Trialling detection dogs as a novel method for finding threatened reptiles

Final report

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This research was undertaken on Wadawurrung and Wurundjeri Country. We acknowledge and recognise the Traditional Custodians and their continuing connection to the land, skies, waters and culture. We pay our respects to their Elders past and present.

Cover photo: A Victorian grassland earless dragon in its burrow. Source: La Toya Jamieson.

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Summary

The Victorian grassland earless dragon (*Tympanocryptis pinguicolla;* VGED) is a critically endangered species of lizard that was recently rediscovered in early 2023 west of Melbourne, after more than 50 years since the last individual was recorded. It is therefore essential that all efforts are taken to both conserve this fragile population and to search for further remaining populations. Exploring novel survey methods, including highly-trained detection dog-handler teams, will be valuable for conserving this, and possibly other, imperilled cryptic reptile species.

In this document, we report on the results of a project exploring how wildlife detection dog-handler teams could be trained to detect live VGEDs and their scat. In addition, we also measured the longevity of VGED scats remaining in situ (including rates of scavenging and degradation) to inform the feasibility and potential value of scat detection surveys.

The period taken to train dogs to detect VGEDs was extensive and was often limited by the rarity of the species. This meant that opportunities to train on wild VGEDs in situ were difficult to predict in advance and were scarce overall. Despite this, the VGED detection dogs, named Kip and Daisy, both successfully and independently detected naturally occurring VGEDs sheltering in previously unknown burrows. Since completing training and initial VGED detectability assessments, these dog-handler teams successfully located 8 wild VGEDs sheltering in previously unknown natural burrows over 11 survey days. This demonstrates the potential of this novel survey method for this highly cryptic species and may provide new opportunities to survey for VGEDs outside of the standard survey periods.

Two additional detection dogs, named Sugar and Moss, were also successfully trained to accurately discriminate between scats from VGEDs and other reptile species. Sugar and her handler, La Toya Jamieson, were then assessed on their ability to detect novel VGED scat samples sourced from captivity in 21 area search assessments. This dog-handler team recorded an overall search sensitivity of 72%, yet they were able to confirm the presence of any VGED scats within a search area in 81.3% of searches in which it was present. This training and assessments process highlighted that fine-scale search strategies were required to maximise the likelihood of detection and reduce the risk of false negative search outcomes. The ability of dogs to generalise their detection to naturally occurring scat from wild VGEDs was likely impacted by the lack of in situ training opportunities in which wild scats could be positively identified as belonging to VGEDs.

We also present the results of a VGED scat longevity trial that was conduct at the same time as the scat detection training project. This trial was designed to determine the rate at which scats were being scavenged or degraded at the rediscovery site. Uncovered (exposed) scat (n = 22) were scavenged more quickly than expected, with more than one-third of samples being scavenged within 24 hours. The high rates of scavenging and degradation and the likely low rates of VGED defecations may result in few scats being persisting in the environment for any great length of time. This further increases the challenge of detection dog teams locating scat; at the same time, it directly increases the value of any scats located.

Overall, our findings highlight the value of employing detection dog teams for VGED surveys, but also the challenges that must be overcome for this method to be safely and effectively employed. As a

result of this research, we have developed a robust training, assessment and survey methodology, which we expect will continue to evolve with future work.

No survey method is perfect. It is therefore essential that we continue to explore the sensitivity and efficiency of this new survey method for VGED and increase our understanding of the search effort required to provide high confidence that any targets present are detected while simultaneously reducing the risk of false negatives. Highly-trained detection dog teams therefore present a powerful method for VGED conservation.

1.0 Introduction

1.1 Species Overview

The Victorian grassland earless dragon (*Tympanocryptis pinguicolla;* VGED) is listed as 'Critically Endangered' under the Australian Government's *Environment Protection and Biodiversity Conservation Act 1999* ('the EPBC Act'). It has been identified as the most imperilled reptile in Australia (Tingley et al. 2019; Geyle et al. 2020; Garnett et al. 2022).

The scientific name of *Tympanocryptis pinguicolla* previously covered the grassland earless dragon complex of 4 species across New South Wales (NSW), the Australian Capital Territory (ACT) and Victoria, which was listed in the Endangered category of the threatened species list under the EPBC Act, effective from 16 July 2000.

Following a 2019 taxonomic review by Jane Melville and others, the *T. pinguicolla* name is applied to the grassland earless dragon restricted to the greater Melbourne region (Melville et al., 2019).

The rediscovery of a VGED on 20 January 2023 was extraordinary news. The species had not previously been seen for just over 50 years despite extensive survey effort, particularly over the past 30 years as Melbourne's urban fringe expanded westwards into its historical range.

Its rediscovery has confirmed that it has survived in an area of private land with a very long history of sheep grazing, droughts, and other significant pressures. Zoos Victoria are leading the recovery effort for the species due to:

- The VGED being one of Zoos Victoria's 27 priority 'Fighting Extinction' species.
- Zoos Victoria's previous experience in surveying for the VGED species.
- Zoos Victoria's role in establishing a successful conservation breeding program for the Canberra grassland earless dragon.

Prior to the rediscovery of VGED, current and future approvals and development in the VGED's historical range were generally occurring in the absence of any or limited consideration for the species, both at a state and federal level. Similarly, survey techniques that have been successful for detecting grassland earless dragon species in other jurisdictions had been completely unsuccessful in Victoria. Fast-tracking the trialling of new techniques such as detection dogs is therefore critical to support the protection and recovery of this species, improving our understanding of VGED distribution, population health and threats as well as efficacy and confidence in surveys to assist those in developing an informed regulatory posture for the species.

1.2 Detection Dog Program Overview

Zoos Victoria's Fighting Extinction Conservation Detection Dog Program harnesses the highly sensitive olfactory capacity of canines to assist threatened species biologists to locate and monitor wildlife, which can enhance and complement current survey methods. Since 2017, the program has been developed to assist with Zoos Victoria's wildlife conservation efforts, in field surveys, research initiatives, and community engagement. Alongside their specialist dog training and handling skills, Zoos Victoria's Wildlife Detection Dog Officers have backgrounds in wildlife biology and ecology and collaborate closely with species experts and land managers to achieve program objectives.

The Conservation Detection Dog program based at Healesville Sanctuary currently includes 5 dogs working on projects with multiple threatened species, including:

- Baw Baw frog: Population monitoring and collection of individuals for conservation breeding.
- Broad-toothed rat: Presence/absence surveys that provide data to support habitat protection.
- Platypus: Assisting non-invasive population monitoring by locating occupied burrows in river banks.
- Tasmanian devil: Detecting changes in hormone profiles specifically related to oestrus and lactation to inform conservation captive-breeding and management of the species.
- The tea-tree fingers fungus: Detecting one of Australia’s rarest and most endangered fungi in situ (this project is in collaboration with the Royal Botanic Gardens Victoria).

The Zoos Victoria Wildlife Detection Dog research team have also undertaken an extensive research project investigating the ability of dogs trained on more common frog species to ‘generalise’ their scent training across to detection of rarer species that they have not had previous exposure to.

The key Wildlife Detection Dog program objective is to have up to 5 detection dogs trained on multiple threatened species and for those dogs to be readily deployable for wildlife monitoring surveys across Victoria. There is also potential for dogs to be rapidly trained and deployed to play a pivotal role in either locating threatened species that are in high-risk fire areas, or when recovering injured individuals or assessing population loss post-bushfire.

Figure 1. Zoos Victoria’s Wildlife Detection Dogs. Pictured left to right: Moss, Sugar, Daisy, Finn and Kip.

Source: Zoos Victoria.

1.3 Project Objective

The aim of this project is to support Zoos Victoria with innovative research in their response to the rediscovery of the cryptic VGED by training the Zoos Victoria Wildlife Detection Dog teams to locate both live VGED and their scat; and to determine best practice methods for training and surveying for this species when working with wildlife detection dog teams.

1.4 Project Description

To support the project objective, we trialled a novel technique in the use of detection dogs to detect a rare, imperilled and highly cryptic reptile.

This research involved training detection dogs and trialling their effectiveness to detect VGED or VGED scats, both at the rediscovery site and at adjacent locations west of Melbourne.

Training for live VGED detection initially took place with VGEDs housed at Melbourne Zoo for the conservation breeding program, then with wild-caught animals, before dogs graduated to in situ detection training and detection trials at the VGED rediscovery site. In addition to live VGED detection dogs, VGED scat detection dogs were also trained and assessed. The project aim was to trial these techniques and to assess their effectiveness as a method that can be used to increase our understanding of this species' extent of occurrence and occupancy in remnant areas of suitable habitat in the greater Melbourne area.

Results of this research will begin to inform future decision-making regarding surveys, research and conservation for this highly imperilled species, as well as other *Tympanocryptis* species.

Future applications will include applying these learnings to training additional live VGED detection dogs, surveys for additional populations, identification of sites/populations for protection, ongoing population monitoring, identification for collection of individual VGEDs for the conservation breeding and insurance program, disease monitoring, ecological knowledge building, and assessment of key fitness indicators such as survival, reproduction, and dispersal.

The following sections describe in detail the 3 components of the study: live VGED detection, VGED scat detection, and VGED scat longevity trials.

2.0 Detection dog teams trained to locate live Victorian grassland earless dragons

2.1 Introduction

Training detection dogs to locate live VGEDs in situ represented a unique project for Zoos Victoria's Wildlife Detection Dog team. To date, detection dog-handler teams working on reptile or amphibian projects is not common, however, this is an evolving space (Grimm-Seyfarth et al. 2021). Training detection dogs on a reptile species, especially one that often resides in burrows, was therefore anticipated to have unique training challenges. Within this project, we explored training, assessment and field survey methods for detecting live VGEDs with 2 experienced detection dog teams.

The primary objective of this project was to determine the feasibility of detection dog teams locating wild VGEDs to complement current survey methods. The specific aims in this component of the project were to:

- Develop a training protocol that will allow detection dogs to be successfully trained to survey for VGEDs.
- Develop an assessment protocol that will measure detection dog-handler teams' search performance—including sensitivity and search effort—when surveying for VGEDs.
- Develop a VGED survey protocol for detection dog-handler teams that would maximise survey efficiency and detection confidence.
- Successfully train detection dog-handler teams to locate VGEDs, including those VGEDs sheltering in burrows, tussocks or other refugia, in situ. This will enable VGEDs and their burrows to be recorded, and potentially for genetic samples to be collected or the individuals themselves collected for conservation breeding, where appropriate.
- During detection dog training and surveys, collate information on active VGED burrow location, appearance and usage characteristics. This information may inform future survey efforts and allow investigation on burrows identified by detection dog teams and those identified during human visual surveys.

All activities completed in this project were conducted in accordance with approval granted by Zoos Victoria's Animal Ethics Committee (ZV20010 and ZV23010) and relevant research permits (Wildlife permit 10010823).

2.2 Methods

2.2.1 Detection dogs

Two of Zoos Victoria's most experienced wildlife detection dogs were selected to detect live VGEDs. Kip, an 8-year-old Kelpie cross, and Daisy, a 6-year-old Lagotto Romagnolo exclusively worked with their handlers Naomi Hodgens and Nick Rutter throughout the duration of this project. These teams were selected due to their experience and for Kip and Daisy's safe behaviour around small animals. Kip is experienced surveying for active platypus (*Ornithorhynchus anatinus*) burrows, Baw Baw frogs

(*Philoria frosti*), and Victorian freshwater turtle nests, in addition to research work detecting Tasmanian devil (*Sarcophilus harrisii*) oestrus from scat samples. Kip is generally reinforced with toy play, food or verbal/physical praise. Daisy is experienced surveying for tea-tree fingers fungus (*Hypocreopsis amplectens*), bird and bat carcasses, and Victorian freshwater turtle nests, in addition to research work detecting Tasmanian devil oestrus from scat samples. Daisy is generally reinforced with food and verbal/physical praise.

As with all Zoos Victoria's operational wildlife detection dogs, both Kip and Daisy undergo annual detectability assessments on a familiar training odour (pieces of rubber from the dog toy brand, Kong, that has a consistent scent) to demonstrate reliable search performance. Both teams also train for and display reliable safety cues to ensure they are safe to work in the field. These assessments are in accordance with those outlined in the Australasian Conservation Dog Network (ACDN) evaluation guidelines (Guidelines - Australasian Conservation Dog Network).

2.2.2 Captive VGED training

To introduce the odour of VGEDs to the detection dogs, we presented live VGEDs in purpose-built PVC containers, or 'pipes' that allowed the odour of the VGEDs to be accessible to the dogs while visual contact between the VGED and the dogs was inaccessible (See Figure 2). Training sessions occurred between March – May 2023 at Melbourne Zoo. Training sessions occurred for up to 30 minutes, to safeguard the welfare of the VGED and to maximise the dogs' learning.

Figure 2. Container used to temporarily hold a VGED to facilitate detection dog training. The container allows odour dispersal while preventing visual contact between the VGED and dog.

Source: Nick Rutter.

Both dogs were exposed to the odour of VGEDs for training using these pipes in one of 2 ways. Pipes were either presented in a linear ‘line up’ or arranged in a ring of pipe ‘stations’ that dogs could investigate or be lead around systematically (Figure 3). In either scenario, up to 10 pipes could be included in a training session, with pipes containing either a live VGED, a distractor odour (e.g. snail, cricket, rubber glove) or were left empty (i.e. blank pipe).

During initial training sessions, dogs were rewarded with food, praise and/or ball games for investigating the pipe containing the VGED. Care was taken when rewarding the dogs around the live VGED to not disturb the animal. Over successive training sessions, handlers applied behaviour shaping principles through positive reinforcement techniques to train dogs to perform a sit alert at the pipe containing a VGED. Training searches could include from zero to 2 VGEDs within a line up or search ring. Training activities varied by day, depending on the requirements of each dog, yet each training day generally consisted of 2 short sessions of approximately 5 to 6 VGED presentations per dog. Throughout their involvement in training, VGEDs were monitored for signs of stress and training was to be immediately ceased if any concerning behaviours occurred. During the training, no VGEDs displayed concerning behaviours.

Figure 3. Daisy and handler, Nick Rutter, completing a container search training session at Melbourne Zoo using the pipe containers to securely house live VGEDs. Pipe containers are secured by bricks to prevent movement

Source: Naomi Hodgens.

2.2.3 Field training

After the completion of the first training milestone on captive VGEDs (whereby dogs demonstrated they recognised the odour of VGEDs as a target), the next training milestone required dogs to locate and alert to VGEDs in their natural habitat. This is an important training step in the overall development of effective VGED survey teams, especially as we are not aware of how the odour of captive and wild VGEDs varied to a dog.

From April 2023 the detection dog-handler teams began attending the rediscovery site, along with members of the field recovery team, on a regular basis. This in situ training focused on 2 main training goals:

- Familiarising dogs to the rediscovery site through conducting training searches for Kong rubber (i.e. their training odour).
- Capitalising on any in situ detection training opportunities on wild VGEDs detected by humans in burrows or traps.

Dog-handler teams worked on site familiarisation training searches in areas near the VGED recovery team members, who were monitoring survey infrastructure including tile grids, artificial spider tube grids, pitfall traps and checking natural invertebrate burrows with endoscope equipment. When a member of the recovery team located a VGED, detection dogs were able to search around those individuals VGEDs (that were contained within natural burrows, under tiles or in artificial spider tubes) and the dogs were reinforced for safely detecting and eventually alerting the dog-handlers to the VGED. These sessions were always completed with a member of the recovery team present to monitor VGED behaviour and were only conducted when the safety and welfare standards of the detection dog team and recovery team could be upheld. As with captive training, care was always taken when rewarding the dogs around the live VGED to minimise any disturbance.

As the number, frequency and nature of training opportunities on live VGEDs in situ was highly unpredictable, it was necessary for handlers to maintain a large degree of flexibility in training methodology and structure when a VGED was located. The team could go for weeks, if not months, without the opportunity to train on a VGED and this does not represent an optimal training schedule for efficient detection dog training. Despite this inherent variability, training sessions could be broken down into 2 categories: 'guided searches' or 'blind searches'. Handlers conducted guided searches in which dogs would be guided to search near where a VGED was located, such as an invertebrate burrow. Alternatively, handlers completed blind searches with their dog in a defined area in which the presence or location of a VGED was not revealed to them. Handlers chose to complete these opportunistic training exercises as either blind or guided based on the specific training goals relevant to the dog-handler team at the time. In general, handlers opted to complete guided training sessions earlier on in the training progression and blind sessions once dogs were more confidently alerting to VGEDs in situ in preparation for initial VGED detectability assessments.

Dogs could be considered to have successfully acquired the skills relevant to this stage of training once they could reliably detect and alert to a live VGED in situ. This does not mean training is complete, as some degree of field training is an important ongoing component of successful detection dog-handler teams.

2.2.4 Initial VGED detectability assessments

After completion of the second training milestone, whereby dog-handler teams could locate VGEDs in training search areas, the next milestone was for dog-handler teams to locate wild VGEDs in situ during single-blind searches of defined areas. (A single-blind search is when an observer knows the location of the VGED, but the dog and handler does not.) A single-blind search is important in understanding how dog-handler teams might locate unknown VGEDs, and in developing an empirical understanding of dog-handler teams' search performance (including sensitivity) and required search effort over multiple search assessments.

Given that the number of opportunities to complete assessments on wild VGEDs in situ is highly unpredictable (due to the rarity of wild VGEDs), handlers sought to complete assessments in as many opportunities as possible during this interim reporting period, while continuing to conduct assessments into the future, where possible. The decision was made that for all initial assessments to be completed with a single-blind design, whereby an observer who knows the location of a VGED was present during each search, rather than a double-blind design in which no one present knows the VGED location. This design permitted the timely and effective reinforcement of dogs that have alerted to a wild VGED, as the observer could immediately confirm the accuracy of alerts when prompted by a handler. Avoiding delays in reinforcement in this stage of training was considered essential for the dogs' overall training progression. This approach also allowed for VGED to be monitored during searches, so it was known that the VGED was still present before the searched commenced.

Handlers and recovery team members worked to locate wild VGEDs in situ that were sheltering in natural or artificial burrows by inspecting burrows with endoscopes or torchlight. The use of a torch was preferred as it does not leave incidental olfactory cues, such as disturbed soil or subtle endoscope odour, which the dogs may inadvertently consider relevant to their target as they consolidate their understanding of the VGED odour profile.

Once a VGED was located, a 10m x 10m (100 m^2) area was flagged out around the burrow, ensuring that it could be situated anywhere within the area (i.e. not always in the middle) and ensuring that the relevant dog-handler team remained blind to the burrow location. To ensure human odour cues were also not an indicator of VGED presence to the dog, the person setting up assessments walked throughout the search area and spent an equivalent amount of time looking in other locations and crouching down at certain spots.

When ready, the dog-handler team then commenced the search, with the handler guiding the dog to effectively cover the search area in a systematic way. This included considering environmental variables such as wind direction and speed while also noting any changes in dog behaviour including, but not limited to, trained alert behaviours (e.g. sit and stare). If the handler believed the dog was alerting them to a VGED during the search, they would declare "found" to the observer, who would then confirm that the alert was either 'correct' or 'incorrect'. The handler could then immediately reward the dog for correct alerts or direct them to continue searching for incorrect alerts before continuing to search the area to ensure it was adequately surveyed. Once the area had been searched to the satisfaction of the handler, they declared the search 'finished'. All assessments were filmed and parameters including search success, search time, number of false alerts and time to detection were recorded.

2.2.5 Live VGED surveys

Once it was established that both live VGED dog-handler teams could effectively locate live VGEDs in situ, teams focused on completing surveys of the rediscovery site and a nearby property. Handlers typically coordinated these surveys by flagging out a search area of approximately 100 m^2 – 150 m^2 at a time. Dog-handler teams then thoroughly searched this area over approximately 8-15 minutes (depending on the habitat and weather characteristics), ensuring dogs effectively smelled all burrows, rock crevices, tussocks and other refugia that may shelter VGEDs. When a dog alerted to the presence of VGED, the handler confirmed presence with a torch or endoscope before the dog was either rewarded (for true positive alerts) or directed to continue searching (for false positive alerts). Burrows screened by dogs during searches that were not alerted to were typically marked with flags or survey tape and handlers would return to these burrows after the search was completed to confirm with an endoscope that no VGEDs were missed (i.e. false negative search result).

Breaks would occur in between searches, as required by the dog, to ensure their welfare and health was safeguarded. Frequent rest breaks were especially important for the dogs' health during high temperature conditions, particularly over summer. During these warmer periods the dogs would wear cooling jackets under their search vests to assist with thermoregulation and water bowls were available near the search area. Searches may have occurred at any time during the day, but closer to midday appeared to be when the VGEDs were most active and therefore likely producing the most odour. Dogs may have searched on or off leash (i.e. a long line), depending on the dog and day, but having them on a 5- or 10-metre-long line did often act as a cue to the dogs that a fine scale search method was required.

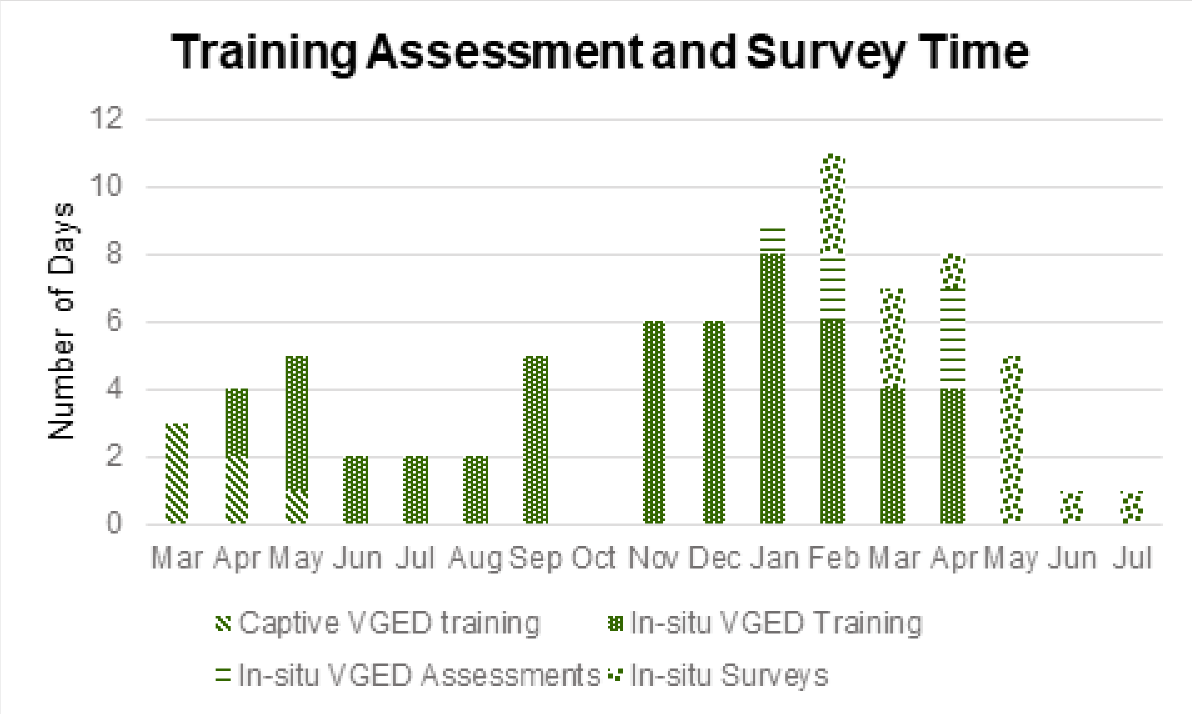
Detection dogs wore a GPS tracking collar during searches that was connected to their handler's GPS handheld system. Dog and handler search tracks were therefore recorded during each search to enable the collection of information on search area coverage, the locations of VGEDs detected or the dogs' alerts. Data on search time required to thoroughly cover the area was also collected.

2.3 Results

2.3.1 Project time investment overview

In total, we estimate that Kip and Daisy participated in live VGED detection training and surveys on a combined total of approximately 160 days (80 days per dog-handler team). Due to the nature of the project, it was deemed necessary for the team to be on site and available for as many training opportunities as possible. Many days on site did not yield opportunities to train on wild VGEDs simply because none were detected by other, traditional survey methods (e.g. tile grids, artificial spider burrows, pitfall traps, endoscope surveys of natural burrows, etc) that could be incorporated into detection dog training. Instead, time was spent learning about the species, the site, traditional VGED survey methods, and also afforded training opportunities for the dogs to search for low odour targets in a systematic manner. A breakdown of time spent on the different phases of this project is presented in Figure 4. Peak field season occurred across summer and autumn and field surveys continue over winter.

Figure 4. Days per month spent on VGED captive training, field training, field assessments and field survey



2.3.2 Captive VGED training

In total, Daisy and Kip were exposed to 8 captive VGEDs during training. Across 6 training sessions in total, both dogs were able to reliably alert the handlers to the VGED among an assortment of non-target containers. After these sessions, both dogs were considered to have achieved the training goal of recognising the odour of captive VGEDs and associating it with a reward, which then enabled field training to commence. The captive training period was intentionally kept short to reduce the likelihood of dogs' becoming specific to the odour of captive VGEDs, thereby increasing the challenge of generalising detection to wild individuals.

2.3.3 Field training

From early November 2023, both live VGED detection dog-handler teams were completing a mixture of training and surveys at the rediscovery site. This typically occurred from one to 3 days per week, until late April 2024. While surveys completed in this time were done prior to initial detectability assessments, handlers were still able to achieve productive outcomes. These continued to focus on search strategy and effective search area coverage in VGED habitat, in between training opportunities that arose from VGEDs located by members of the recovery team. Both the outcomes of training and associated data collection for this field training period focused largely on qualitative, rather than quantitative outcomes.

Before the more intensive search period began, Kip independently located his first VGED in late-September 2023. This individual was originally outside the burrow when dog-handler Naomi Hodgens noticed a change in Kip's behaviour. The VGED was subsequently confirmed to be under the rock that Kip had shown interest in. This detection highlights the importance of handlers being able to read their dog's behaviours, even when a trained alert is not performed, especially in the early stages of field training. Had Naomi not noticed Kip's change of behaviour, this VGED would not have been located.

Figure 5. Not just an ordinary rock: the rock and surrounding habitat where Kip's first VGED detection was made

Source: Naomi Hodgens.

Towards the end of this field training period, a 'hot spot' of active VGED burrows was found at the rediscovery site. Over a two-month period, this 'hot spot' provided an excellent opportunity for dog-handler teams to complete highly valuable training sessions on VGEDs that could be confirmed in burrows by torchlight (without leaving endoscope or large amounts of residual human odour). Here, dog-handler teams participated in approximately 21 training sessions in which they could detect and alert to wild VGEDs in situ that were sheltering in natural burrows. These sessions consisted of both guided and single-blind training sessions, depending on the training goals of the dog-handler team at the time. Almost all of these training opportunities arose from VGED detections that were made by detection dog team members using torches to spot individuals in burrows.

A table cataloguing these detections and burrows is presented in Appendix A.

2.3.4 Initial VGED detectability assessments

After a highly successful period of training on wild VGEDs, the team began completing initial single-blind search assessments after a VGED was detected in situ, without the assistance of an endoscope and without being handled by humans. As these opportunities are low in frequency and difficult to predict, a total of 9 assessments have been completed to date between Kip (n = 4) and Daisy (n = 5). These assessments will continue to be conducted where possible and the data from these additional searches will be combined with that presented here at a later date.

During these initial assessments, dog-handler teams recorded a combined search sensitivity of 100%, detecting 9 out of 9 VGEDs within the search area (Table 1). Dog-handler teams were able to locate the VGED quickly, with the average time to detection (i.e. when the dog alerted to the VGED) being just over 3.5 minutes (Table 1).

Table 1. Results of single-blind dog-handler team searches for live VGEDs sheltering within unknown invertebrate burrows.

Dog-Handler team (dog and handler)	Trial Number	VGED located by dog-handler team	Time to Detection (mm:ss)	Total search effort (time) (mm:ss)
Daisy and Nick	1*	Yes	1:56	7:06
	2*	Yes	1:36	13:35
	3	Yes	0:37	7:25
	4	Yes	2:00	5:57
	5	Yes	7:02	n/a
	Mean		**2:38**	**8:30**
Kip and Naomi	1*	Yes	4:34	n/a
	2*	Yes	4:32	10:15
	3	Yes	5:09	16:02
	4	Yes	5:59	14:29
	Mean		**5:03**	**13:33**
Total mean			**3:39**	**10:34**

*Area contains burrows recently checked with an endoscope

An unexpectedly high total of 18 false positive alerts were recorded by the dogs in these 9 search assessments. Interestingly, 13 (72%) of these false positive alerts were recorded in each of the dogs' first 2 searches, in which VGEDs were detected in burrows via a camera endoscope. Importantly, the majority of these false alerts were directed to several of the other burrows that had been checked with the camera endoscope prior to the dogs commencing searching. In response to this high false positive alert rate in these first 4 searches, handlers decided to only search for in situ VGEDs for assessments using torchlight within a known hotspot. This does not disturb soil or leave other potentially salient residual odours within a survey area as camera endoscopes do. In these last 5 assessments using this new method, only 4 false positive alerts were recorded. We expect that as false positive alerts are almost always associated with human influence within a site, such as residual endoscope odour, these instances will further reduce when surveying novel areas.

2.3.5 Live VGED surveys

Since completing initial detectability assessments, the dog-handler teams have completed 11 days of surveys for live dragons. This includes one day of surveying on a property nearby to the rediscovery site and 10 days surveying across the rediscovery property. While survey efforts have been spread across much of the rediscovery site where teams were permitted access, efforts to date have largely been focused across 3 main areas of the site associated with known VGED activity.

To date, the detection-dog handler teams have located a combined 8 VGEDs within naturally occurring burrows (Figure 6 and 7), all of which have been found across 2 main areas of the property. All detections were made in previously unrecorded burrows. As no VGEDs were excavated postdetection to facilitate an individual VGED's identification (whether it was a recapture or to collect for the conservation breeding program), it is unclear whether these detections represent 8 unique VGEDs or if a portion of individuals had been previously detected. Regardless, this represents a very important milestone in the project and validates detection dog-handler teams' abilities to detect wild VGEDs in their natural habitat. These detections are especially important as they continued to be made in the winter period when VGEDs are less likely to be detected via alternative survey methods (the species goes into torpor during winter).

Of these 8 burrows, 3 could be characterised as being surrounded by grassy vegetation at the time of detection, which was observed to visually occlude the entrance to some degree. A catalogue of these burrows can be found in Table 2. The locations of these burrows in relation to those identified by human surveyors and in detection dog training can be seen in Map 1.

Figure 6. Detection Dog Daisy independently alerting to a VGED in a new burrow during a survey (endoscope in background). This was the first time a dragon had been located in this burrow.

Source: Nick Rutter

Figure 7. Kip and Naomi celebrating with a ball game after Kip independently located a VGED within a new burrow

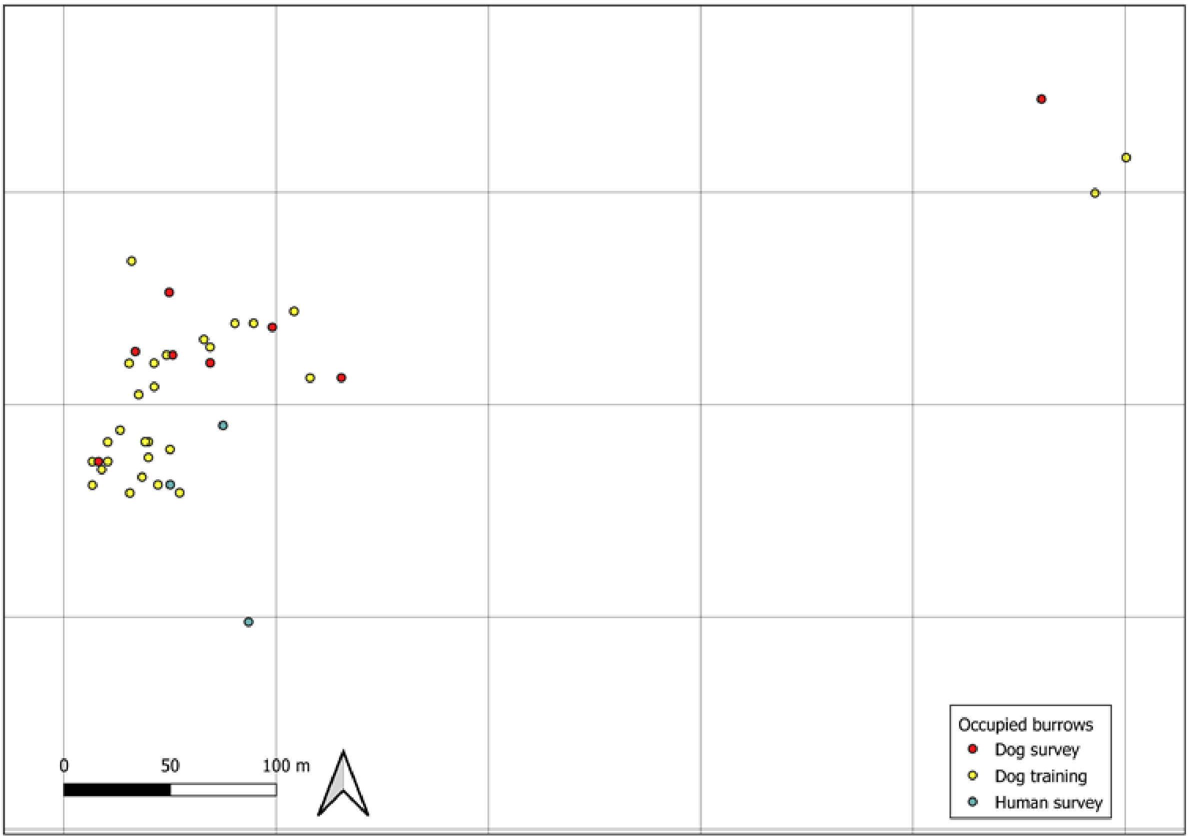
Source: La Toya Jamieson

Table 2. Catalogue of burrows sheltering VGEDs found by detection dog-handler teams during surveys

Date	No.	Photograph	Date	No.	Photograph
7/05/24	1		13/05/24	2	
13/05/24	3		16/05/24	4	

Date	No.	Photograph	Date	No.	Photograph
22/05/24	5		18/05/24	6	
18/05/24	7		18/05/24	8	

Map 1. Distribution of natural burrows housing live VGEDs across the rediscovery property. Burrows detected through human surveyors (triangle), detection dog training (circle) and detection dog surveys (square) are included



Source: Map created by Sakib Kazi (Zoos Victoria). Note that at least 6 burrows sheltering VGEDs found by human surveyors and during detection dog training are absent from this map. Identifying landscape features have been redacted to preserve the anonymity of the site for security purposes.

Detection dog handlers also located a total of 3 wild VGEDs outside of burrows, including one hatchling and 2 adults. While these detections did not result from the searching activity of a detection dog, they are reported here as they incidentally arose from training and survey activity.

One adult individual could not be captured before sheltering in a burrow but was suspected to be a recapture (tail appeared to have been marked). The hatchling and other adult had not previously been captured and either the recovery team or handlers were able to complete the appropriate data collection and processing of this individual before it was released back at the site.

2.4 Discussion

2.4.1 Captive VGED training

Initial training on captive dragons in PVC pipes represented a relatively small component of the overall training time spent on this project. Both dogs were familiar with learning new odours being presented in various containers and successfully incorporated VGED odour into their detection repertoire. While the method of presenting detection dogs with new target species housed in containment employed in this study is a commonly used training technique, we contend that this step was of limited value in the overall preparation of Kip and Daisy to search for wild VGEDs in situ. It is possible that the odour profile of VGEDs begins to change during their time in captivity due to changes in diet, habitat and incidental exposure to background odours of their enclosure. Our anecdotal experience with this training step leads us to conclude that the goal of rapidly preparing live VGED detection dogs could be fast tracked by beginning training on VGEDs sheltering in burrows in situ if these training opportunities are available. It is entirely possible that other detection dog-handler teams may indeed benefit from training on captive VGEDs, especially if in situ training opportunities are limited or unavailable, as is often the case in threatened species detection. It is important to highlight that being able to detect VGED in containers does not necessarily translate to detecting wild VGEDs in a variety of different presentations, including in tussocks, under rocks or within burrows.

2.4.2 Field training and assessments

The unpredictable and disjointed nature of opportunities available to train on live VGEDs in situ, combined with their cryptic nature and low odour volatility meant that this important and productive training stage was often quite challenging. Training progression over these months was very often nonlinear.

The cryptic and low-density nature of VGEDs in the wild meant that early in situ training opportunities on wild VGEDS almost always followed a large amount of human activity and associated residual odours around the VGED location (such as kneeling, using an endoscope, measuring burrow characteristics, etc.). This inadvertent and largely unavoidable pairing of VGED odour with human odours meant that dogs learned that the presence of VGEDs was often paired with other olfactory cues, which may have been more salient than the VGED odour itself. This information guided a review of training that focused on creating more training opportunities to ignore these other odours associated with VGEDs and to focus on VGED odour alone. This resulted in improvements in the dogs' understanding of the task and increased performance overall, particularly in the reduction of false positive alerts.

These training and assessments have also provided insights into effective detection dog search strategy for VGEDs. Importantly, it seems that at this stage in their progression that dogs need to have their nose within centimetres of a burrow entrance to detect a VGED (currently the most likely detection location). Consequently, teaching the dogs to conduct fine scale pattern surveys and to actively check potential burrows has proven incredibly important in successful VGED detection to date. As the dogs gain more experience, we anticipate that both the detection distance and survey speed could increase, depending on environmental variables (e.g. wind speed, air humidity and temperature), and vegetation density. We also expect that the odour profile and odour availability of VGEDs could display seasonal changes and we continue to capitalise on training opportunities that incorporate as many of these variables as possible in order to promote the dogs' understanding of VGED odour year-round. We plan to incorporate the use of radio telemetry equipment to monitor released VGEDs on the rediscovery site and this will enable us to compile a sizable dataset of single- and/or double-blind searches by which we can gain a strong empirical understanding of team search performance and requirements.

Both live-animal detection dogs have also had several training opportunities to search for VGEDs concealed in tussocks or moving through the grassland in which the dogs demonstrated recognition of the target odour and were able to track moving VGEDs across a distance of several metres after they had sought shelter. We anticipate that there may be more opportunities for dogs to detect VGEDs sheltering in tussocks or other refugia in the upcoming spring/ summer season when VGEDs are more active. It is possible that dogs could detect VGEDs in these scenarios from a greater distance. We will continue to explore these factors over successive surveys.

It is important to highlight that dogs have consistently ignored other reptile species naturally present within the VGED habitat throughout all field activities in this project. This demonstrates that dogs' alerts are specific to VGEDs, and false positive alerts observed in this project are more likely a result of odours associated with human activity, rather than a failure to discriminate between VGEDs and other reptile species. As dogs' learning, training and behaviour is constantly ongoing and subject to reinforcement, we have been focusing on reducing false alerts by not rewarding interest in these odours and redirecting dogs to continue searching, as well as providing improved learning opportunities. We anticipate false alerts to these odours will continue to decrease with experience.

Whilst the dog-handler teams are now in the surveying phase of the project, it is important that 'top up' training sessions continue to occur on wild VGEDs to maintain the dogs' motivation and understanding of the search task, especially after prolonged periods without VGED detections.

2.4.3 Field surveys

The detection of 8 VGEDs within 11 survey days are highly promising and demonstrate the potential benefits and success of this new VGED detection method. This also validates the training and assessment methods employed, even though modifications to these methods continue to occur based on increased knowledge.

We have demonstrated that detection dog teams are able to non-invasively provide evidence of species presence and VGED location in real time, including those sheltering within burrows, tussocks and under rock formations. Unlike other detection methods, trained detection dog-handler teams do not require installation at a site and can instead begin surveys straight away. Detection dog-handler team surveys may therefore be a beneficial method of screening new locations to determine whether it would be valuable to install traditional survey infrastructure. Detection dog-handler teams therefore present a valuable complementary VGED detection method that will further add value to, and hopefully assist with gaps in, current survey methods.

Based on our findings, detection dog surveys may be potentially valuable during cooler periods when VGEDs are less active and therefore less likely to be detected through passive and active survey methods which rely on the lizards moving. This may also be relevant during periods when vegetation is dense. Nearly half of the VGEDs (38%) detected by dog-handler teams were found in burrows with entrances surrounded by grassy vegetation. While vegetation levels are subject to seasonal variation and grazing pressure, increased vegetation can visually occlude burrow entrances, making them difficult to spot and therefore check with an endoscope. As dogs are detecting VGEDs within burrows using olfaction rather than vision, these factors are less influential on their detection success. Dogs may also be less subject to potential human survey biases, such as the perception that particular burrow characteristics may lead to a higher or lower probability of VGED detection. Ongoing data collection and comparative detection trials are required to determine if there are differences in the occupied burrows detected by detection dogs and human visual surveyors.

It is possible that we were able to detect VGEDs that were less likely to be captured by other monitoring techniques due to individual preferences in habitat or burrow refugia. Similarly, trained detection dogs have enabled the detection of VGEDs not sheltering in artificial refuges, which may have been missed by existing survey methods. It is possible that the probability of a given survey method detecting a VGED is influenced by environmental variables, such as those associated with daytime temperatures. VGEDs may preferentially use roofing tiles as shelter during early morning to assist thermoregulation, for example (P. Robertson, personal communication), whereas they may then choose to avoid sheltering under tiles during the heat of the day in preference for foraging, seeking shelter in tussocks or in underground burrows where detection dogs may be more successful in locating them. In this way, the overall chance of detecting VGEDs in a given area may be greatest when multiple detection methods, including detection dog-handler teams, are deployed.

Over the course of this project, the human members of the detection dog team have been trained in VGED handling and sample collection, which has enhanced the capacity of the overall survey team on the rediscovery site. The handlers have also been trained to complete trap checks, which has further added value. We see great value in this skill development, and it will further enhance the contribution of the detection dog team to VGED conservation when surveying additional properties.

Whilst we have demonstrated many benefits of employing detection dog-handler teams, no survey method is perfect. Completing field training and surveys relatively consistently over the last 12 months has highlighted some limitations to detection dog surveys for VGEDs. High temperatures are a particularly relevant limiting factor to dog search duration and likely search performance. High temperatures typically decrease detection dog efficacy and performance and increases the required search time (de Oliveira et al. 2021; Deak et al. 2020). The optimal search temperature and the extent to which dogs are influenced by higher temperatures is likely dependent on the individual dog (Deak et al. 2020). We aim to gain a more complete, empirically-based understanding of these influences on detectability over the coming warmer period associated with Spring, Summer and Autumn when VGEDs may be more active (September 2024 – May 2025). To assist with overcoming this limitation and maximise effective survey time, the detection dog teams worked to complete surveys in the early morning before temperatures exceeded 25 to 30°C. By combining this with measures such as shorter search duration, dogs wearing evaporative cooling vets, and maintaining a schedule of sufficient rest and hydration, we have developed a strategy of completing surveys with the dogs in these warmer temperatures (approx. 30°C) whilst maintaining the high welfare standards of the Zoos Victoria detection dog team. While it is possible that these conditions are more favourable for VGED activity, it is not yet understood how this influences detection dog performance or their ability to detect VGEDs. Finally, we are currently demonstrating that it is possible to detect VGEDs sheltering in burrows outside of peak activity seasons associated with warmer weather. Indeed, dog-handler teams have recently detected 3 VGEDs on cooler winter days that were sheltering in their burrows. As we learn more about this, effective conditions for detection dog team surveys may be extended to cooler months.

Now that it has been established that dogs can be successfully trained to detect VGEDs, it may be possible for these dogs to generalise this training to other grassland earless dragon species, such as *Tympanocryptis lineata*, *T. osbornei* or *T. mccartneyi*. In separate studies, detection dogs have been able to successfully generalise their training to similar, yet distinct species, to their original target (Rutter et al. 2021; Jamieson et al. 2024). If dogs were able to show evidence of this olfactory generalistion in *Tympanocryptis* sp., it could represent a valuable means by which detection dogs could be employed to assist in the conservation of other imperilled grassland earless dragons. This would likely also result in a short timeline between training and successful field deployment. This would therefore be a valuable future direction of research.

Moving forward, the Zoos Victoria team plan to add 2 additional dogs to the live VGED detection team to continue to build survey capacity across new sites in Victoria, as well as collaborating with interstate agencies to explore using the dogs to detect other *Tympanocryptis* species. The learning and subsequent methodologies developed during this project will be invaluable in streamlining training for the next cohort of dogs trained to locate live VGEDs. We believe that training 2 new live VGED detection dogs can be done in a way that avoids extraneous odour cues by focusing on VGED hot spots during periods of increased seasonal activity. Capitalising on these opportunities has enabled relatively fast and effective training of the current VGED detection dogs over a period of approximately 2 months. We anticipate that if we have access to a similar density and duration of VGEDs, that this training opportunity could enable the 2 new live VGED detection dogs to be trained and surveying in a similar timeframe.

3.0 Detection dog teams trained to locate Victorian grassland earless dragon scat

3.1 Introduction

Outside of live trapping individual VGEDs and collecting any scats they may drop when being handled, there is no current survey method for VGED scat. This is due to several factors including the small size of the scat; the time investment required to survey for specimens this small; the lack of distinctiveness between VGED and other similar sized reptile scats; and the prioritisation of detecting live individuals in a species only recently rediscovered. However, there are several advantages of collecting VGED scat, including ability to determine diet and species presence in a non-invasive way.

There is extensive evidence that highly trained wildlife detection dog-handler teams are capable of detecting small target samples, with a high level of precision. This includes species from plant, animal, fungi and bacteria kingdoms (see Grimm-Seyfarth, Harms and Berger, 2021 for a review). The key objective of this part of the project was to determine if detection dog-handler teams would be an effective survey method for VGED scat detection.

Our aims were to:

- Measure detection dogs' sensitivity and precision to VGED scat when presented among non-target scats
- Measure detection dogs' ability to generalise from captive to wild VGED scat
- Determine search style required to effectively detect VGED scat
- Measure search time required and search sensitivity for novel VGED scat
- Determine factors that may influence field survey success of VGED scat detection dog teams.

3.2 Methods

3.2.1 Detection dogs

Two detection dogs were involved in this study. Sugar was to be the primary VGED scat detection dog. At the beginning of this project, Sugar was a 3-year-old German shorthaired pointer cross Springer spaniel, with experience in scent detection (including brief periods of field surveys for truffles or serrated tussock (*Nassella trichotoma*) prior to her involvement with the Zoos Victoria Wildlife Detection Dog Program). Whilst we originally only intended to have one scat detection dog, given the challenges of visually discriminating VGED scat samples from other similar sized reptiles, it was determined that it would be beneficial to have a second dog to further validate Sugar's detections. Moss was to be the support VGED scat detection dog, due to his experience and success on scat projects and his natural specificity to his targets. At the beginning of this project Moss was a 4.5-year-old Labrador with experience surveying for broad-toothed rat (*Mastacomys fuscus*) scat and active platypus burrows, in addition to research experience detecting oestrus in Tasmanian devil scat.

Both dogs were familiar with and had a reinforcement history using the training equipment employed in this project, and both dogs had been previously trained to perform a passive alert behaviour to their target sample (i.e. sit and stare at the target). Sugar was motivated by food, toys and verbal/physical praise, and Moss was motivated by food and verbal/physical praise. A variety of these reinforcers were utilised throughout training, assessments and field surveys. Both dogs were trained and handled by their primary trainer, Dr La Toya Jamieson, who they had a strong working history and relationship with.

3.2.2 Scat samples

During initial stages of training, scats from a variety of VGEDs, including sub-adults and adults, were collected from the VGED population at Melbourne Zoo that had recently been brought into captivity to establish a conservation breeding program. Samples were initially frozen to prevent odour degradation, including mould growth, and to keep the samples as fresh as possible to preserve their natural odour as much as possible. Later in training, however, unfrozen samples were used. It was determined that if scats were allowed to naturally air dry then mould growth would not occur unless the samples were wet during training.

Samples used in the early stages of training, especially during odour imprinting, were collected from the VGED enclosures using clean metal tweezers and were placed into individual clean glass jars with airtight lids. The jar was then labelled with the individual the samples was collected from, the date of collection, whether the individual was an adult or sub-adult and sex (if known). At later stages in the training, samples were collected and stored in a variety of containers, including plastic zip-lock bags, paper bags, travel mugs, etc, to ensure the dogs were not becoming specific to only detecting VGED scat that had been collected with metal tweezers and stored in glass.

To further provide sample variety, samples were presented to the dogs after they had been wet, become mouldy or had begun to degrade, in addition to fresh intact samples.

Scats were collected from wild VGED, either after they had recently been brought into captivity and it was their first defecation (therefore the diet within the scat would have been from wild sources) or when wild VGED were trapped and were being temporarily held to collect morphometric measurements (measurements of size, shape and form). As these wild VGED scat samples were very rare to collect, and that all wild VGED scats were being sent for diet analyses, these scats could only be trained on in containers to prevent the scats being unintentionally damaged during training.

Non-target scat samples (i.e. samples that are not from VGED) were collected from both captive and wild reptiles. Some of these species included blue-tongued lizard (*Tiliqua scincoides*), red-barred dragon (*Ctenophorus vadnappa*), frilled-neck lizard (*Chlamydosaurus kingii*), eastern striped skink (*Ctenotus robustus*) and tussock skink (*Pseudemoia pagenstecheri*). As with the VGED scat, these samples were stored in a variety of containers, both frozen and non-frozen.

3.2.3 Odour imprinting and discrimination training

Odour imprinting is the process in which a detection dog is trained to associate the smell of a target (in this case, VGED scat) with a reinforcer. Both Moss and Sugar have completed this training process on several targets previously and were therefore experienced with the approach.

For this training, frozen VGED scat samples were housed in PVC containers, with mesh lids (called scent pots), on a metre long wooden board (called a scent board). When the dogs approached the

VGED pots they were reinforced for smelling and eventually alerting (sitting and staring at the sample) to the VGED scat. Empty pots were added to the scent board so the dogs had to make a choice about which to alert to before being reinforced. Once the dogs were reliably alerting to VGED scat from multiple individuals, other non-target samples, including reptile scats (non-target scats), were then introduced for the dogs to learn to discriminate. Dogs were only reinforced for alerting to the VGED scat and reinforcement was withheld if they alerted to non-target samples.

VGED scats used initially in training were frozen. When additional fresh scats were available for training they were introduced after completing generalisation assessments (see section 3.2.4). Finally, dogs were trained on wild VGED scats.

Training sessions were short, typically up to 5 minutes in length, however, multiple training sessions could occur in a day. Sessions were filmed and data including the session number, samples used (including what VGED scat the sample was from and whether that was a novel individual or sample), and the dogs alerts to target and non-target samples were collected. Training was initially completed indoors, before moving outside once the dogs were reliably alerting and discriminating to VGED scat.

3.2.4 Target discrimination and generalisation assessments

3.2.4.1 Training interim assessment

Before Sugar transitioned from detecting VGED scat on the scent board to detecting in a search area, her search performance was assessed. This training interim assessment occurred across 5 trials (i.e., 5 scent board searches). Each trial involved 5 pots that either contained a target or non-target sample. Only one target pot was ever present on the scent board. The target and non-target samples used were novel to Sugar, with the order of the samples being randomly allocated by a research assistant. Assessments were single-blind, with Sugar's handler being blind to the order and location of the target sample. When Sugar would alert, her handler would say "found" before the research assistant would confirm whether the alert was correct or not. Sugar was reinforced for correct alerts only.

This assessment occurred once inside and once outside (at the location where Sugar's outdoor VGED scat training would begin). Across each assessment, Sugar was exposed to 5 target samples and 20 non-target samples (10 targets and 40 non-targets combined across both assessments). For Sugar to progress to training on VGED scat not contained on the scent board, she was required to alert to all available VGED scats and perform no false alerts (i.e. perfect sensitivity and precision).

3.2.4.2 Generalisation assessments

Both dogs were assessed in a similar way to the method outlined above, where scent boards were used to present Sugar and Moss with novel target and novel non-target samples. Similarly, all assessments were single-blind. These assessments were completed to measure how the dogs were performing on their current training type (e.g. frozen VGED scat) before assessing whether the dogs would generalise on first exposure to a novel target type (e.g. fresh VGED scat or wild VGED scat). Before use in assessments, pots were cleaned in hot water to remove organic materials, were sterilised with 70% isopropanol spray and were left to dry.

Assessment 1 was comprised of 6 searches of the scent board – 5 searches contained novel frozen VGED and non-target reptile scats, and one of the searches (order randomised) contained a non-frozen, fresh, novel captive VGED scat.

Assessment 2 was completed in the same manner to assessment one1, except the dogs were assessed over 5 trials using the fresh captive VGED scat and on one randomly organised single trial, a wild VGED scat was used. As with the training interim assessment, the dogs' alerts were recorded, and a research assistant responsible for organising the assessments would confirm whether the dog was correct after the alert was called by the handler, before the dog was reinforced.

Throughout the course of the project, the dogs were tested, as above, each time a new wild VGED scat was available for training. Once an initial generalisation assessment was completed with the wild VGED scat, it was then used for training.

During all assessments, the dogs' alerts were confirmed and recorded by a research assistant, and they were rewarded for true positive alerts.

3.2.5 Fine-scale search training

3.2.5.1 Training odour (Kong)

In conjunction with odour imprinting and discrimination training, Sugar also received training to complete fine-scale searches. Moss did not require this training as he was already experienced in this search style. Based on the size and low odour volatility of the VGED scats, it was assumed that fine-scale searches employing a high sniffing frequency would be required for the scat to be detected, especially in complex environments. Building a detection dog's skills and stamina associated with fine-scale searches is essential for search performance and also welfare. Detection dogs that are not accustomed to this search style and intensity will tire quickly, potentially posing an increased risk of overheating, and may have a reduction in search motivation. Therefore, building this stamina before field deployment is essential.

To begin this training, our training odour (Kong rubber) was used. Small pieces of Kong, ranging from 3 mm to 5 mm, were used in small flagged out areas (typically 5 m x 5 m, or 25 m^2 areas). The smallest pieces of Kong were used so Sugar would have to search slowly in order to detect the odour. A different search cue was also given to highlight the difference in the search style requested of Sugar – the handler would double-tap the ground with her fingers at the spot where Sugar was being asked to begin searching, and the word "search" was the verbal cue. Search times started out very short before Sugar would either detect the scat or the search was cued to finish ('Sugar, finished!'). As the action of searching provided a great deal of reinforcement to Sugar, care had to be taken to not always finish a search after locating the target and to allow her to continue searching after the find. Search times continued to increase until Sugar could reliably and comfortably perform fine-scale searches in a single area with a high sniffing rate for 15 minutes before having a break.

To assist with our understanding of search effort required to detect small pieces of Kong rubber, that may be similar to search effort to detect VGED scat, single-blind area assessments were completed before VGED scat search training began. These assessments also provided an opportunity to evaluate the reliability of Sugar's emergency cues, as outlined in the Australasian Conservation Dog Network's Detection Dog Team Evaluation Guidelines (https://conservationdognetwork.com.au/guidelines/). Assessments were completed across twelve 5 m x 5 m (25 m^2) flagged areas, containing zero to 2

Kong pieces. Time to detection, total search time, Sugar's alerts and performance of safety cues were recorded.

3.2.5.2 VGED scat training searches

Once Sugar successfully completed her interim training assessments, the VGED scat was then moved from the pots on the scent board and were placed in the natural environment for training sessions. Training initially prioritised ensuring that Sugar recognised the odour of the VGED scat without the stronger smell of the PVC plastic pot. Early training stages therefore involved training techniques whereby her handler would verbally 'mark' her as correct as soon as her nose was close to the VGED scat before reinforcing that behaviour. Training also focused on shaping Sugar's alert so that it was close enough to the scat that the handler could visually locate it, which is very difficult in the environment, without the scat being moved or damaged. Sugar's alert behaviour therefore had to be slightly modified so that her nose hovered approximately 1 to 2 cm from the scat, which is closer to the target than her previous alert behaviour (Figure 8).

Figure 8. (L) Sugar sitting and alerting to a VGED scat in a rock crevice. (R) Sugar alerting by pointing her nose to the VGED scat.

Photo credit: La Toya Jamieson

Area training then progressed to having the scat in slightly more complex environments, where longer search times were required before detection. To assist Sugar to complete the fine-scale searches more effectively, she was placed on a lead when searching, which became another cue to her for the search style required to locate the target. A rope that circled the perimeter of the area was also used instead of flags, so Sugar had a clearer visual barrier that defined her search area. Due to the level of search intensity required to detect the VGED scat, Sugar received frequent breaks during searches, as with the Kong training, and her search stamina gradually increased without reducing search sensitivity.

Scat search training was originally completed in the Coranderrk Bushland Reserve at Healesville Sanctuary, across multiple habitat types (this is not thought to be part of VGED historical distribution but is a private area accessible to the Wildlife Detection Dog program for dog training), in addition to training on captive scat samples at the VGED rediscovery sites after familiarisation and habituation training.

3.2.6 Field training and surveys

Initial field training and later surveys began at the VGED rediscovery site late-October 2023, with field surveys intensifying from January 2024 onwards. As with training, field searches typically involved a 5 m x 5 m (25 m^2) area roped off at a time for the dog-handler team to thoroughly search, before moving the rope into another area.

Due to the intense nature of the searches, Sugar would rest briefly on a mat in between searches whilst a new area was being selected. A water bowl was often available just outside the search area so Sugar could choose to have a drink as required during searching, which was particularly important over summer.

Search areas around active VGED burrows were prioritised to maximise the likelihood of a scat being present in the general area. Due to the low target density, training VGED samples were intermittently placed out in search areas to firstly allow Sugar to be reinforced for her work, and secondly to confirm she was still working effectively.

3.2.7 Area search assessments

Search assessments were completed concurrently with field surveys. These assessments mimicked those of the training odour Kong assessments and the way in which the VGED scat detection dog team was currently searching in the field. Assessments were completed across May – July 2024.

Twenty-one search assessments were completed within 5 m x 5 m (25 m^2) areas, with zero to 3 targets present. Novel adult captive VGED scats were used and were placed out by the research assistant in the roped search areas a minimum of 30 minutes before searches began. Scats used during the scat longevity study (see Section 4) were used during this study, in addition to scats that were not degraded, to determine if Sugar would generalise her learning to degraded scats. The research assistant walked randomly across all areas, including blank areas, as well as stopping and spending an extended time in multiple locations to ensure Sugar was not using human odour as a cue to where the scats were located.

Assessments were filmed and search location, sample type (fresh or degraded), how the sample was stored, date sample was collected, environmental conditions (wind speed, relative humidity and temperature), search start time, time to alerts, total search time, targets found, targets missed, and false alerts were collected. At the end of the search, the handler was asked what percentage of the area they believed was searched and how many targets they believed were present based on the dogs' behaviours.

3.3 Results

3.3.1 Target discrimination and generalisation assessments

3.3.1.1 Training interim assessments

Sugar completed the training interim assessments after completing 8 training sessions. Across both the indoor and outdoor assessments Sugar achieved 100% sensitivity and 100% precision, alerting to all available VGED scats and completing no false alerts (i.e. alerts to non-target samples).

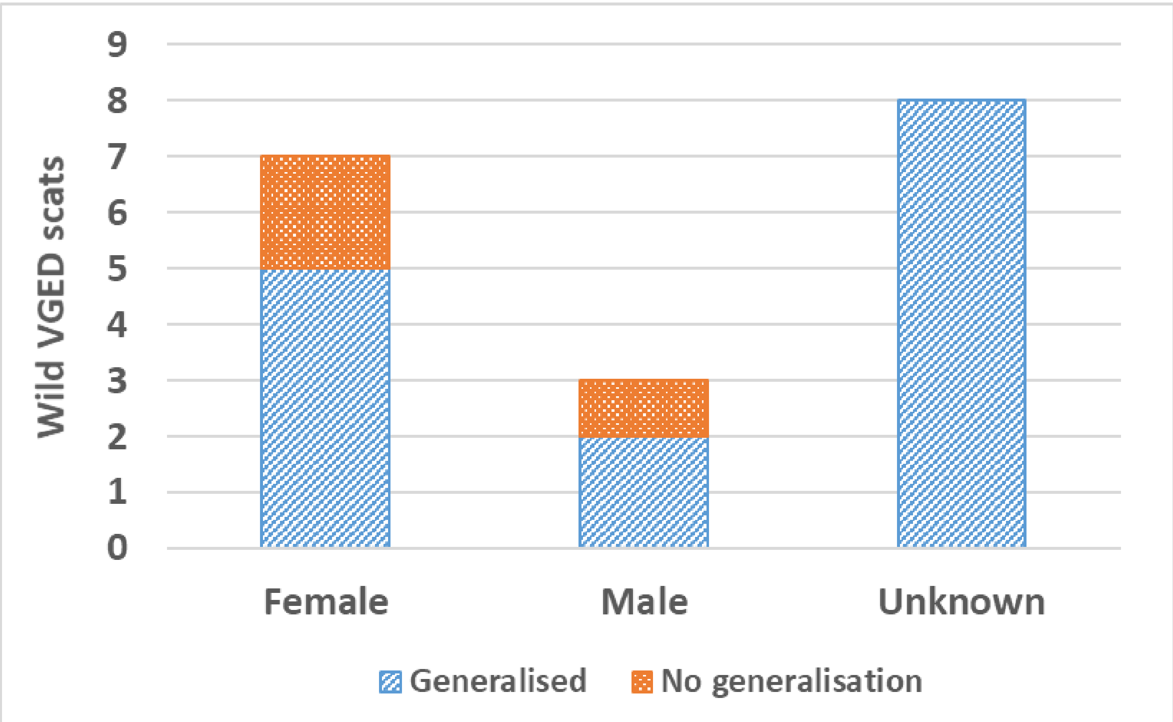
3.3.1.2 Generalisation

Assessment 1: Both dogs correctly alerted to all 6 targets across the searches and completed no false alerts, therefore achieving a sensitivity and precision of 100%. Both dogs were able to generalise on the first exposure to a captive VGED scat that had never been frozen, after only being trained on scats that had been frozen.

Assessment 2: Both dogs correctly alerted to the 5 captive VGED scats and completed no false alerts during these trials. However, during the one trial involving wild VGED scat, neither of the dogs showed evidence of olfactory generalisation (i.e. they did not alert) to this wild scat. This indicates that there is an evident difference in the odour profile of captive vs wild scat, potentially due to dietary differences.

Wild VGED scat: Sugar was tested on 10 wild VGED scats and generalised on 8 (80%) trials. Moss was tested on 8 wild VGED scats and generalised on 7 (87.5%) trials. The sex of the VGED that the scat was from did not appear to influence generalisation ability (Figure 9).

Figure 9. Number of times Moss and Sugar combined generalised or failed to generalise to VGED scat depending on whether the scat was from a female, male or unknown sex



3.3.2 Fine-scale search training

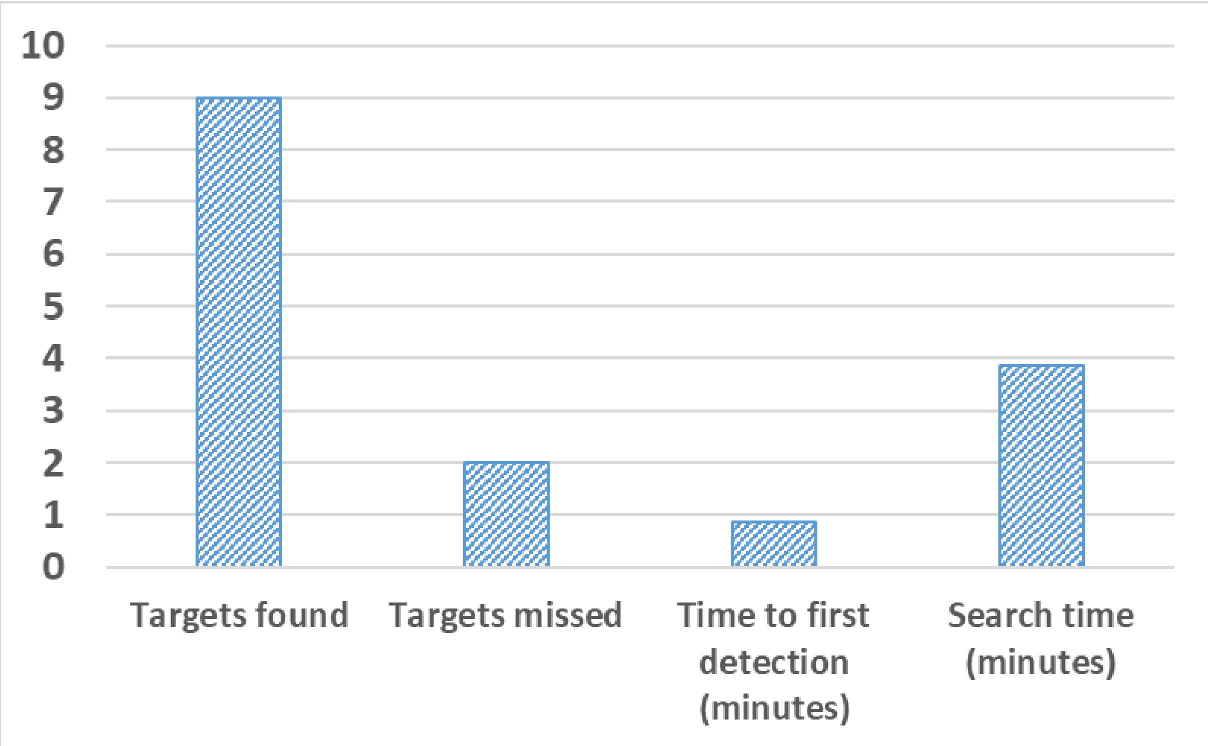
3.3.2.1 Training odour assessments

After completing 12 area searches (all conducted off-lead), Sugar recorded a sensitivity of 81.8%, detecting 9 of the 11 Kong pieces (Figure 10). Whilst Sugar did miss 2 targets, she always confirmed whether an area contained any targets at all and only failed to detect the second target in the area. We took this as a learning opportunity to increase the number of targets within training areas in the future.

Sugar completed no false alerts, even in the 4 blank area searches (i.e. areas with no targets), and therefore scored 100% precision. Additionally, Sugar performed 100% of her cued safety behaviours (2/2 recalls and 2/2 emergency stops) whilst off-lead.

Sugar's average time to first detection was 52.6 seconds (range: 10 – 127 seconds), and average total search time was 231.2 seconds (range: 150 – 338 seconds). The average time searched before the handler declared an area 'blank' was 170.2 seconds (range: 150 – 215 seconds). Overall, Sugar's search effort was 15.4 minutes per 100 m^2.

Figure 10. Total number of targets detected and missed during Kong assessments, and the average time to first detection (minutes) and average search time (minutes)



3.3.2.1 VGED scat training searches

Throughout VGED scat training, it was evident that a high level of search intensity was required for either Sugar or Moss to detect the scat within the environment. Dense grass, tussocks or wet vegetation increased search times required before detection, especially when ambient temperatures were lower. Typically, there also had to be a very small distance between the sample at the dog's nose before they would first detect odour.

Whilst a high sniffing rate was required for detection, Sugar was able to search areas very thoroughly and was able to alert the location of the scat with a high level of precision. Sugar's success at this stage of training, with a large variety of VGED scats, was the catalyst to begin field training.

3.3.3 Field training and surveys

Typically, the goal of field training is to bridge the difference between training on samples that had been influenced by humans (e.g. captive scat or wild scats in containers) and samples naturally occurring in the environment. Unfortunately, due to the difficulties in visually confirming whether a wild reptile scat found in situ was deposited by VGED or a similar sized reptile, this stage in training was not possible. In the absence of this training opportunity, field training and surveys continued, in addition to training on captive scats within search areas and new VGED scats in containers, with the hopes of increasing the dogs' likelihood of naturally generalising to wild VGED scats. Field training and surveys occurred similarly to training, where 5 m x 5 m (25 m^2) search areas were roped off, and the area thoroughly searched (Figure 11).

Field training and VGED scat surveys occurred predominantly between December 2023 and April 2024. Over 100 search areas were covered during this period. The average search time was 8 minutes and 40 seconds. Search duration was typically limited by the high temperatures and the style of searching required to maximise search sensitivity.

Early in the season when the ambient temperature was warmer, we began searches early in the morning before temperatures rose. However, we soon realised that VGEDs were typically more active nearer midday during these warmer hours. We therefore anticipated that there would be a higher chance of scat being deposited during these times, so searches were moved to typically late-mornings and midday.

Safeguarding the detection dogs' health and welfare during these searches was essential. Having these short searches, followed by short breaks before searching again was essential. Evaporative cooling vests were placed under the dogs' search jackets to further assist with cooling and water bowls were placed outside of the search area for the dogs to access as required. The dogs' behaviours, including panting rate and early signs of lethargy, were constantly monitored to ensure overheating did not occur.

Areas where active VGED burrows were recently discovered were prioritised for scat surveys to maximise detection success during the short window of optimal search conditions and stamina during the day.

Sugar's handler could tell when she had found a non-target reptile or bird scat during a search, based on her sniffing intensity and could observer that she had 'checked' the scat and decided it was not the target. Later in the season, when Sugar had not yet alerted to scats in situ, any scats that Sugar spent a long-time checking, but did not alert to, where still collected to be analysed in the future to determine if they were from a VGED. On several occasions, if a scat looked very similar to a VGED scat, but Sugar did not alert, we would then search the area with Moss to see if he believed it was VGED scat. On one occasion Moss did alert to a scat and this sample was collected.

Figure 11. Sugar surveying for VGED scat at the rediscovery site within the roped search area

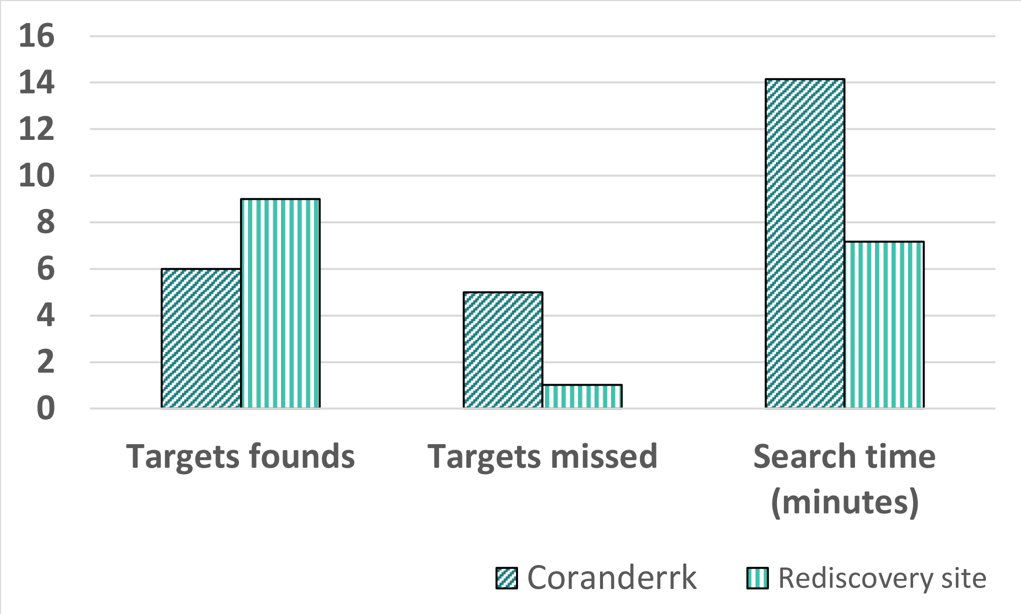
Source: La Toya Jamieson

3.3.4 Area search assessments

Of the 21 assessments, 11 were completed in the Coranderrk Bushland Reserve and 10 were completed at the VGED rediscovery site. When all search assessments were combined, Sugar achieved a sensitivity of 72.7%, locating 17 of the 22 scats (Figure 12). When the assessments were separated by location, Sugar had a higher sensitivity at the rediscovery site (n = 10, sensitivity = 90%) in comparison to the Coranderrk Bushland Reserve (n = 11, sensitivity = 58.3%).

Sugar had a combined mean search time of 10.83 minutes. Sugar's mean search time at the VGED rediscovery site, however, was lower (mean = 7.18) than in the Coranderrk Bushland Reserve (mean = 14.15).

Figure 12. Total number of VGED scats detected and missed at the assessment sites, and the average search time (minutes)



We found that the freshness of the scat did not influence Sugar's likelihood of detection, as she spontaneously generalising to 50-day-old, degraded scat, nor did the use of degraded scat lower her detection ability. All scats during assessments had, however, been in storage for several months, with no fresh scats used.

Detection distance (i.e. distance from the VGED scat when Sugar first detects odour) across all searches was very small. Sugar typically had to have her nose directly over the scat or a maximum of 30 cm away, to detect odour. This small detection distance is correlated with the search time required to complete searches. Detection distances appeared to be especially small during searches in the Coranderrk Bushland Reserve when the ground was wet and the grass thick.

3.4 Discussion

3.4.1 Training

Based on the training methodology used, both dogs were able to learn to discriminate between VGED scat and other reptile scat in controlled trials with a high level of precision. Having this high level of specificity is essential during field surveys when scats cannot be visually identified from other reptile scats of similar sized species. There is a potential, though, that including non-target reptile scats during training in these presentations may influence how the dogs respond to these samples in the field, as presenting them in training may increase the potential value and relevance of these samples. At this stage, however, our field results do not show evidence of this occurring.

Throughout training it was evident that a fine-scale search strategy would be required to successfully detect VGED scat and minimise the chances of false negative search outcomes. Dogs therefore are required to move slowly and systematically when searching to avoid missing targets. These types of searches are highly strenuous and search stamina needs to be increased gradually. Due to the challenges of this search strategy, it is beneficial to employ dogs in this role who are motivated to search in this style. Having dogs that were not suited to completing fine scale searches would have likely changed the chance of search success and may have also increased frustration in the dogs.

Training also highlighted the importance of dogs reliably preforming a passive alert to their target in order to pinpoint exactly where the scat is located. These scats can blend into a natural environment exceptionally well, so being able to have our dogs point precisely to the samples with their noses was highly valuable.

Whilst Moss received fewer training sessions than Sugar, both dogs were able to demonstrate a high level of performance during scent board discrimination assessments. Based on the challenges Sugar had generalising from captive to wild VGED scats, it was hoped that spending fewer sessions on captive scat would have benefited Moss's ability to generalise to wild scat. It is unclear from our results if this was helpful, as neither dog successfully generalised to wild VGED scat on first exposure. However, this was the only instance where Moss did not generalise to a novel wild VGED scat on a scent board (successfully generalising to all other scats he was assessed on), whereas Sugar only began generalising on her third wild VGED scat presentation.

If sufficient samples were available, it would be recommended to only train on wild VGED scat in the future during the imprinting phase of training, before introducing captive scat to be used during searches (if wild VGED scat could not be trained with outside of containers). It is hard to determine how valuable these scent board presentations of new wild VGED scat are, in comparison to how these scats would smell if they were not housed in containment. Having a limited amount of training samples is, however, typically the reality of training detection dogs on critically endangered species. Continuing to increase the dogs' likelihood of generalising to wild VGED scat is therefore highly beneficial, as is having a second dog to further verify any scats detected in situ.

3.4.2 Assessments

Sugar was able to perform with a fairly high level of sensitivity during the VGED scat area assessments and was generally able to confirm species presence in a site, even if a second or third scat sample was not detected. It is important to examine the sensitivity scores in relation to search time, especially at the 2 different locations. Sugar had a higher search effort and lower sensitivity within the Coranderrk Bushland Reserve compared to the rediscovery site, where her required search time was almost half, and she had a near 100% sensitivity. This difference may be due to several factors, including environmental and habitat factors. The wind speed was typically much faster at the rediscovery site during assessments, which may have increased the possible detection distance, and therefore reduced the amount of time required to cover the area. Similarly, the vegetation within the assessment sites at the rediscovery site was much sparser and drier than at the Coranderrk. Having drier, more open vegetation likely aided in odour dispersal and accessibility.

Whilst the search time required at the rediscovery site was lower, overall, the search times required to thoroughly survey the area was high. Increasing the search area size typically naturally reduces the time spent searching per square metre, however, it is currently not known how that would influence search success. Choosing whether to increase the area size, and potentially reduce detection sensitivity, would depend on the purpose of the surveys. If the aim was to detect and collect as many scats as possible, then having a higher search effort to increase likelihood of detection is warranted. If the aim is to determine species presence, then searches would be able to cease after the first detection, which typically was within several minutes or less. Considering that there is no other current scat survey method, these search times might be considered less relevant.

Overall, the assessments highlighted that Sugar was able to reliably detect novel VGED scats with her handler having no knowledge of sample presence or amount. These results, especially the findings at the rediscovery site, provide an estimate of how Sugar would perform during operational VGED scat surveys if wild scat was present.

3.4.3 Field training and surveys

After extensive training on captive VGED scats in search areas and wild VGED scats in scent line ups, we began field training. Ideally, field training would involve a human locating a VGED scat sample (and leaving it undisturbed) and then bringing the detection dog to that location to search and immediately reinforcing the dog at the wild scat. This helps the dogs generalise their knowledge of the target and ensure that they are not becoming specific to only VGED scat that has been handled, collected and stored in certain ways. This, unfortunately, was not a training possibility, so field training focused on searching around potential VGED burrows where scat may be present, with the aim of the dogs naturally generalising. This lack of field training opportunity has likely further increased the challenge of detecting wild VGED scat. Having 2 dogs trained on this species is valuable though, as if we believe that a VGED scat has been detected we can confirm this with a second dog in situ or after collection.

Whilst Sugar is yet to detect a wild VGED scat during surveys she has been able to search very thoroughly and systematically in the field and has been successfully detecting any VGED scat that we place in the area. Moss has alerted to a single scat near an active VGED burrow. However, this sample is yet to be verified though genetically analysis.

Another potentially useful training approach to help dogs to generalise to wild VGED scats is to train them on scats of similar grassland earless dragons. Training dogs on a broad category of wildlife targets, with the aim that they would generalise to endangered or harder to source species has been successfully explored in previous research (e.g. Rutter et al. 2021; Jamieson et al. 2024). Having the dogs trained to detect scats of several grassland earless dragon species may further increase their knowledge of their target and increase the likelihood of generalisation.

Given the low rate of defecation observed in captive VGEDs and the high rate of scat scavenging/degradation (explored further in Section 4), it is therefore likely that a low density of scat is present in the environment, even when VGEDs are active. This directly increases the challenge of this project and further explains why more scats were not detected. Currently there is no survey method for VGED scat, outside of collecting scat from captured individuals, so having detection dogs capable of locating wild VGED scat remains highly beneficial. This study has highlighted that it is feasible to train detection dogs to both detect VGED scat and discriminate it from similar scats in a variety of environments. It is important to highlight the high amount of search effort required to detect these scats in certain contexts.

3.4.4 Limitations and future research

This main limitation in this study was sample size, with only 2 dogs being involved in the research. It is important to therefore highlight that if different dogs and handlers were involved in future work, and at different locations with varying habitat, training and search success may vary. Search sensitivity and search effort provided should therefore be used as a guideline, rather than something expected for every search. Regardless of the detection dog team employed, environmental and habitat factors will always be influential on search success and efficiency.

Future research would be valuable in maximising detection dogs' abilities to generalise from training samples of VGED scat (including captive VGED scat samples) to wild VGED scat samples.

At this stage, we contend that detection dog-handler team surveys may still represent a viable and valuable VGED scat survey method. However, training methods that explore ways of transitioning the dogs confidently from captive VGED or contained wild VGED scat, to wild VGED scat still require additional exploration.

At this stage, we would recommend that VGED detection dogs are trained to detect both live animals and scats, further increasing the likelihood of detection and the potential for greater data collection opportunities with only a moderate increase in training requirements.

4.0 Victorian grassland earless dragon scat longevity

4.1 Introduction

It has been identified that collecting VGED scat will be beneficial for increasing our understanding of this cryptic species. The ways in which scat can be analysed and the data that can be obtained from it is also evolving, including for reptile scat projects (Ackel, 2016). Scat detection may therefore be a valuable, non-invasive means through which important contributes to the conservation of this imperilled species can be made or informed. Whilst detection dog-handler teams have been demonstrated to be a potential detection method, there are multiple factors that will influence how feasible this method is, including the rate at which VGEDs defecate and the longevity of naturally occurring scat within the environment.

Based on our findings from the VGED detection dog project, particularly around signs of scat being scavenged quickly after being placed in certain environments, we believed it would be valuable to determine the rate at which VGED scat degrades or is scavenged. These findings will also help guide detection dog surveys and whether live VGED detection should be prioritised over scat detection.

The aims of this component of the project were to:

- Measure the physical (i.e. size and weight) variation in captive VGED scat.
- Determine the rate at which VGED scat is scavenged or degrades in a natural environment.

4.2 Methods

4.2.1 Study area

The study was completed at the VGED rediscovery site, west of Melbourne. The location of this site remains anonymous in order to ensure it remains secure. The study area was located within a section of the site that provided suitable vegetation and habitat structure for VGEDs, however, VGEDs have yet to be detected at this area. It is therefore estimated that this area provided a suitable representation of VGED habitat for the purpose of this project.

4.2.2 Scat samples

4.2.2.1 Source

Scat samples used were collected from the Melbourne Zoo VGED conservation breeding population. Scats were collected across 3 days (from 29 to 31 January 2024) to ensure enough samples were available for this study. Samples were only collected from adults and were collected using metal tweezers and stored in airtight glass jars with metal lids. The jars were labelled with the sample collection date and whether the samples were collected wet or dried.

4.2.2.2 Scat descriptions

Prior to being placed in the environment for the study, each scat was measured (length and width), weighed, and photographed to record the condition of the scat. The current physical state of the scat was also recorded, including whether the urea had already been separated from the scat or if the scat was in multiple parts. This was important in ensuring that any degradation was accurately recorded.

At the conclusion of the study, the remaining scats were weighed to measure moisture loss. These scats were then stored in glass jars until being used in the area search assessments (see Section 3).

4.2.3 Study design

4.2.3.1 Layout

Forty-four scat samples were randomly assigned to 2 conditions: 'contained' or 'exposed' (uncontained). These 2 categories allowed for the natural degradation rate of the scat samples to be determined without influence from scavenging rate by excluding the possibility of disturbance from other animals (as per Sanchez et al. 2010). Fine stainless mesh tea strainer balls were used to contain the scat, which still exposed the scats to environmental conditions while preventing scavenging (Figure 13). The exposed scats therefore provided an estimate of scat longevity when subjected to both scavenging and degradation.

Figure 13. An open mesh tea-strainer ball that contains a VGED scat

Source: La Toya Jamieson

The scat samples were stationed along a transect line with 1 m distance between each station. The stations alternated between contained and exposed scats and each station was marked by a flag. The exposed scats were placed in between 2 flags so they could be easily observed throughout the study if they were still present (Figure 14). Contained scats were pegged to the ground though a loop in the closed metal strainer to prevent it from moving in high wind.

Figure 14. An exposed VGED scat in between 2 flags

Source: La Toya Jamieson

4.2.3.2 Study duration

The study commenced on 31 January 2024, with scats being placed in the environment that evening. Scat ages at the beginning of the study therefore ranged from less than one day to 3 days old.

All remaining scats were collected after 50 days (20 March 2024), at the conclusion of the study.

4.2.3.3 Data recording

The scats were checked for the first 3 days after being stationed, and then weekly after that. Checks involved recording the status of the scat: whether it was intact (i.e. still present and intact enough to be identified), scavenged (i.e. the scat had been fully removed from the location and could not be detected within a radius of approximately 100 mm from the station) or degraded (i.e. the scat had degraded beyond the point of identification or recovery). The sample was then photographed. Environmental conditions, including temperature and rainfall were also recorded from the closest Bureau of Meteorology weather station throughout the study.

4.3 Results

4.3.1 Scat descriptions

The adult VGED scats showed a large degree of variation in size. The average scat length was 9 mm (range: 5 mm – 16 mm) and width was 3 mm (range: 1.5 mm – 5 mm).

Obtaining accurate scat weight was limited to the sensitivity of the available scales (which recorded a minimum weight of 0.03 g). Therefore, the exact weight of 5 samples could not be obtained and these were instead classed as <0.03g. The scat weights ranged from <0.03 g to 0.23 g.

4.3.2 Environmental conditions

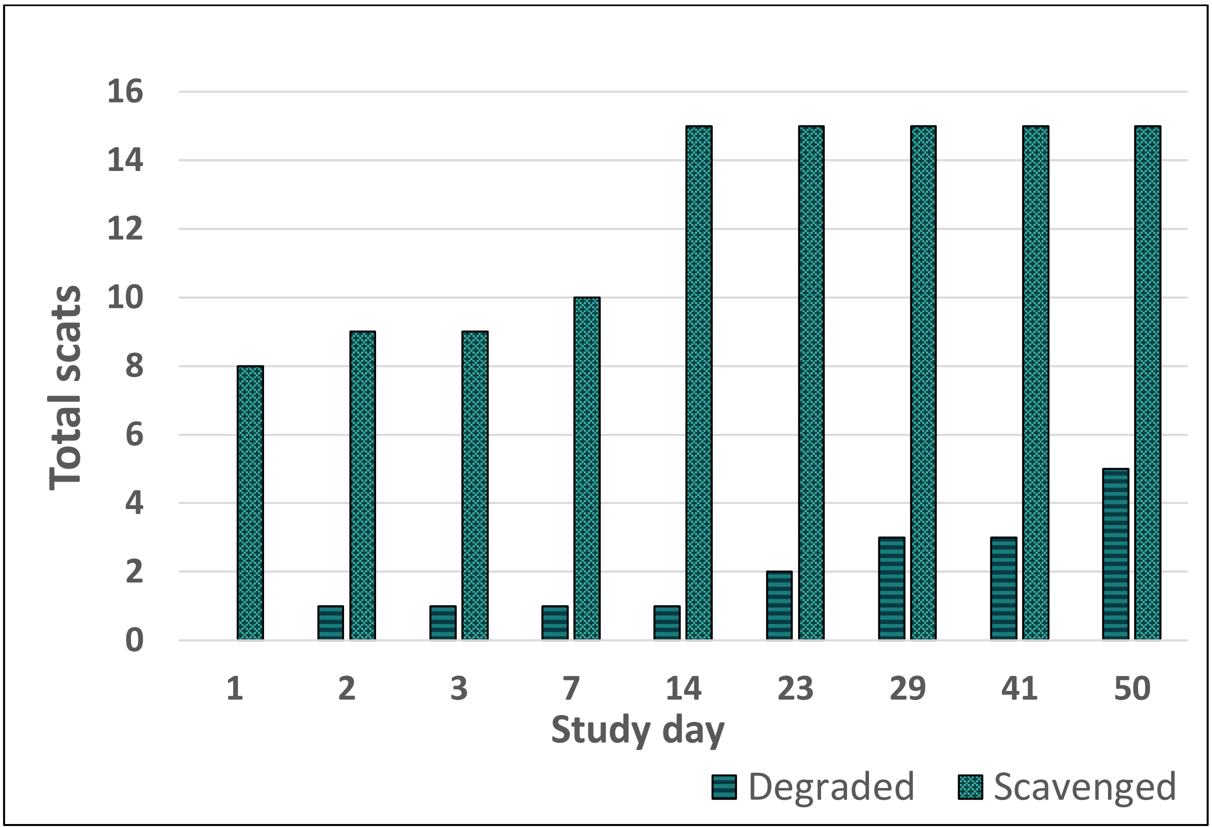
Very little rainfall was recorded over the course of the study, with a total of only 3.2 mm over the 50 days. Rain only occurred on 5 days, and the maximum within a day was 0.8mm.

The average minimum temperature was 14.1°C, and the average maximum was 27.6°C. There was a large range in temperature: from 7.3°C to 38.5°C.

4.3.3 Exposed (non-contained) scat longevity

Scavenging of the exposed scats occurred quickly, with invertebrates seen investigating the samples that had been placed out whilst the rest of the stations were still being placed. In under 24 hours, over one third (36%) of the scats had been scavenged, reaching 40.9% by day 3 (Figure 15). By day 14, 68.1% of the scats had been scavenged, which then remained stable for the remainder of the project. Comparatively, the exposed scats degraded at a slower rate, with the first scat being degraded by day 2, while the second scat did not degrade until day 23. The rate of degradation did slowly increase over the course of the study though, unlike scavenging which plateaued after day 14. Within a month, 82% of the scats were recorded as either degraded or scavenged, and by day 50 only 2 scats remained. At the project conclusion, 25% of the exposed scats had degraded and 75% were scavenged.

Figure 15. Total number of exposed scats that degraded or were scavenged within the study, ending on Day 50

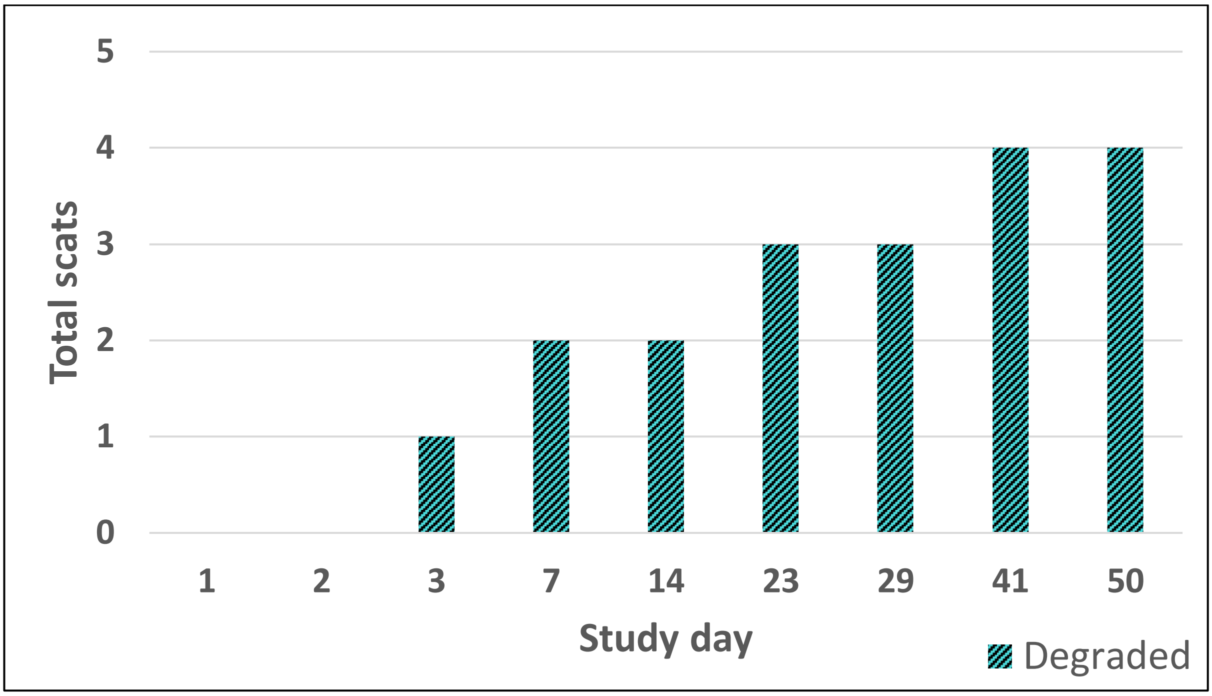


4.3.4 Contained scat longevity

The rate of the contained scat degradation was similar to that of the exposed scats (Figure 16). The first scat was observed to have degraded within 3 days. By day 7, 9% of the scats had degraded and within a month, 13% were degraded. At the conclusion of the study, 18% of the contained scats had degraded, which is similar to the level of exposed scats degradation.

The scats that were still intact at the conclusion of the study had a weight loss of 62% on average, with 54% of the scats weighing less than <0.03g.

Figure 16. Total number of contained scats within the study that degraded, ending on Day 50



4.4 Discussion

Overall, the scats in the longevity study took longer to degrade in the natural environment than expected. This may be due to the very low rainfall experienced across the 50 days, with the most rainfall received in one day being 0.8 mm. In a similar scat longevity study of Texas horned lizards (*Phrynosoma cornutum*) over summer, authors reported that scats remained intact after 90 days, however, these scats were housed in open petri dishes and were brought inside overnight or during rainfall events (Huerta et al. 2023). In another similar coyote (*Canis latrans*) and bobcat (*Lynx rufus*) scat longevity study, authors reported that the unprotected (exposed) scats had an average survival rate (including degradation and scavenging) of 11 days, reducing to 4 days over June-July (Sanchez et al. 2010). Comparatively, the VGED scats had much faster rates of degradation and scavenging than both these previous studies. It may be beneficial to repeat this study with daily 'watering' of the scats to simulate rainfall in order to determine whether this would increase the rate of degradation. This current study was limited to only one season (summer), so it would also be beneficial to explore if degradation rates change across different seasons.

Scats were scavenged very quickly overall, particularly within the first 24 hours. Scavenging only occurred within the first 14 days of the study, which was possibly influenced by the freshness of the scats. Scats aged over 14 days may therefore be less palatable to scavengers. Palatability may also be related to scat moistness, with increased rainfall possibly also increasing scavenging rates. Over one-third of the non-contained scats were scavenged in the first 24 hours, but it is unknown how far these scats were scavenged and how quickly they degraded. Repeating this study across different seasons may also modify the rate of scavenging, potentially changing invertebrate (the presumed scavengers) activity or accessibility to other food resources. Future research could also explore whether detection dogs can locate these scavenged scats or if detection is not feasible.

The variation of the scats in this study, particularly length and width characteristics, further highlighted the challenges of visually discriminating VGED scat from other local reptile scat. Having the dogs accurately discriminating between scats through olfaction (smell) is therefore essential.

Based on the relatively low defecation rate of the VGED (Melbourne Zoo specialist keepers currently estimated up to one scat per day in captive VGED during their peak activity season), the low-density nature of the target species and the high scavenging rate within the first 24 hours, it is likely that there is not a large amount of scat available or persisting within the environment. This means that while any scat located in the field is valuable, a large amount of search effort is required for it to be detected. We therefore see the most benefit in detection dog teams being trained to locate both live VGED and VGED scats, rather than scat alone. This may slightly increase handler uncertainty if the dogs were to alert and we cannot visually locate anything (i.e. they could be alerting to degraded VGED scat particles and not falsely alerting), but this can mostly be mitigated in training by only reinforcing for detecting intact scats and not separate scat particles.

5.0 Conclusion and recommendations

Following months of training, surveys, and assessments throughout this first phase of research into training and deploying detection dog-handler teams to search for the critically endangered VGED, we can confirm that both of the VGED detection dogs successfully detecting multiple individual lizards in the wild. This has been achieved in addition to a substantial amount of training, field trials and subsequent routine searches. We have developed a robust search protocol, using endoscopy to confirm VGED presence in a burrow following a dog alert. The challenges posed by both the rarity of the species and the limited scent dispersion from burrows to the surface should be acknowledged, yet seasonal changes in VGED activity levels and their behaviours (such as emerging from burrows for foraging and breeding) provide increased opportunity for detection. While we have been unable to verify that the 2 VGED scat detection dogs detected VGED scat at the rediscovery site, our study has highlighted the high degree of specificity the dogs can be trained to and the search effort required to detect these tiny, fragile scats.

The success of this research prompts our recommendation to conduct a second project phase, that would allow us to expand detection dog-handler team surveys to new sites across the Victorian Volcanic Plains west of Melbourne that host potentially suitable habitat for VGED. We also see high value in extending the scope of this work to include other closely related and endangered species of grassland earless dragons within the genus *Tympanocryptis.* This would involve training Zoos Victoria's detection dogs to detect the odour of live individuals of *T. lineata* (the Canberra grassland earless dragon ('Canberra GED')), firstly in a controlled setting with odour samples and live animals sourced from a captive population, then graduating to surveys in suitable habitat across several sites within the species historic range in the ACT and or NSW. The timing of this work is critical considering the perilous situation the Canberra GED program finds itself in with very few individuals detected this past field season, and the urgency to detect and bring additional individuals into the conservation breeding program at Canberra University and Tidbinbilla Nature Reserve.

While little is known about any differences in odour profile that may exist across grassland earless dragon species, and we would to some extent be taking a proof-of-concept approach to this component of the project, we expect there may be sufficient similarity to allow the detection dogs to generalise their scent training from the Victorian and Canberra GEDs to *T. osbornei* (Monaro grassland earless dragon) and potentially *T. mccartneyi* (Bathurst grassland earless dragon). The detection dog method could then be implemented in surveys for any of the 4 *Tympanocryptis* species of grassland earless dragon.

Key priorities for Phase 2 research include:

- Expanded searches for the VGED at new sites west of Melbourne.
- Testing the ability of the VGED detection dogs to 'generalise' their scent training across to the other 3 GED species: *T. lineata* (Canberra GED), *T. osbornei* (Monaro GED) and *T. mccartneyi* (Bathurst GED), as well as undertaking direct training on these species with the cooperation of colleagues in NSW and the ACT who have expressed a strong interest in collaboration.

- Incorporating radio telemetry and eDNA analysis as complementary technologies to detection dog training, assisting in guided searches when dogs are being trained, and collection of further data on burrow occupancy.
- Begin training 2 additional detection dogs for live dragon detection (increasing our capacity of operational detection dogs).
- Potential PhD project with a focus on the use of dogs for detection of *Tympanocryptis* species.
- Publish scat detection and scat longevity study in 2025 and continue collecting data for live animal detection study for publication in 2025/26.

Figure 17. A Victorian grassland earless dragon

Source: Peter Robertson

Appendix A

Table A2. In situ training opportunities available for dogs to detect live VGEDs sheltering in naturally occurring invertebrate burrows

Date	No.	Photograph	Date	No.	Photograph
1/02/2024	1		1/02/2024	2	
14/02/2024	3		12/03/2024	4	
12/03/2024	5		12/03/2024	6	
12/03/2024	7		13/03/2024	8	
13/03/2024	9		13/03/2024	10	

Date	No.	Photograph	Date	No.	Photograph
19/03/2024	11		19/03/2024	12	
19/03/2024	13		19/03/2024	14	
20/03/2024	15		26/03/2024	16	
26/03/2024	17		8/04/2024	18	
8/04/2024	19		8/04/2024	20	

Date	No.	Photograph	Date	No.	Photograph
8/04/2024	21				

Glossary

Term	Definition
Alert / Indication	A trained behaviour detection dogs perform to indicate to their handler that they have detected a target, e.g. a VGED
eDNA	Environmental DNA
VGED	Victorian grassland earless dragon (*Tympanocryptis pinguecula*)
WDD	Wildlife detection dog

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