Museum Victoria Science Reports 5: 1–10 (2002)

ISBN 0 7311 7246 9 (print); 0 7311 7259 0 (on-line)

https://doi.org/10.24199/j.mvsr.2002.05

Further studies on the Giant Gippsland Earthworm (*Megascolides australis*) population at Loch Hill, South Gippsland, Victoria.

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Abstract Van Praagh, B.D., and Hinkley, S.D. 2002. Further studies on the Giant Gippsland Earthworm (*Megascolides australis*) population at Loch Hill, South Gippsland, Victoria. *Museum Victoria Science Reports* 5: 1–12.

As part of ongoing investigations into the Giant Gippsland Earthworm population at Loch Hill, a site occurring within the vicinity of proposed roadworks for the South Gippsland Highway, further investigations into earthworm density and population structure were conducted over a four month period from February to June 2002. Preliminary investigations into release methods for worms and possible release sites for translocation near Loch Hill were also made.

Introduction

Museum Victoria was contracted by Vic Roads in October 2000 to investigate the potential impact on the Giant Gippsland Earthworm, Megascolides australis, of road and bridge construction works on the South Gippsland Highway around Loch and Bena (Van Praagh and Hinkley 2000a, b & c, 2001). During these surveys, a population of M. australis was located at Loch Hill (Van Praagh and Hinkley 2000b) (Appendix 2).

A preliminary study of the distribution, soils and biology of the Giant Gippsland Earthworm at Loch Hill was conducted between September and November 2001. A large earthworm population was found to occur over an area of approximately 2500m2. Worms appeared to be widespread within their area of distribution. Worm density within this area was very high ranging from 4.1 to 17.9 worms per m3 with an average of 8.5 worms per m3 calculated. This indicates a very high density of worms at Loch Hill. Due to the limited sample size and extended cooler weather conditions, further monitoring of the site was considered beneficial before roadworks begin to obtain more baseline information about the population including breeding, density and population structure.

Museum Victoria was further contracted (February 2002) to continue monitoring the population at Loch Hill for approximately five months (Feb to June 2002) to obtain more information about the population at Loch Hill. Objectives of the project were to to obtain more information on:

- Population structure.
- Breeding.
- Density.

Further objectives were to conduct preliminary investigations into:

- Capture and release methods for individual worms.
- Potential translocation sites at Loch Hill.
- Specific veterinary aspects of worm management (see Appendix 1).

Methods

Worm Density and Population Structure. Field work was conducted approximately every three weeks from February 13 to June 5th. Information regarding earthworm density and population structure was obtained by extensive digging of large quadrats to obtain individual specimens. Quadrats were randomly located within the area of known earthworm distribution at Loch Hill in an attempt to determine more accurate information on earthworm density and distribution within the total area occupied by worms. However, sampling was somewhat biased towards accessibility. Once the soil was exposed, the site was searched for wet burrows. Wet burrows were then followed carefully until the worm was found and part of its body exposed. Once exposed, the entire worm was slowly dug out of the burrow, using a shovel and trowel. Individual worms were then measured and weighed and their reproductive status recorded. It is difficult to obtain an accurate estimate of worm size due to the worms' ability to expand and contract. Weight possibly gives a better comparative estimate of size, although this may vary according to amounts of soil consumed and that voided during capture. Depth of the worms was also recorded by measuring the depth in the soil when the worm was first observed. Worm density was estimated by calculating the number of worms located in a given area of soil dug.

Worms were classified into juveniles, subadult and adults based on the number and position of clitella banding (Van Praagh 1994). These small, light coloured bands occur on the ventral surface of the worm between segments xvii and xix. Juvenile worms have no external banding, subadults one or two and adults have three bands on segments xvii, xviii and xix. This information could only be obtained from worms that were fully dug up, or at least had their anterior segments exposed far enough so the clitella banding could be examined.

Unfortunately, even when great care is taken, individual worms can be killed or damaged through the excavation process. As the worms are very fragile, even bruising can result in death. Any worms injured or killed were fixed in formalin and preserved in 70% alcohol and retained in Museum Victoria collections for further research. Worms were relaxed in a mixture of 10% Magnesium sulphate and Magnesium chloride added to water, fixed in 10% formalin and stored in 70% alcohol. All uninjured worms were released at site of capture.

Not all worms sighted could be dug up. If worms were found very deep in the soil or where a large number of worms were found together, only the number and depth of the worms found was recorded as the risk of injury to other worms was considered to outweigh the successful capture and release of a worm.

Collected egg cocoons were taken back to the laboratory and kept in a container with moist soil from Loch Hill at a temperature of 16oC. The embryos were then observed under a microscope using optic lighting to determine the size of the worm.

Worm density is expressed per m3 of soil. To take into account the hillslope when digging a quadrat, the average of the highest and lowest depth of the quadrat dug is used for the calculation. The estimation of the number of worms at Loch can also be given per m2, assuming that most worms are in the top 1m of soil.

Mean densities were converted to logarithms to calculate the 95% confidence limits. Log transformations are used on small sample sizes where the sample is not randomly distributed over an area (Elliot 1977) as is the case with the Giant Gippsland Earthworm at Loch Hill. The confidence limits indicate that if one went back to sample the site, the mean density would fall between the confidence limit range given 95 % of the time.

Samples of Giant Gippsland Earthworm were also taken on one occasion for some trial genetic analyses by Dr Dave Runciman.

Methods for release of worms. Two methods were explored: release of worms into existing burrows, and creation of burrows using metal rod.

In the first method, uninjured worms were gently placed, anterior first into existing burrows of appropriate size. Sticks or a metal rod was used to check that the burrows were not blocked or did not end abruptly. However, this was often difficult to establish as the burrows were not straight. Worms were guided into burrows as far as possible This was usually only a few centimetres. The posterior section of the worm was supported by either a hand made ledge below the burrow on which the worm body could rest or with a build up of soil. The exposed section of the worm was gently buried with loose soil as far as possible to reduce the likelihood of desiccation and exposure to predators. The soil and burrow into which the worm was placed was dampened with tap water.

Artificial burrows were made using a thin metal rod, 1 cm in diameter hammered into the exposed soil face of a trench using a rubber mallet.

Possible future release sites at Loch Hill for translocated worms. During the translocation of worms and/ or egg cocoons at Loch Hill, it is proposed that at least one or more receptor sites should be located at Loch Hill in areas that will not be directly impacted by the road works. These sites may include i) areas near by that do not already support populations of the worm, preferably adjacent to existing sites and ii) restored habitat at Loch Hill.

A preliminary search for sites adjacent to known worm habitat was undertaken to determine suitable receptor sites for translocated worms. The boundaries of known Giant Gippsland Earthworm distribution around the bottom of Loch Hill were surveyed to determine where the worms "petered out". Small quadrats were dug to examine the soil for Giant Gippsland Earthworm burrows. Burrows are easily identified and, if wet, represent burrows that are actively being utilised by the worms. If the ground is wet, presence of the worms can also be established by banging the ground with a spade and listening for gurgles, the sound that is made when the worms retreat down their wet burrows. Large quadrats were dug at some sites to obtain an indication of worm density.

Results

Biology and Density. Eight quadrats were dug at Loch Hill over four months to further examine the density of earthworms and estimate their depth in the soil profile (Table 1a-1c). These sites were randomly located within the area of known worm distribution. All quadrats had Giant Gippsland Earthworm burrows and worms were recorded in all but one quadrat. A total of 20 Giant Gippsland Earthworm were recorded comprising 6 adults, 2 subadults/adults, 5 juveniles, and 7 worms of unknown size class.

Four egg cocoons were collected, including one hatched cocoon. One of the egg cocoons was accidentally chopped so the developmental stage of the embryo could not be ascertained. The two other egg cocoons were taken back to the laboratory where their progress is being monitored. If they hatch successfully, they will be released back at Loch Hill during the translocation phase of the project. The first egg cocoon collected on February 13th appeared to be fairly recently laid. The cocoon itself was relatively hard and opaque and it was difficult to observe the embryo inside. It was obviously very small at this point and thought to possibly be dead. The cocoon was examined several weeks later with a similar result. However, 3.5 months later, the cocoon was examined and the young worm was observed easily through the more transparent cocoon. It was found to have grown considerably and was around 3.5 times the length of the egg cocoon and around 16 cm in length (Plate 1). The second cocoon collected on June 5th contained an embryo of approximately 8cm in length (Plate 2). At present it is not possible to correlate embryo size with the age of the developing worm as this data is very difficult to collect and relies on the collection of freshly laid egg cocoons. Also development time varies according to temperature, with higher temperatures speeding up development. As these cocoons are being kept at 16˚C, the temperature is higher than that of the soil (around 10˚C) and therefore accelerated growth would be expected. Estimates of growth of cocoons from fertilisation to hatching is around 12 months at 12˚C (Van Praagh 1994).

No breeding adults were found (ie adults with swollen clitella).

Worms were found at a mean depth of 37cm.

The number of worms found ranged from 0 to 9 with the mean density per m3 ranging from 1.4 to 28.5 (Table 1b). The mean density recorded from the second sampling season was 7.3, a similar density to that recorded from the first sampling season (8.5 see table 1a). The mean density from both seasons combined is 7.9 per m^3.

A total of 43 worms were found from both study periods with all developmental classes recorded (Table 1a-c).

Release of worms into existing burrows. Several worms were released into existing burrows. One such released adult worm was followed up two weeks after release. It had died and was in a similar position to that which it was left in, with little progress into the burrow. A relatively recently hatched juvenile worm was successfully released into a small burrow (Plate 2a-d). Whilst the long term survival of this worm isn't known, its quick disappearance down a burrow is promising.

Although released worms were not severely injured, there is still stress involved in the capture process, often resulting in small abrasions and 'pressure' on the body (Plate 3).

Creation of burrows using metal rod. This method proved to be unsuccessful due to difficulties in hammering in the rod far enough and being able to remove it from the soil

Future sites for translocation of Giant Gippsland Earthworm around Loch Hill

Two sites that may serve as potential Giant Gippsland Earthworm translocation sites were discovered during this study.

Site 1 was located in the next major hillslope gully just to the east of Loch Hill in the property owned by Christina and Gerry Norbergen. A small patch of Giant Gippsland Earthworms were found virtually at the head of the gully. This area was terraced and very moist. Worms were restricted to a small area of 5-10 m wide by approximately 15 to 20 long. They were not found in the surrounding hillside area. Burrows were quite dense in the top section of this area with a juvenile worm located indicating a breeding population occurred at the site. Burrows then became less dense toward the lower end of the occupied area

Site 2 was located at the bottom of the hillslope at Loch Hill, to the west of the tree fern. It occurred on the border of the established worm distribution and had a very low density of burrows. Burrows disappeared further west.

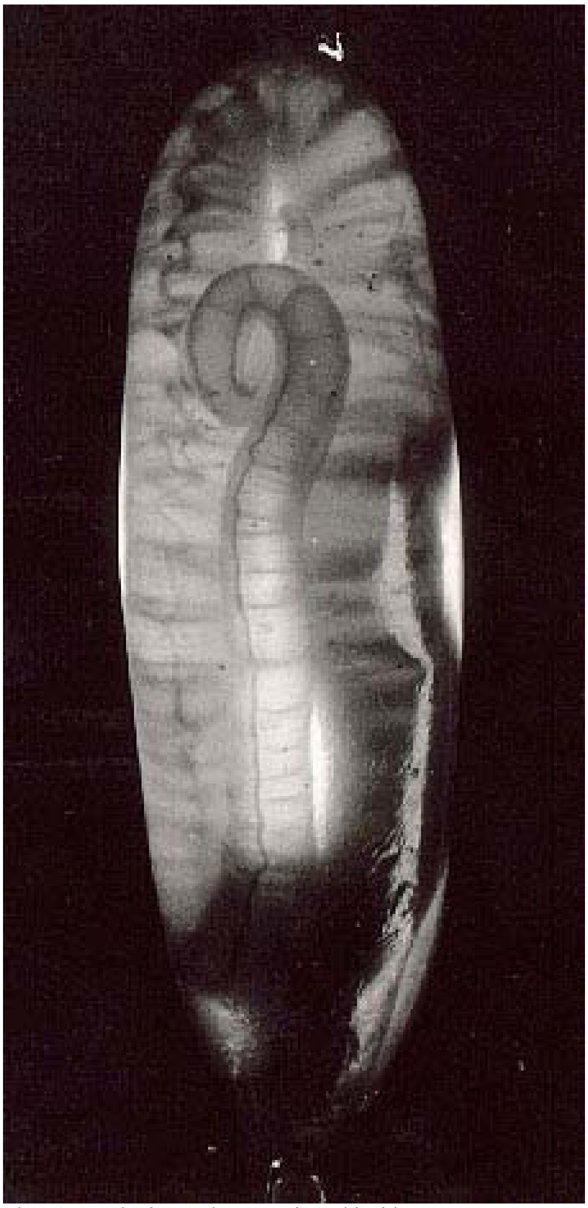

Plate 1 Developing embryo as viewed inside egg cocoon

Table 1a Number, depth and density of *M. australis* recorded at study sites at Loch Hill from October to end of November (previous study). "Unknown" indicates that a worm was observed but not completely dug out so that its age class could not be determined; ? indicates that weight was not recorded. Immature indicates either juvenile or subadult worm.

Date	**Site**	**Lat long**	**No of worms**	***Age class and weight of known worms**	**No and depth of egg cocoons**	**Ave depth of worms (cm) (range)**	**Area dug m^3 Ave depth = the average of the highest and lowest depth of the quadrat taking into account hillslope**	**Ave density of worms per m^3 (95% C.L)**
10 Oct 01	1	38º 22' 720" 145º 43' 570"	1	Juvenile (95 g)	0	20	0.8 x0.35x0.2 =0.056	17.9
10 Oct 01	2	38º 22' 722" 145º 43' 563"	5	3 Unknown, 1 subadult, 1 adult	0	55.8 (50-68)	1.20 x1.10x 0.50 (ave depth 0.58) = 0.8	6.5
17 Oct 01	3	38º 22' 725" 145º 43' 567"	3	3 juveniles (18g, 33g, ?)	0	25 (20-35)	1.20x1.50x0.55(ave depth 0.33) =0.59	5.1
17 Oct 01	4	38º 22' 763" 145º 43' 55	2	1 adult (150g), 1 unknown	0	59 (54-64)	1.13x 0.90x 0.90 (ave depth 0.47) = 0.48.	4.1
13 Nov 01	5	38º 22' 739" 145º 43' 548"	3	1 immature, 1 adult, 1 juvenile (50 g)	1(hatched)	28 (10-50)	110x90x60 (ave depth 0.35)=0.34	8.7
28 Nov 01	6	38º 22' 725" 145º 43' 567"	8-10 (9)	2 Juveniles (30 g, ?), 4 adults (215g, 250g, 220g, ?) and 4 unknowns.	2 (hatched) @ 25 and 15 cm	35 (15-55)	1.5x1.5x 0.80 (ave depth 0.45) =1.01	8.9
			23		**3**	**39.3±16.6**	**Average density of worms**	**8.5 (4.9-14.6)**

Table 1b Number, depth and density of M. australis recorded at study sites at Loch Hill from February until May (present study). "Unknown" indicates that a worm was observed but not completely dug out so that its age class could not be determined; ? indicates that weight was not recorded. Immature indicates either juvenile or subadult worm.

Date	**Site**	**Lat long**	**No of worms**	***Age class and weight of known worms**	**No and depth (cm) of egg cocoons**	**Ave depth of worms (cm) (range) ±S.D**	**Area dug m^3 Ave depth = the average of the highest and lowest depth of the quadrat taking into account hillslope**	**Ave density of worms per m^3 (95% C.L)**
13 Feb 02	1	38 25 365 145 47 556	0	-	0	-	0.90 x1.20 x 0.80 (average depth 0.60) =0.6	0
13 Feb 02	2	38 22 734 145 43 559	3	2 adults (125g, ?) 1 unknown	1 * (15cm) taken back to lab	44(40-47)	1.45x 0.95x55 (ave depth 0.50) =0.69	4.3
13 March 02	3	38 22 734 145 43 570	1	1 adult (g?) Possible post clitellate	0	50	1.5x1.05 x 0.80 (ave depth 0.45) = 0.7	1.4
13 March 02	4	38 22 733 145 43 566	2	2 adults 155g, 170g)	0	37.5 (35-40)	1.0x 0.9x.8 (ave depth 0.50) =0.45	4.4
03 April 02	5	38 22 737 145 43 558	1	1 subadult/adult 155 g 2-3 bands present	3 (1 empty, 1 damaged, 1 undamaged) (35, 26, 24 cm depth respectively)	45	1.28x0.6x 0.7(ave depth 42.5) =0.33	3.0
23 April 02	6	38 22 741 145 43 538 (edge of worm distribution)	1	1 Subadult/adult 125 2-3 bands present	0	40	1.13 x.85x.50 (ave 0.30) =0.28	3.5
23 April 02	3	38 22 737 145 43 545	3	1 juvenile (30g), 2 unknown	0	26.6(10-55)	0.8 x 0.90 x 0.52 (ave 0.31) =0.22	13.6
22 May 02	8	Near above site	9	4 juveniles (52, ?), 1 adult (140 g), 4 unknows	0	38.3 (20-50)	1.0 x 0.8x 0.6 (ave 0.35) =0.28	28.5
Total			**20**		**4**	**36.9±15.4**		**7.3 (3.1-17.2)**

Table 1c. Total number, average depth and density of *M. australis* recorded at study sites at Loch Hill from both studies combined

Total No of worms	***Age class**					**No of egg cocoons**	**Ave depth of worms (cm) (±S.D.)**	**Ave density of worms per m^3**
	Juv	**Sub-adult**	**Immature**	**Adult**	**Unknown**			
43	11	3 ?	1	13	15	7	38.3±15.6	7.9 (4.8-12.9)

Plate 2a. Juvenile worm prior to release into existing burrow.

Plate 2b. Juvenile worm moving into existing burrow.

Plate 2c. Juvenile worm moving into existing burrow.

Plate 2d. Juvenile worm moving into existing burrow.

Plate 2e. Juvenile worm moving into existing burrow. The process was completed over a 5-10 minute period.

Plate 3. An adult worm showing physical stress of capture. Generally caused by worm wedging itself into burrow so that it cannot be pulled out of its burrow.

Discussion

Biology and Density. The Giant Gippsland Earthworm was found to be widely distributed over its known range at Loch Hill. All but one of the randomly located quadrats supported worms and all quadrats contained burrows. The suggestion of very high worm densities at Loch Hill as indicated in the first study was supported by the results in this study. The overall density of 7.9 worms per m3 remains higher than the previous average density of 2.25 worms per m3 recorded from other studies (Van Praagh 1992, 96). Generally worm density drops off markedly during the summer months when it can be very difficult to locate worms. However, at Loch Hill, worms were easily found over the summer period. This may be due to the mild weather conditions this year and the absence of prolonged periods of hot weather over summer. It could also indicate that the site is wet throughout the summer. Worms were found at similar depth to the initial study at an average depth of 37.2 cm.

All age classes (juvenile, subadults and adults) were recorded at the site. In particular, a large number of juvenile worms and several egg cocoons. This indicates the presence of a healthy, breeding population of earthworms. Worms with swollen clitella, indicative of breeding were not recorded in the last survey, which was thought surprising given that sampling occurred during the breeding season (Sept to Feb). It was thought that the prolonged cooler weather conditions encountered in spring and summer 2001 may have delayed breeding. However, no breeding worms were found in the latest survey. Studies have found that adult worms need to be at least 180 g with an average weight of 255 g before breeding can occur (Van Praagh 1996). The largest adult recorded in the present study was only 170g, well below the average adult weight and known breeding threshold as determined from studies at a nearby creek bank at Loch (Van Praagh 1994). Studies at this site, where sampling occurred fortnightly over a three year period, found the average adult weight was around 210 g with the largest worm weighing 380g. The average weight of adults from the first and second survey at Loch Hill was 208 and 147g respectively with the overall average weight from both surveys being 178g (n=8). The largest adult found weighed 250 g. Therefore average adult weight was lower than that found from other studies. It may simply be a sampling bias in that they were deeper down in the soil and therefore not completely dug out for weighing. Other possibilities include low sample size, the high densities recorded may cause competition resulting in smaller worms or that the quality of the habitat is not as good as that found elsewhere.

Release Methods. Both release methods met with varying degrees of success. Releasing worms into established burrows is a possibility though it is difficult following up the success of this method without killing or injuring the worm. Also the identity of the released worm is difficult to establish until appropriate marking techniques are developed. One of the difficulties with releasing worms may be that the method of extraction, rather than the release itself, causes the most problems for the worms. It is very difficult to extract worms by hand, regardless of how careful one is, without causing some physical stress to the worm. Thus it is not known whether the death of worms after release results from the injuries caused by capture or occurs because the release method was inadequate and the worm could not proceed down the burrow for some reason. If worms were extracted without injury, they may have a better chance of survival using this method.

One method of extraction suggested for the trial in translocation involves using machinery to move large blocks of soil supporting worms into pre-dug holes or removing worms individually from soil blocks and releasing into burrows. While some worms on the edges of these blocks are vulnerable to injury, it is possible that those in the centre of the block may remain unharmed.

Given the apparent success of releasing a juvenile worm (although its survival after being released was not monitored) it appears likely that young worms and egg cocoons represent the best chance of successful translocation. This may be partly because it is easier to extract young worms out of the soil without injury because of their small size.

The creation of artificial burrows was unsuccessful due to the difficulties of hammering the steel rod into the ground far enough and then removing it. The use of a screw auger may be more beneficial as it would be more adaptable for the purpose of creating holes. However, Giant Gippsland Earthworm burrows are not entirely vertical or horizontal but rather form a connected network of burrows radiating in different directions (Kretzschmar and Aries 1992).

Translocation Sites. At this stage it is unknown whether the most appropriate translocation sites should already support worms. If sites do not support Giant Gippsland Earthworm then it is likely that the habitat is unsuitable for the species and therefore may not sustain released worms. A possibility may be to attempt to manipulate this sort of habitat to suit worms. However, at this point the feasibility of this is unknown. This will be the subject of some pre-translocation investigations. The two sites found in this study represent sites that have a small population or are on the edge of the worms' distribution. Site one has a very discrete, small population of worms. This site may be able to be extended somewhat to support translocated worms and since the existing population is small, the new worms may be easier to distinguish. However there may be some accessibility problems for this site given that it occurs high up on a very steep hillslope. Site two exists on the eastern border of worm distribution at Loch Hill. It is not known whether this is because the site represents marginal habitat or simply because the worms have not dispersed that far yet.

Loch Hill supports an extensive Giant Gippsland Earthworm population with very high densities recorded. All age classes occur at the site with a relatively high number of juvenile worms noted. This, along with the collection of several developing and hatched egg cocoons, indicates a healthy breeding population, although no breeding adults were found. Adult earthworms were overall somewhat smaller than that recorded from similar studies. This may be a result of small sample size or because the high densities are causing competition amongst the population. Another possibility is that the habitat at Loch Hill is not as conducive to the development of larger sizes as the creek bank site where previous studies have occurred. Time consuming and damaging techniques of hand collection result in small sample sizes so that only limited interpretation of population structure can be made. Further information on a scale never before possible will be obtained during the translocation phase of the project at Loch Hill. The use of machinery and the movement of large amounts of soil will allow many more individual earthworms to be examined allowing more solid data to be collected. This information, together with the results of the translocation of specimens, will aid in our understanding of the species and its conservation.

Acknowledgements

This work was funded by Vic Roads. Help of Vic Roads staff, in particular Graham Embry and David Gellion is much appreciated. Giant Gippsland Earthworms were collected under Research Permit to Dr. A. Yen under Wildlife Act (1975) and Flora and Fauna Guarantee Act (1988) No. 10001631

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Appendix 1 – Giant Gippsland Earthworm Project – Preliminary investigations into veterinary aspects of worm management

By Dr. David Middleton. (Healesville Sanctuary's Wild Animal Clinic)

Background

As a result of decisions taken at the initial planning meeting on May 1st, 2002 at Loch, Victoria, undertakings to investigate specific veterinary aspects of worm management were made. The provisional estimate document, 13/5/02, listed 11 components allocated to three distinct parts – Basic Health Care Techniques, Immediate management objectives and Laboratory based investigations (Appendix 2). Five of the eight components of Parts One and Two were initiated with some encouraging and useful information emerging.

These aspects included reviewing the anatomy of the species, identifying clinical parameters useful for assessing health status, trialing various sedatives, examining a range of tissues histo-pathologically (especially the skin and its response to injury), establishing a small colony in captivity for the development of health monitoring protocols and the design and implementation of methods for identifying individuals visually. In response to these objectives a field trip was conducted on 22 May 2002. Two sub-adult worms were retrieved for clinic based trials of sedatives, anaesthetics and euthanasia agents. These specimens provided the basis for anatomical, haematological and histological studies.

Items investigated (component numbers are consistent with the provisional estimates document)

Anatomy. The anatomy of Giant Gippsland Earthworm has been described in reasonable detail in Baldwin Spencer, (1888) to be supplied. However clinical anatomical descriptions are lacking. We investigated various accessible blood sampling points, coelomic fluid sampling points and administration sites for various agents.

Health parameters. The components of the blood have been described in Jones et al. (1994) and Stephenson (1930). But the response to injury, to metabolic disturbance and to infection does not seem to have been investigated in detail. We have conducted preliminary haematological investigations and submitted blood films and digital images to a specialist clinical pathologist for more accurate identification of cells and their functions. We have also sampled quantities of a clear serous fluid which resembles lymph. The identity of this fluid remains uncertain as the exact site of origin within the worm is not known. It may be a transudate of gut origin and it may have some relationship to the pale fluid reported to be expressed through the dorsal pores as a lubricant for subterranean motion (Spencer 1888). It is possible that this fluid may be valuable in health monitoring.

Histopathological investigations. A range of tissues were submitted for histopathological investigation. Although no specific abnormalities were reported, a more detailed account of the structure of tissues of particular interest (skin for example) has not yet been received. We intend to assemble a library of normal tissue sections for future reference.

Sedation, anaesthesia and euthanasia methods. Various substances were trialled as sedatives under both field and laboratory conditions and four routes of administration were used (topical, intra-coelomic, intra-muscular and intra-venous). These trials were preliminary as only 3 worms were used and a number of substances administered in series. This meant that individual worms received more than one substance and observations about single drug effects could not be made in most cases. Some initial response to diazepam was noted in the field however this was not repeatable with any reliability. Isoflurane, ketamine, zolazepam/tiletamine mixture, lignocaine, bupivicaine, medatomidine, suxamethonium chloride, midazolam and pentabarbitone were all tested in a preliminary manner. No substances were considered appropriate for sedation or euthanasia. Following advice from invertebrate specialists in UK (Cooper, 2001), further trials will include electrolyte solutions, carbon di-oxide, tricaine and benzocaine topically. Whilst direct access to the circulation of the worm was possible using fine needles (30g) this was not reliable enough to guarantee IV administration. Further work on access sites is required. Euthanasia methods are not easy to evaluate because of the difficulty in establishing the point of death. Further research is required in this area.

Marking and identification. These aspects have not be investigated to date. Plans to trial a number of mechanisms including dermal dyes and implants are planned for the future.

Thermal imaging trials. These have not been conducted. An introduction to the technology is to take place at Healesville in the near future to ascertain its applicability to worm research.

Provision of materials for genetic investigations. A genetics representative was present during the field trial and samples of various kinds were collected for analysis.

Establishment of a laboratory colony. Two specimens were retrieved for euthanasia and investigation. No immediate plans exist for the establishment of captive colony. It is recommended that this goes ahead in the near future however. Specialist invertebrate staff are available at Healesville to oversee the construction of a facility and provide day to day husbandry.

Summary of progress and future directions

If the Giant Gippsland Earthworm colony at Loch Hill is impacted by road construction, further investigations are required on all aspects outlined. Specifically, more recently recommended agents of anesthesia need to be trialled and a captive colony established at Healesville Sanctuary's Wild Animal Clinic to support further studies including health monitoring and response to injury and infection.

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Appendix 2 – Giant Gippsland Earthworm Project Provisional Estimates for Health Investigations

By Dr. David Middleton (Healesville Sanctuary's Wild Animal Clinic)

Introduction

Healesville Sanctuary's Wild Animal Clinic (AWH) provides veterinary services for the health, welfare and conservation of Australian wildlife. AWH is currently engaged in a number of endangered and threatened species recovery projects including those for Orange Bellied Parrots, Helmeted Honeyeaters, Eastern Barred Bandicoots, Tiger Quolls and Eastern Quolls. AWH also provides professional services in support of wildlife management objectives such as koala and kangaroo fertility control, flying fox translocation, mortality and disease investigations, bird of prey rehabilitation and wildlife rescue.

Our veterinarians are involved in research at many levels including original investigations, training of investigators, capture and restraint techniques, health monitoring and welfare advice. They also provide support for various government and university entities such as ethics committees, steering committees, technical advisory bodies and panels established for such tasks as the drafting of codes of practice for wildlife. The AWH is accessible 365 days a year for assisting veterinarians and other professional and technical individuals to solve problems associated with wildlife from the individual animal level through populations to issues relevant to the survival of the species.

Healesville Sanctuary is uniquely equipped to interpret and summarise conservation projects and present them to the visiting public. In this way, our organization is able to generate community support and interest for various programs which seek to provide long term security for wildlife and wildlife habitat. Our mission is to encourage enduring relationships between people, wildlife and the environment.

Giant Earthworm Project – Basic Health Care Techniques

Review of current knowledge of earthworm health and survival including consideration of environmental factors influencing health.

Collection and analysis of basic health parameters.

Histopathological investigation of normal and abnormal tissues with a view to characterizing the response to injury and infection.

Giant Earthworm Project – Basic Support of immediate management objectives

Investigation of appropriate sedation and euthanasia methods.

Investigation of marking and identification techniques.

Trialing of survey and monitoring techniques involving specialized thermal imaging.

Provision of appropriate tissues/fluids for genetic investigation.

Establishment of artificially maintained colony at Healesville Sanctuary.

Giant Earthworm Project – Laboratory based investigations on artificially maintained worms

Refinement of husbandry and maintenance protocols.

Trialing of management techniques relevant to field management.

Design and construction of public display.