterranean fauna. Bores that are blank-cased through potential habitat strata and only screened at depth in non-target aquifers should be avoided. The sites selected for sampling should be the best available, aiming to minimise construction or other artefacts that may compromise the adequacy of the survey.

7.2 Sampling effort

Sampling effort should be determined when planning the initial survey design. The sampling effort should be sufficient to adequately characterise the faunal assemblage of the proposal area present at the time of sampling.

Repeat sampling is required for detailed surveys. Depending on the survey objective, repeat sampling may also be required for targeted surveys or for monitoring.

The survey effort should be reviewed following completion of the sampling, based on the habitat and taxa information recorded.

7.2.1 Stygofauna

Sampling effort for detailed surveys for stygofauna requires consideration of:

Scale of the impact: Impact areas (e.g. predicted extent of groundwater drawdown) that are small relative to the size of the remaining suitable habitat for stygofauna, considering both the vertical and lateral extent of aquifers, and only intersect a single aquifer system are likely to require a lower sampling effort. However, large impact areas, such as proposals that will affect a high proportion of the available habitat or more than one aquifer, including cumulative impacts, will require sampling a greater number of sites to adequately characterise the fauna and habitats.

Time since borehole construction: the suitability of recently drilled boreholes for stygofauna sampling is likely to be based on the nature of the geology, groundwater flow rate, and the suitability of borehole construction specifications and methods used. In high-flow karst settings, stygofauna specimens, including stygal vertebrates, have been collected from bores drilled less than a month prior to sampling. In low transmissivity settings, or where bore development has been poor or construction specifications less appropriate, a longer period of several months may be required before stygofauna can reliably be sampled.

Given this variability, the above considerations must be taken into account when planning the survey to ensure adequate time has elapsed since borehole completion. Generally, sampling six months after borehole construction provides time for stygofauna to colonise new boreholes and has been shown to reliably record stygofauna. In the event that sampling of likely stygofauna habitats is conducted within six months of bore completion, and no or few stygofauna are collected, then repeat sampling would be required.

Number of sampling phases: It is typical for new taxa to be recorded from multiple sampling phases, with species accumulation increasing with the number of sampling rounds. In areas where there is suitable habitat and a reasonable expectation of encountering stygofauna, a minimum of three phases of sampling is expected for detailed surveys. Sampling phases should be approximately three months apart to allow sufficient time between sampling for other fauna to enter the borehole, and to capture changes in aquifer conditions. Undertaking a minimum of three phases of sampling aims to account for any seasonal or other artefacts where nil or reduced captures are recorded. Based on the outcomes of the initial three phases of survey, additional sampling phases through targeted surveys may be required to inform the assessment where uncertainty or knowledge gaps remain.

Sites that are sampled repeatedly over multiple phases will provide a more adequate characterisation of the stygal assemblage present and record changes over time and take into account possible seasonal changes in taxa abundance and distributions. Multiple sampling phases also provide the opportunity to sample more broadly in the study area, particularly for large proposals, or to sample other prospective habitats.

Number of sampling sites: The number of sites sampled should be based on the scale of impact; the extent and variation of the prospective habitat including changes in water chemistry parameters; and the number, type (e.g. mineral exploration or hydrogeological bores) and location of the bores available for sampling (e.g. mine pits and borefields).

For detailed surveys, a minimum sampling effort of at least 10 impact sites is recommended. Generally, the larger the impact area or where there is habitat variation across the proposal area, a higher number of bores would be expected to be sampled. In addition, a greater sampling effort would be expected where the desktop study or baseline surveys suggests that the area may have significant stygofauna values.

Where possible, it is recommended that at least as many samples are taken in reference areas, selected outside of the impact area, as those within and that the reference habitats sampled should be similar to those within the impact area. The selection of reference sites for stygofauna should consider areas that are hydrologically connected, geologically similar, and within the same aquifer system or local surface water catchment as the impact. For example, stygofauna reference sites should include those within the same aquifer, but outside of the area of predicted groundwater drawdown to provide context for habitat suitability and extent beyond the impact zone.

Seasonality: Ideally, one of the sampling phases should be timed to occur following significant rainfall in the catchment and aquifer recharge. While there are no quantified relationships between major recharge and stygofauna sampling yields, anecdotal information and some studies suggest these events may result in improved specimen yields or records of taxa not collected during previous sampling of the same site. For example, sampling in January in the Pilbara may yield more diverse assemblages of stygofauna than other months (WABSI 2021); and studies in the Yilgarn suggest that rainfall may influence stygofauna species composition and abundance in calcretes, through changes in the watertable and introduction of nutrients (Hyde et al. 2018).

If one of the sampling phases is conducted following dry conditions - where there has been little or no rain for a period of more than three months - and no stygofauna are recorded in suitable habitat, additional sampling would be expected and the results and limitations would need to be discussed.

7.2.2 Troglifauna

Sampling effort for detailed surveys for troglifauna requires consideration of:

Scale of the impact: Impact areas (e.g. predicted extent of mineral extraction) that are small relative to the size of the remaining suitable and connected habitat for troglifauna, are likely to require a lower sampling effort. However, large impact areas such as proposals that will affect a high proportion of the available habitat, or impacts over multiple areas, or that may fragment connected habitat, including cumulative impacts, will require sampling a greater number of sites to adequately characterise the fauna and habitats.

Time since borehole installation: Troglifauna sampling may not be affected by some of the drilling artefacts associated with stygofauna sampling (e.g. slow groundwater flow rates and high turbidity), but the drilling process still disturbs the environment above the watertable. Drilling fluids may reduce the adequacy of a sampling site in the short term, particularly for haul net sampling. However, traps deployed in uncased bores one month after drilling have reliably yielded troglifauna. Therefore, a minimum period of one month after completion of drilling is recommended before sampling. If sampling of suitable habitats is conducted less than one month from drillhole completion, and no specimens are collected, then the sampling phase would not be considered adequate and additional sampling would be required.

Number of sampling phases: It is typical for new taxa to be recorded from multiple sampling phases, with species accumulation increasing with the number of sampling rounds. In areas where there is suitable habitat and a reasonable expectation of encountering troglofauna, a minimum of three phases of sampling is expected for detailed surveys.

Sampling phases should be approximately three months apart to allow sufficient time between sampling for other fauna to enter the drillhole, and to capture any changes in conditions. Undertaking a minimum of three phases of sampling aims to account for any seasonal or other artefacts where nil or reduced captures are recorded. Additional sampling phases through targeted surveys may be required to inform the assessment where uncertainty or knowledge gaps remain.

Sites that are sampled repeatedly over multiple phases will provide a more adequate characterisation of the assemblage present and record changes over time.

Number of sampling sites: The number of sites sampled should be based on the scale of impact areas; the extent and variation of the prospective habitat including geology; and the number, type (e.g. mineral exploration or hydrogeological bores) and location of the sites available for sampling (e.g. mine pits and borefields including groundwater reinjection).

Considerably more sites need to be sampled and more samples are required to be collected for troglofauna than stygofauna (WABSI 2021). Therefore, for detailed surveys at least 30 impact sites should be sampled. Generally, the larger the impact area or where there is habitat variation across the proposal area, a higher number of bores would be expected to be sampled. In addition, a greater sampling effort would be expected where the desktop study or baseline surveys suggest the area may have significant troglofauna values.

Where possible, it is recommended that at least as many samples are taken in reference areas, outside of the impact area, as within it, and that the reference habitats sampled should be similar to those within the impact area. The selection of reference sites should take into account the likelihood that species turnover across the landscape is very high, particularly in troglofauna, and recognise that areas isolated from the proposal impact areas are unlikely to support the same suite of species.

Each site should be sampled using both trapping and scraping methods, depending on the suitability of the bore type and construction. Combining both methods will increase the effectiveness of sampling and contribute to incremental knowledge gain on the applicability of the two primary sampling methods in different settings (WABSI 2021).

Seasonality: While there are few data demonstrating seasonality in troglofauna, anecdotal evidence and some studies suggests that troglofauna sampling results are generally greater following periods of substantial rainfall. For example, sampling in March in the Pilbara may yield more diverse assemblages of troglofauna than other months (WABSI 2021). This may be a function of the influx of water and nutrients to subterranean environments that occurs during recharge events. Conversely, areas of known troglofauna habitat have yielded little or no fauna after lengthy periods without rain, despite adequate sampling. For example, sampling in October and November in the Pilbara may contribute to high community distributions of troglofauna (WABSI 2021).

If sampling is conducted under dry conditions, where there has been little or no rain for a period of more than three months, and no troglofauna are recorded in suitable habitat, additional sampling would be expected or the results and limitations would need to be discussed.

8 Habitat connectivity

Determining whether there is habitat connectivity between impact and reference areas requires an evaluation of the homogeneity of the geological, hydrological and hydrogeological features, including evidence of barriers that may isolate species and prevent species from dispersal, and consideration of the morphological characteristics, dispersal attributes and distribution of widespread species in relation to these features.

The interpretation of habitat connectivity is only appropriate in situations where adequate sampling has already been completed and the data suggest taxa may be restricted to impact areas.

Subterranean fauna surveys commonly record many taxa represented by only one (referred to as singleton taxa) or a few specimens (see Figure 2). Some taxa may be in high abundance within relatively small ranges, others may be found at multiple sites but at low densities or are difficult to sample using current methods and it can be challenging to determine how widely distributed a taxon may be where many taxa are represented in sampling data sets by few or single records. Therefore, it can be difficult to distinguish those taxa that are truly rare and restricted in distribution, from those where the effects of sampling have created the appearance of extreme short-range endemism.

Determining whether the record represents a population of a taxon that is restricted to the proposal impact area includes evaluating the likelihood of physical and biological connectivity.

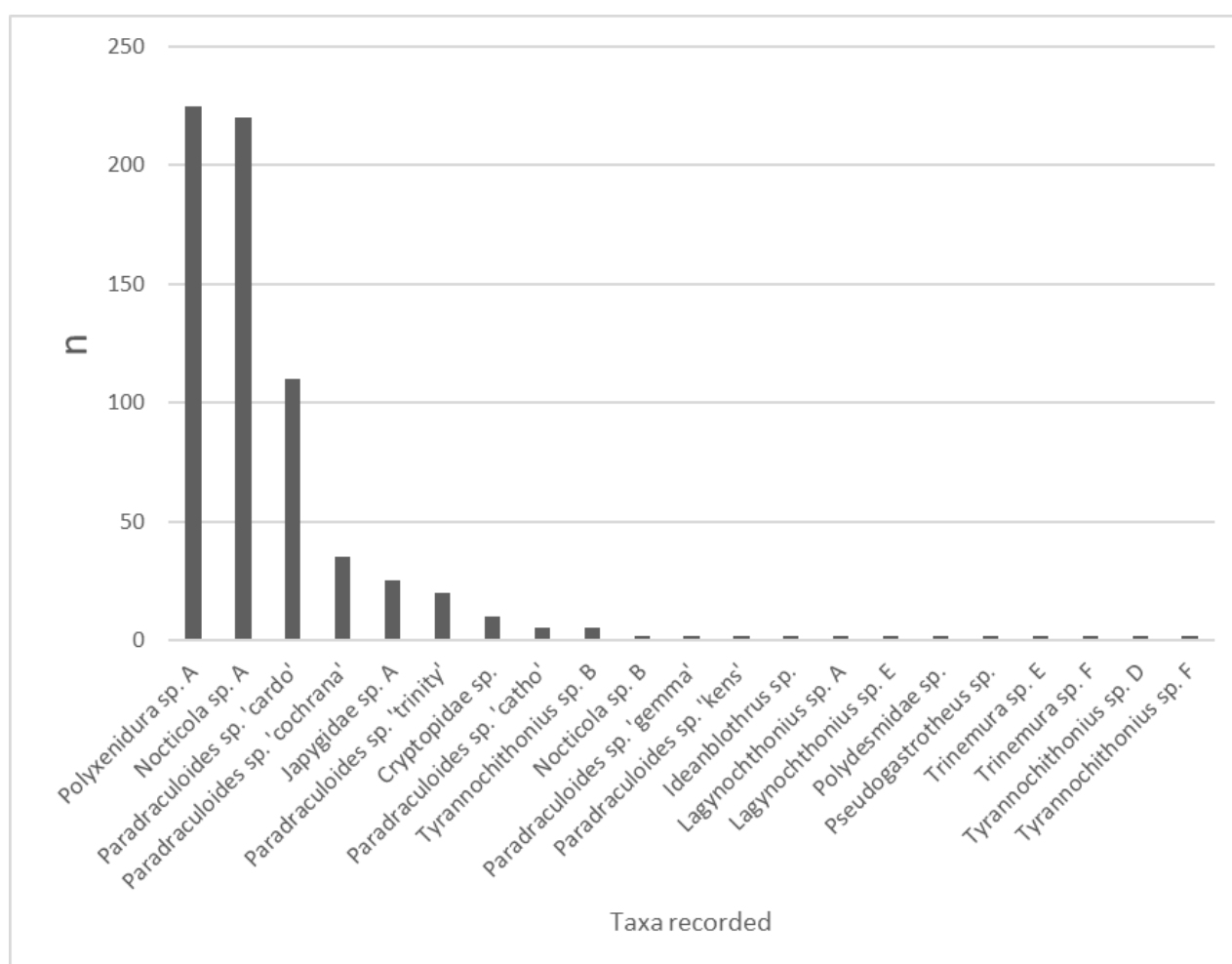


Figure 2: Example subterranean fauna rank-abundance plot from a Western Australian EIA survey (n=number of specimens of each taxon)

8.1 Physical connectivity

Demonstrating the relationship between impact and reference areas increases confidence that habitat is connected and that singleton taxa are likely to have a wider distribution. Geological and/or hydrogeological data can be assessed to determine if there is physical evidence for potential habitat connectivity.

For example, if a singleton specimen is recorded from a habitat that is extensive and well-connected to equivalent habitat occurring more widely outside the impact area, then, by inference, the population of that species might also occur more widely. Conversely, if the singleton record comes from an area of habitat that is isolated by significant geological features that typically represent barriers to subterranean fauna movement, and thereby gene flow, then the likelihood that the taxon is truly restricted to the area is considerably higher.

The broadest data set available is regional geological mapping which may not be sufficient to provide a sound basis to demonstrate habitat connectivity. Finer scale geological mapping, specific to the proposal area is preferred and usually provides a more spatially accurate framework to evaluate connectivity.

However, detailed surface geology mapping is only of value if the mapped units continue with depth and are representative of the subterranean fauna habitat. Where different underlying stratigraphic units characterise the habitat, the use of drillhole log data, geological cross-sections, 3D modelling (e.g. Leapfrog), core samples and specific mapping of these subsurface strata will be required to assess the level of habitat connectivity. The presence of faults, dykes or other structural features, particularly major changes in geomorphology, should also be considered in relation to their influence on species distribution.

Hydrogeological studies in the proposal area should be used to determine connectivity; particularly for stygofauna or where pump testing and other assessments of transmissivity and aquifer connections have been completed. Any known or inferred aquicludes that may represent habitat discontinuities should be noted.

The physical attributes of the habitat should be considered in parallel to the biological attributes of a taxon.

8.2 Biological connectivity

Where suitable data exists, evidence for habitat connectivity may be further informed by the biological and ecological attributes of a specimen or taxon.

Singleton records can be compared to other members of the same taxonomic group, taking into account their morphology, size and mobility, known distributions and habitat characteristics of other members of the same group, and whether that group is known to exhibit short-range endemism. For example, if many taxa of the same genus are known to be short-range endemics, and the majority of other taxa recorded from the proposal area appear to be restricted, this may indicate that the taxon in question is also restricted and likely to be a short-range endemic.

The distributions of other specimens collected from the same sampling site can also be considered in relation to the proposal area and their wider regional distribution. For example, if several other taxa have been recorded from the same sampling site and all show wider distributions, then this may suggest that the singleton record from that site in question is also unlikely to be restricted in distribution at the same scale.

Inferences may also be made by considering genetic data in combination with morphological evidence.

To demonstrate genetic continuity between two areas, measures of the level of local gene flow between other members of the same taxonomic group can also be considered. Multiple lines of genetic evidence are required and interpretation of genetic information to infer connectivity or gene flow should be undertaken by qualified persons with experience in genetic analysis for subterranean fauna.

There are limitations to making inferences on biological connectivity, as some species within a genus can be widespread, while other congeners may be short-range endemics. Therefore, it is not acceptable to infer the likelihood of a taxon being restricted on the basis of biological connectivity alone. Habitat connectivity based on physical, biological, ecological and genetic data must also be considered.

8.3 Demonstrating adequate consideration of habitat connectivity

Habitat connectivity should be supported by demonstrating that the information has been adequately considered, this includes:

- presentation of suitable physical evidence such as drillhole logs and core samples, hydrogeological or geological cross-sections, mapping of subsurface geology and groundwater levels, 3D modelling (e.g. Leapfrog), groundwater physiochemical data, and the known extent of aquifers or hydrated zones
- identification and mapping of any potential barriers to dispersal e.g. aquitards, faults, dykes, changes in geology or water chemistry (see Table 1 and Table 2)
- use of appropriate examples of widespread species i.e. those that:
 - have similar biological and ecological attributes to the singleton species in question
 - have been recorded from the same site and multiple sites within the study area, or adjacent areas
 - are not taxa that are known to have broad regional or cross-regional ranges
- maps or figures illustrating the locations of widespread species, between impact and reference areas, in relation to the habitat and proposed impact areas
- state the taxa recorded within the study area, the habitat they were recorded from and the maximum geographic distances between recorded locations or known distribution
- genetic analysis to demonstrate geneflow within the study area or between sites.

9 Specimens

9.1 Identification

All specimens collected should be identified to the lowest taxonomic level possible and assigned to the described species, where known, or the equivalent morphotaxa or taxonomic code available.

Taxonomic frameworks for many subterranean groups in WA are relatively poorly resolved and diagnostic tools are not available for all groups. Due to limited regional-scale surveys across much of WA and inherent difficulties in sampling for subterranean fauna, a specimen often represents an undescribed species.

The most appropriate techniques and tools for the group in question should be used to assign specimen identifications and resolve taxonomic uncertainty. Robust analysis of putative species boundaries is gained from the combined consideration of molecular data and morphological diagnosis, using suitable reference material available.

9.1.1 Morphological

Conventional specimen identification methods rely on morphological characteristics diagnostic of the relevant taxonomic group. For most taxa, this requires specimens to be dissected, slide-mounted or examined under a high-resolution microscope. Diagnostic characteristics are scored against relevant keys, where these are available, to assign the specimen to a species.

Intact adult specimens of a particular sex are often required for morphological species determination for many taxa. A common limitation with morphological identification is that relatively high proportions of juvenile and/or damaged specimens are often collected, meaning they cannot be identified based on morphology even if keys and suitable taxonomic frameworks exist.

Specimens should be compared with the appropriate material from the region e.g. specimens held at the WA Museum or other reference collections, where accessible.

Species-level morphological identifications should only be completed by zoologists familiar with the fauna groups in question. Identification reports should include the name of the person making the identifications, the methods used, such as what morphological characteristics were used to determine the ID, and what specific keys or reference collections were used.

9.1.2 Genetics

The use of molecular tools to identify specimens is a valuable technique in EIA and offers additional benefits to using morphology in isolation.

Specimens, including juvenile, partial and damaged specimens not useful for morphological identification, may be suitable for genetic identification, subject to specimen condition and adequate preservation of DNA. Tissue sampling for molecular analysis should follow the WA Museum Taxonomic Services Submission Guidelines (WA Museum 2018) for the relevant taxonomic group to ensure specimen sub-sampling does not compromise morphological diagnosis.

Each specimen sequenced should have a unique identifier or number and be kept separated from other specimens i.e. in an individual vial, to maintain the link between the specimen and its sequence data.

Genetic studies should be designed by practitioners with the appropriate expertise, and analyses should use standard peer-reviewed methods. Analyses should be reported in detail including methods such as genes sequenced (e.g. cytochrome oxidase 1 (CO1) or 16S and 12S), primers used, programs

and algorithms used to align sequence data and build phylogenetic trees, and the reference library used (e.g. WA Museum; GenBank) should be stated.

A variety of high-throughput molecular methods, usually loosely referred to as 'Next Generation' sequencing, are now available. Next Generation sequencing offers a powerful method of completing phylogenetic analysis, but the method is often expensive and requires specialist expertise and knowledge. Proponents and consultants who are considering the use of Next Generation techniques for EIA should seek advice from qualified experts on the design and methods to be used.

Any third-party reports outlining the specimens and methods used for molecular analysis should be provided as an appendix to the survey report.

9.2 Nomenclature

All data and reporting on surveys for EIA should follow current published taxonomic nomenclature and taxonomic hierarchies for specific groups, such as those available from the WA Museum, with the authority and publication cited in the report.

For taxa that have not yet been formally described or named in the scientific literature, the use of the WA Museum alpha-numeric code system is preferred for those groups where a system exists e.g. schizomids, millipedes, pseudoscorpions. This code system provides a consistent taxonomic framework and enables a taxon, whether named or not, to be placed in a regional context. For those groups where there is no WA Museum alpha-numeric code, phrase names and morphological-codes should be consistently applied by the practitioner and include relevant descriptors, such as taxonomic group, consultancy name or project name. Codes should be unique for each identifiable taxon with no duplication of names, and be traceable e.g. linked to the field number, WA Museum registration number and GenBank number.

Where specimen identifications have changed over time, such as updated taxonomy, earlier names or codes should be listed in the report to allow the taxonomic history of the specimen to be tracked and enable alignment of future identifications.

9.3 Specimen vouchering and lodgment

Specimens collected must be offered to the WA Museum for lodgment into the State collections, in particular those specimens that represent a potential newly identified taxon or range extension, or where new codes have been assigned to an undescribed taxon. Genetic data, such as sequence data, barcoding genes, or phylogenetic trees, should be submitted to a publicly available database (e.g. GenBank, BOLD or Treebase).

The lodgment of specimens is essential to incremental knowledge gain in Western Australia. This enables ongoing taxonomic revisions; refinements to the understanding of taxa distributions, including the alignment of equivalent taxa separately recognised in independent surveys; and overall knowledge of subterranean biodiversity of the bioregions.

Evidence of submission of data and accession of specimens to the WA Museum should be provided with the report (e.g. registration numbers, chain of custody). Registered specimens should be provided to the WA Museum for lodgment within six months of completion of the EPA's report on a project.

The WA Museum Taxonomic Services Submission Guidelines (WA Museum 2018) outlines the requirements for specimen preservation, labelling and data submission, including preservation of genetic material.

10 Data analysis

Considering data analysis prior to conducting field surveys will aid in development of an appropriate survey design. It is important to ensure that the analyses and data presentation:

- are commensurate with the type of survey or study
- provide evidence that the work was conducted in accordance with EPA guidance , or adequately justifies deviations from the guidance
- are sufficient to allow robust assessment of the impacts.

The types of analyses should be appropriate for the data available. For example, desktop studies may simply include species lists, whereas reports for detailed surveys may include species-by-site and species-by-habitat matrices. Analyses should incorporate abundance information whenever suitable data have been collected. Summary statistics can include species richness and/or diversity and similarity matrices. Care should be taken to ensure that the underlying assumptions of analyses are valid.

This guidance does not cover all possible analyses for the range of survey types used in EIA. Ecological data analysis methods evolve continuously, and it is the survey leader's responsibility to ensure that the analyses are suitable. The efficacy of any novel analysis or models used, e.g. for prediction of habitat, should be demonstrated. Involving biostatisticians in the design and analysis of surveys and monitoring programs can be beneficial.

10.1 Assessment of the reliability and veracity of data

The reliability of data should be critically evaluated. For example, an absence of records of a species does not necessarily indicate the absence of that species and may instead be due to information gaps. Information gaps may be:

- spatial – if some areas have been better surveyed than others (e.g. due to access restrictions or location of sites within the project footprint)
- taxonomic – if surveys or methodologies used focused on one subterranean fauna group
- ecological – if surveys omitted some habitat types or failed to account for species rarity or temporal variation in abundance or distribution.


Checking data for errors such as misspellings of taxa names, incorrect identifications or changes of identifications between reports, and erroneous data entry is essential before analysis.

10.2 Assessment of survey effectiveness

Survey adequacy should be determined by demonstrating that the sampling effort undertaken:

- is reasonable to predict the species richness and assemblage present
- can confidently predict the distributions of taxa, particularly when concluding that species are likely to be found outside the areas of impact.

Plotting the cumulative number of species recorded against effort, either as trapping effort or individuals, provides a species accumulation curve. Species accumulation curves, and richness estimators, can be useful in estimating total species richness and the proportion of species caught during a survey. Programs used are based variously on the functions of the number of species in only one or two samples, e.g. Chao 2 and Jackknife, the number of species with only one or two individuals across all samples, e.g. Chao 1, or the proportions of samples that contain each species,



e.g. Bootstrap (Chao et al. 2005, Green et al. 2009, Magurran 2004). As effort increases, an under-surveyed assemblage will continue to show a rapid rise in the number of species, while a well-surveyed assemblage will have fewer new species added and the curve will begin to level out. Due to the difficult nature of sampling for subterranean fauna it is unlikely that species curves will reach asymptote or level out, even after substantial survey effort applied. Therefore, the robustness of the information available to inform the assessment of impacts from the proposal should also be considered.

For the purposes of determining survey adequacy for EIA, the following information should be considered:

- the desktop study used contemporary and appropriate datasets and sources of information relevant for the proposal area
- the surveys follow contemporary guidance
- the surveys are reflective of the scale and nature of the impacts of the current proposal
- targeted surveys have been used to increase sampling effort where early surveys were deficient
- all relevant impact areas have been sufficiently surveyed with a reasonable coverage and number of sites relative to the size of the impact areas and the habitats
- the number and location of reference sites surveyed are adequate to provide context outside of the impact areas
- knowledge gaps have been investigated and outstanding issues, such as the application of genetics to resolve unidentified specimens or the use of targeted surveys to increase confidence in the predictions of species distributions
- the distribution of singleton taxa can be confidently predicted using appropriate physical and biological evidence
- the outcomes of the survey reflect the assumptions of habitat prospectivity, or where there are differences, the limitations and any reasons for them have been discussed.

10.3 Data retention

All raw data collected during surveys (e.g. dates, locations, specimen records, habitat details) should be retained in the form it was originally collected. Derived datasets and analysis outputs should also be retained. This ensures that subsequent surveys can be adequately designed, survey limitations are transparent to data users and the surveys themselves are verifiable and auditable.

11 Mapping

Reports should contain maps that adequately illustrate the existing environment, key survey information, and support the interpretations and conclusions of the studies and surveys.

Clear and appropriately scaled maps should be used to show the distributions of taxa and/or assemblages, placing these into spatial context in relation to the known and inferred habitat mapping appropriate to the fauna group illustrated.

Maps can be used to present the following information, where relevant:

- the extent of the study area in a regional context (e.g. major roads, rail, Local Government Area boundaries)
- IBRA bioregion and subregion, land systems, soil and geological mapping, hydrology and water chemistry, significant habitat features (e.g. aquifers, aquicludes, fractures, faults, shear zones)
- the proposal area (development envelope), indicative footprint and areas of impact (i.e. groundwater drawdown) in relation to the local and regional fauna values
- the extent of the desktop study database search area, and locations of previous surveys included in the literature review or otherwise discussed
- locations of regional fauna records based on the desktop study, relative to the proposal area
- the locations of sampling sites in relation to the extent of geological and hydrological features and the proposal, including sampling sites that recorded null results and with distinction between stygofauna and troglofauna
- the locations of fauna records from the current survey, in relation to the extent of geological and hydrological features and the proposal, and identification of the locations of rare (e.g. singletons) or significant fauna.

The above information should be presented over multiple figures in a report. However, information can be combined where it is appropriate, such as to show comparisons between surveys, provided figures remain legible and are at a reasonable scale.

Maps should be legible and include the most current information. Aerial imagery should be the base layer for most maps with the subject of the map overlaid with transparent colours and labelled features. Colours of features and/or shapes of symbols should be readily distinguishable from one another. The colours or textures used to indicate recurring features (e.g. impact footprint) should be consistent for all maps within the survey report.

As a minimum, all maps should include an explanatory title, legend or labels, scale bar, north point, grid or graticules, coordinate system number, figure number and date or version. Maps should use MGA projections with the GDA2020 datum or decimal degrees, as appropriate to the scale. Maps should be north-oriented unless there is a compelling reason to do otherwise.

The scale may vary depending on the size of the study area, spatial heterogeneity of data layers and overall amount of information that needs to be displayed and presented at an appropriate scale where they are easily readable. Large map sets should also include an overview map for orientation and reference, using an inset to show the map extent relative to the whole study area on individual maps.

Insets can also be used to focus on areas with high numbers of records to improve the clarity of the data presented or highlight important areas. Alternatively, several maps can be used to illustrate the locations of species by taxonomic group. For example, where many taxa have been recorded, it is useful to present a series of maps by taxonomic group, rather than all species on one map.

12 Reporting

The structure, content and detail of the survey report should be based on the objectives of survey. The survey report should accurately reflect the information obtained through the survey, include a rational interpretation of the results, and demonstrate that contemporary techniques and guidance have been used.

Reports should be comprehensive, contain all relevant data and stand alone as the definitive source of information for a given survey. The survey results should be presented using appropriate figures, tables and summaries. The report should be prepared by a zoologist involved in planning and conducting the survey, where possible, or any significant changes made to the report by those who were not involved in the survey should be justified. Any additional data interpretation should be completed by a similarly experienced professional. Generally, stygofauna and troglofauna should be considered separately throughout the report.

12.1 Executive summary and introduction

The executive summary should be a succinct overview of the purpose of the survey objectives, techniques, key results and conclusions.

The introduction should summarise the relevant background information, including the nature and location of the proposal and key contextual data from the desktop study. It should contain a clear statement of the objectives of the survey, including specifying the type of survey, identifying the extent of the study area, and defining any terminology used that provide context to the survey.

12.2 Methods

The methods should outline the scope, phases and timing of the survey, and the sampling methods and identification techniques used. Justification for any deviation from this guidance should be provided.

The reports, publications, databases and other sources used for the desktop study should be stated. Documentation of the survey methods should include, but not be limited to, the survey dates and phases, survey level, rationale for survey design and site selection, weather prior to and during the survey, techniques used and survey effort. The survey effort should be broken down by sampling technique and group (e.g. stygofauna or troglofauna) using meaningful units (e.g. number of sites sampled and survey phase). The methods and references used for identifying specimens including the methods used for any molecular analysis should be clearly stated. Detailed descriptions of data analysis methods or any modelling methods should be provided.

Protocols followed for specimen tracking, identification and data management and analysis, should be outlined, including the reconciliation of data from the desktop study with records obtained from the survey, and any data omitted and the basis for which it was omitted.

The personnel involved in the survey should be listed and their roles, qualifications and experience should be outlined. Third parties that contributed to the report or analyses, their details and roles should also be stated.

Any survey-specific issues should be addressed in a limitations section. These may include:

- the availability and reliability of relevant contextual data and information
- competency/experience of the survey team
- scope of the survey

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- disturbance that may have affected results e.g. existing drawdown
 - sampling site artefacts that may have affected the results e.g. borehole age or casing
 - any factors reducing the certainty that species-level taxa could be determined e.g. specimen condition, historical records, taxonomic framework or DNA sequencing issues affecting the results
 - adequacy of the survey intensity and proportion of survey achieved, e.g. whether the potential habitats of the study area were adequately sampled
 - access problems
 - timing, weather and season
 - problems with data and analysis, including sampling biases.

12.3 Results

The results from the desktop study, habitat assessment and surveys should be collated and fauna data should be presented quantitatively, wherever possible. An overview should be provided summarising the significance of the fauna and habitat values within the proposal area.

Survey results should include tables and figures summarising the survey effort and captures (by site including geographic coordinates, survey phase or dates, and survey techniques used), climatic records before and during the survey.

The suitability of each habitat type to support subterranean fauna should be described, including the key characteristics of the habitat and, where available, accompanied by photographs (e.g. drill cores), maps and schematic figures of the extent of habitats. Any observations regarding the sensitivity of the habitat to specific impacts should be included.

The results should provide a summary of the fauna recorded, including the taxonomic group, family or higher rank taxa it belongs to, the number of specimens of each taxon, any listed species recorded, and known distribution. Species recorded should be discussed in a regional context, including the presence of short-range endemics or species for which the project area is at the limits of the known range, or where the record is an extension of the previously known range. The main body of the report may present fauna assemblage data in summary, with raw data provided in appendices, but provide a detailed discussion of significant species or groups.

Specimen identification including the relevant field identification, voucher specimen or WA Museum lodgment numbers should be included in the report, where appropriate. This will enable identifications to be verified and ensure that biodiversity data are adequately stored.

Data collected during the survey should be clearly differentiated from data gathered from the desktop study, and sources of information used should be clearly referenced in the report.

12.4 Discussion

A detailed discussion should be provided on the values and significance of the subterranean fauna, assemblage and habitat values identified within the study area in relation to the proposal and regional context.

The discussion should consider the adequacy of the survey and state whether the studies and surveys meet EPA guidance. The discussion should address:

- presence and distribution of any significant species recorded
- singletons and restricted taxa, likelihood of short-range endemism, and likely distributions

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- habitat suitability and extent, relative to impact areas
 - evidence of habitat connectivity, including examples of widespread taxa and any potential barriers to dispersal or habitat isolation.

The composition of the recorded assemblage, and study area locality and habitats, should be used to identify whether important subterranean communities may be present.

Where subterranean fauna were identified in the desktop study as potentially occurring in the study area, but were not recorded during the survey, the report should discuss possible reasons for the absence of fauna and justify the survey type and effort used. The influence of survey limitations on the results should also be addressed.

12.5 Conclusions

The survey report should conclude with a summary of the key findings and consider recommendations. All conclusions should be substantiated by the data and/or reference to the literature. This should include consideration of whether adequate data has been collected to ensure that the EPA objective can be met, or state whether additional information is required to inform the assessment. The influence of the survey limitations should be noted.

The conclusion should identify issues that may need to be mitigated in planning a proposal within the study area, including the actual or predicted restriction of taxa or habitats to the proposal impact area, or where further survey is required to inform an assessment.

12.6 Appendices

Appendices containing species lists should be presented in tabular format, organised taxonomically and grouped by class and family, with significant species identified. At minimum, appendices should include:

- evidence that the statutory licenses required to conduct the survey were obtained
- a complete list of all the subterranean fauna specimens recorded during the survey; the sites from which they were recorded (including site name/number; grouped by taxonomic hierarchy; number of specimens per taxon; date of collections, sampling method and sampling site locations)
- the location and co-ordinates of all sites sampled during the survey
- outputs of any relevant database search results (if not already included in the report).

Appendices should be prepared and submitted per the guidelines for reporting above. All data sources should be cited and attributed to the original author, including maps, spatial data, figures and tables copied or adapted from other sources.

If a report relies substantially on information contained within another document, such as geological or hydrogeological information, specialist taxonomic identification reports or reports on DNA sequencing and molecular analysis, then that document should be provided as an appendix. Any other substantial information that supports the main report or results should be appended.

12.7 Provision of electronic datasets

Data collected from surveys for EIA should be publicly available to support assessments. Survey reports and associated raw data must be supplied electronically via the IBSA Submissions portal. Upon submission, all reports containing field survey results should be accompanied by an electronic data package prepared according to the [Instructions for preparing data packages for IBSA](#).

13 Glossary

Aquifer	groundwater contained within an underground layer of water-bearing permeable rock or unconsolidated materials such as gravel, sand or silt. The aquifer may be confined or unconfined.
Aquiclude	a solid impermeable area that prevents the flow of groundwater from one layer to another.
Aquitard	a geological formation with low hydraulic conductivity that restricts the movement of water between aquifers.
Bioregion	see Interim Biogeographic Regionalisation of Australia (IBRA)
Calcretes	composite rock deposits formed in arid environments by groundwater evaporation which causes the cementation of superficial gravels by calcium carbonate.
Congener	another species belonging to the same genus.
Dewatering	extraction of water from an aquifer such that the watertable is lowered in part or all of the aquifer.
Endemic	restricted in known distribution to a particular geographic area. Short-range endemic (SRE) species having a distribution of less than 10 000 km ² (see Harvey 2002).
Fauna	species or assemblages of subterranean fauna, either troglofauna or stygofauna.
Groundwater	any water located below the surface of the ground. For purposes of subterranean fauna assessment it does not include surface expressions of groundwater (eg. rivers, springs, seeps).
Hyporheic zone	the area adjacent or beneath a streambed or near-stream aquifer through which water flows and where there is a mixing of groundwater and surface water.
Interim Biogeographic Regionalisation of Australia (IBRA)	categorisation of the Australian continent into regions of like geology, landform, vegetation, fauna and climate (Commonwealth of Australia 2012).
Impact area	the areas that will be or potentially be directly or indirectly disturbed or affected by the proposal activities and any adjacent indirect disturbance or impacts that may result from operation, including changes to hydrology. For a definition of impacts see EPA 2016.
Karst	an area of exposed limestone with distinctive features such as caves, caverns and sinkholes and often with underground streams.
Local and regional context	an understanding of the study area in relation to the local area or region. Local context should be considered at a scale that allows comparison of survey data and any detailed surveys found at desktop study. Regional context is considered at a broadscale, defined by existing bioregional studies.
Palaeochannel	a remnant of a stream or river cut in older rock and filled by sediments of younger overlying rock.
Playa	an ephemeral inland salt lake.
Phreatic zone	the part of an aquifer that is saturated or below the watertable.
Proposal area	the project area encompassing the development envelope and proposal footprint boundaries. See also Impact area.

Reference area	the areas that will not be directly or indirectly impacted by a proposal activity (non-impact areas), which may be inside or outside of the proposal area.
Sampling site, site	the location where a sample or specimen has been collected. Usually a single drillhole or borehole.
Short-range endemic (SRE)	for the purpose of this guidance, SREs are defined as animals that exhibit poor dispersal powers; confinement to discontinuous habitats; and have naturally small distributions of less than 10,000 km ² , after Harvey (2002).
Singleton	a species or other taxon only represented by a single specimen from a single location.
Stratigraphy	the order and relative positioning of rock strata with depth.
Study area	the boundary used for the study or surveys, which may include areas inside and outside of the proposal area.
Stygofauna	aquatic fauna which inhabit various types of groundwater.
Taxon	a taxonomic group of organisms of any rank, such as a species, genus or family. Plural taxa.
Troglofauna	air-breathing fauna which inhabit air-filled voids or caves below the ground.
Vadose zone	the zone between the terrestrial surface and the groundwater table, also referred to as the unsaturated zone.
Void	an air-filled space in rock.

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