



EchidnaCSI: Engaging the public in research and conservation of the short-beaked echidna

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The short-beaked echidna is an iconic Australian animal and the most-widespread native mammal, inhabiting diverse environments. The cryptic nature of echidnas has limited research into their ecology in most areas; however, from the well-researched and endangered Kangaroo Island echidna population, we understand that the threats include habitat loss, roads, and invasive species. To obtain more information about echidnas Australia-wide, we established the Echidna Conservation Science Initiative (EchidnaCSI) citizen science project. EchidnaCSI calls on members of the public to submit photographs of wild echidnas and learn to identify and collect echidna scats for molecular analysis. To facilitate participation, we developed a smartphone application as well as ongoing social and traditional media activities and community events. In 3 y, more than 9,000 members of the public have downloaded the EchidnaCSI app, collecting 400 scats and submitting over 8,000 sightings of echidnas from across Australia. A subset of submitted scat samples were subjected to DNA extraction and PCR, which validated the approach of using citizen science for scat collection and viability for molecular analysis. To assess the impact of the project through public participation, we surveyed our participants ($n = 944$) to understand their demographics and motivations for engagement. Survey results also revealed that EchidnaCSI served as a gateway into citizen science more generally for many participants. EchidnaCSI demonstrates the potential for using citizen science approaches to collect high-quality data and material from a cryptic species over a very large geographic area and the considerable engagement value of citizen science research.

citizen science | monotreme | Australia | scat | DNA

The short-beaked echidna (*Tachyglossus aculeatus*) is one of Australia's most-iconic mammals and is of both evolutionary and ecological importance. Echidnas and platypuses form the unique group of egg-laying mammals (monotremes), which is the most-ancient surviving mammalian lineage, diverging from all other mammals ~187 million y ago (1, 2). Both monotremes are seasonal breeders with breeding season around June through September each year. During this time, echidna trains are observed where several males follow one female (Fig. 1A) (3). The short-beaked echidna (hereby referred to as “echidna”) is characterized into five subspecies found in Australia and parts of New Guinea and is the most-widespread native Australian mammal, inhabiting environments from snow to desert and tropical regions (Fig. 1B–D) (4, 5). Because of their cryptic nature and wide distribution, we lack basic population information, making echidnas difficult to study in the wild (6, 7). The subspecies *Tachyglossus aculeatus multiaculeatus*, which inhabits Kangaroo Island (a large island off the coast of South Australia), is the only extensively researched and monitored echidna population, which has resulted in this subspecies' recent listing as “endangered” under the Environment Protection and Biodiversity Conservation Act 2015 (“Conservation Advice *Tachyglossus aculeatus multiaculeatus* Kangaroo Island echidna”). However, the IUCN

(International Union for Conservation of Nature) Red List database lists the short-beaked echidna as a species of “least concern” due to the lack of knowledge on basic ecology in most parts of Australia [<https://www.iucnredlist.org/> (8)]. The greatest threats to echidnas identified from the Kangaroo Island population, like for many Australian mammals, are feral cats, vehicle strike, and habitat loss (9). These threats exist on mainland Australia (along with additional predators such as foxes, pigs, and dingoes) and have been further exacerbated by the recent devastating Australian bushfires in 2019 and 2020 (10). It is therefore a matter of urgency to obtain more information to determine the conservation status of echidnas across Australia. As far as we are aware, there are no concerted efforts in place to ascertain and monitor echidna populations. As echidnas are difficult to study in the wild, gaining Australia-wide information on their populations is a very challenging task. Echidna sightings have been reported in the past via paper-based reports to citizen science “Echidna Watch” projects hosted by Wildlife Queensland

Significance

Echidnas are iconic egg-laying mammals threatened by environmental changes. However, their cryptic lifestyle and Australia-wide distribution renders a citizen science approach the only feasible way to obtain continent-scale information. With EchidnaCSI (Conservation Science Initiative), we used a citizen science approach to learn more about echidnas, which is remarkably powerful and continually provides data and material for molecular and ecological research. Here, we describe and discuss the establishment and performance of EchidnaCSI, which has produced high-quality echidna-sighting data and fecal material. In addition, a survey of participants provides insight into the motivation and engagement outcomes in citizen science and conservation. EchidnaCSI demonstrates the enormous potential of citizen science to advance the data and knowledge base to enable echidna conservation outcomes.

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The authors declare no competing interest.

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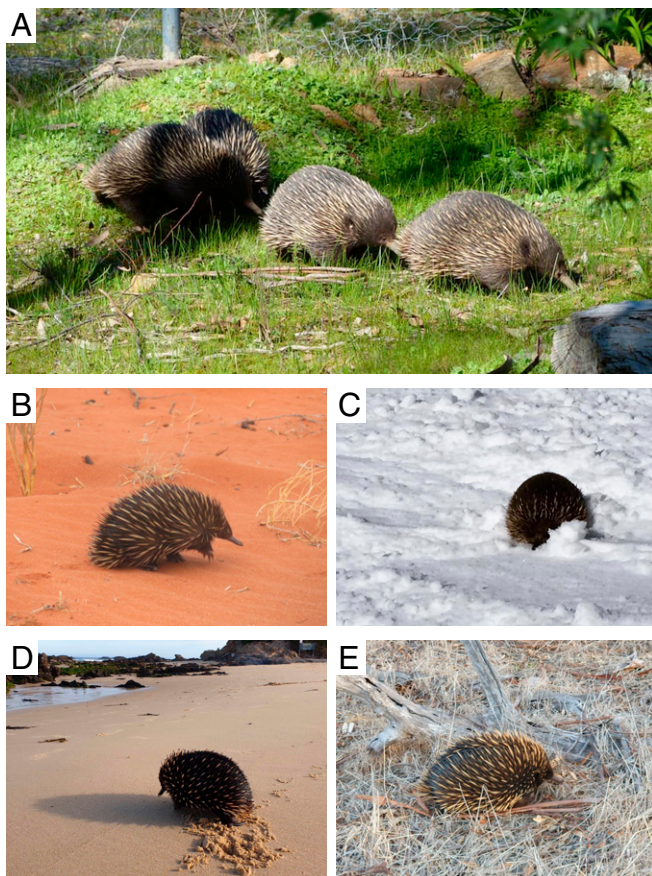


Fig. 1. Subset of echidna photos submitted to EchidnaCSI by the public. (A) An echidna train formed during breeding season (June through September), in which a female is followed by multiple males. (B–E) Echidnas seen in all types of environments including desert (B), snowy alpine (C), coastline (D), and bushland (E).

[<https://wildlife.org.au/echidnawatch/?v=13b249c5dfa9> (11)] and by leading echidna ecologist Dr Peggy Rismiller, which shows promise for using these types of approaches for nationwide and more coordinated data collection long term.

The citizen science approach is increasingly recognized as producing valuable and large datasets in environmental biology as well as many other fields (12). Citizen science is an excellent platform for research, as it has the potential to enlist thousands of members of the general public to collect data over large geographic and time scales, which is not usually possible for researchers without significant time and costs associated (13). In addition, advances in technology and social media have vastly increased the reach and reliability of citizen science research, at least for those with access to online technology (14, 15). The smartphone has revolutionized this approach, and many projects use apps, providing validation and additional information (photo, date, time, and location) as well as limiting user error in data submission (16–18). Apps can also be linked directly with larger databases (in Australia the Atlas of Living Australia [ALA]: <https://www.ala.org.au/>), allowing accessibility to the general and scientific community. Social media have also been powerful in advancing citizen science research, facilitating engagement of new and existing audiences over large geographic scales (such as entire countries) (19, 20). Importantly, citizen science is an effective engagement and education platform that can be used to increase the public's knowledge of science and raise awareness for environmental issues and conservation (21, 22).

A critical part of designing a successful citizen science project is how to recruit participation and sustain engagement over

time. In order to ensure good communication and outreach for targeted audiences, many citizen science projects have evaluated the demographics and motivations of their participants through surveys to gain a deeper understanding of why they volunteer their time for certain scientific tasks (23–25). By evaluating how project participants rank these motivations, project leaders can better implement targeted strategies to increase engagement and diversity in participation (23). It is important to engage diverse audiences in citizen science, especially for biodiversity and conservation-type projects, as direct involvement can empower individuals to make significant changes in their attitudes and behaviors around environmental and sustainability issues, including sociopolitical views (26–28). Citizen science is effective at engaging people in many forms of science; therefore, it may be a powerful avenue for aiding science learning in often underrepresented groups such as rural, regional, and indigenous communities (29). Furthermore, species of conservation interest often live in nonurban environments, such as the echidna, and so engaging with these communities is crucial for achieving appropriate research outcomes.

Many citizen science projects focus on data collection of plants and animals for biodiversity and conservation purposes (30–34). These studies have led to important ecological milestones such as gaining baseline population information (35), finding pockets of habitat (36), and showing distribution changes (37) or declines in species numbers (31), ultimately leading to tangible conservation outcomes (38). While many citizen science projects are created to assist with producing research outcomes, others are formed to enable directions and perform science that could not be done through the traditional research route. An exciting avenue of citizen science is incorporating material collection for analyses (such as genomic or microbiome studies), which greatly increases the information that can be obtained from the species of interest (39–42). Animal scats (feces) contain DNA and hormones that can provide information about sex, population genetics, diet, stress level, microbiome gut health, and reproductive activity of the species of interest (43–47). Analyzing animal scats has become increasingly used in field studies over the past 20 y due to improved technologies and more-robust techniques, which allow valuable information about an animal to be gained in a noninvasive way (48). However, fecal material collection from animals is rarely used in citizen science projects and thus could provide a powerful avenue for wildlife research.

With the Echidna Conservation Science Initiative (EchidnaCSI), we established an innovative citizen science project that incorporates both echidna-sighting submission and scat collection, through a dedicated phone app, with the aim to record echidna sightings and obtain scat samples Australia-wide. After 2 y, we surveyed participants of the project to determine the demographics and motivations behind their participation. Here, we provide an overview of EchidnaCSI design, roll out, and performance. The main aims of the project were to 1) generate the largest database for echidna sightings in order to develop a baseline distribution map to track population changes in the future, 2) collect echidna scats from across Australia and validate their use for future molecular work in order to show the feasibility of incorporating scat collection in large-scale citizen science efforts, 3) engage the public in scientific research to raise awareness of the importance of echidna conservation, and 4) determine the demographics and motivations of participants in order to evaluate the project and develop strategies to increase future public engagement and participation.

Results

EchidnaCSI Participation and Engagement. From the launch of EchidnaCSI on 4 September 2017 until 4 September 2020, EchidnaCSI had 9,079 downloads of the dedicated app, resulting in the submission of 8,090 echidna sightings and 406 echidna

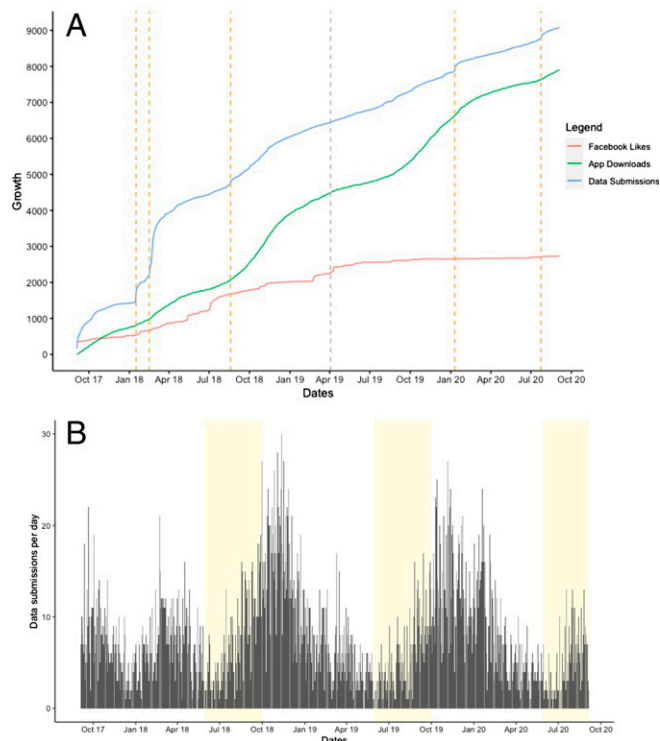


Fig. 2. Change over time for app downloads, social media reach, and data submissions. (A) Accumulative growth of number of EchidnaCSI app downloads, submissions of data (either echidna sightings or scats), and Likes on the EchidnaCSI Facebook page. Orange dotted lines indicate dates of large increases in app downloads associated with media and events (SI Appendix, Table S1). Gray dotted line indicates launch of EchidnaCSI Twitter and Instagram accounts. (B) Number of data submissions per day; submissions can fluctuate between 0 and 30 submissions per day in a cyclic trend. Echidna breeding season is indicated in yellow shading. Data are visualized from 4 September 2017 until 4 September 2020.

scats. A total of 2,816 users had submitted data (either photo sighting or scat collection); most participants (56.8%) had submitted data once, 33.2% had submitted data between two and five times, and 10% had submitted data more than five times. Although app downloads, Facebook Likes, and data submissions continually increased, large increases in app downloads were mostly associated with nationwide media broadcasts, in particular news articles (SI Appendix, Table S1 and Fig. 2A), while increases in Facebook Likes were associated with viral social media posts. Data submissions varied from zero to 30 submissions per day, with a cyclic trend (Fig. 2B). Although echidnas are most active during breeding season (June through September), these were not the months with the highest data submissions. Instead, September to February were the most-active months (Australian spring and summer/holiday season).

The main forms of communication with participants were through bimonthly email newsletters and the Facebook page (Twitter and Instagram accounts were also launched on 3 April 2019). Since the launch of EchidnaCSI until 4 September 2020, the Facebook page grew to 2,734 Likes (Fig. 1A), consistently engaging growing audiences. Responses from the survey indicated that Facebook was the most-common mode for participants to be introduced to EchidnaCSI, followed by “word of mouth.” Facebook posts have a high engagement rate (maximum: 68%, minimum: 3%) with the base rate considered as “good” engagement by marketing standards being 1% (49); one EchidnaCSI post “reached” over 100,000 people, according to Facebook metrics.

Echidna Sightings. In total, 8,090 echidna sightings were received from across Australia (Fig. 3A), with data submitted from every state and territory. Many submitted sightings are from densely populated areas, city fringes, and even within major cities (Fig. 3A). Users are asked to submit sightings of both live and deceased echidnas; 314 sightings (4% of total sightings) were recorded as deceased, and of those, 82% were due to vehicle strike (Fig. 3B). Users were asked to self-report the kind of environment in which the echidnas were sighted: 35% were in native vegetation, 26% roadside, 23% agriculture or farmland, 11% urban, and 3% coastal (Fig. 3C). Although size of echidnas does not generally correlate with age or maturity, if echidnas were described as able to “fit in one hand,” then these sightings were attributed to juvenile echidnas; only 2% of total sightings were considered as juvenile. Mating trains were seen frequently during breeding season; however, the actual mating was rarely observed (1% of total sightings).

Echidna Scats. Collection of echidna scats has never been attempted before and only few citizen science projects include material collection. Through EchidnaCSI, 406 echidna scats were collected from across Australia, providing the largest collection of echidna material to date. Citizen scientists collected invaluable samples from remote locations such as the Kimberley in Western Australia, APY (Anangu Pitjantjatjara Yankunytjatjara)-lands in central Australia, Arid Recovery in South Australia, and far north and central

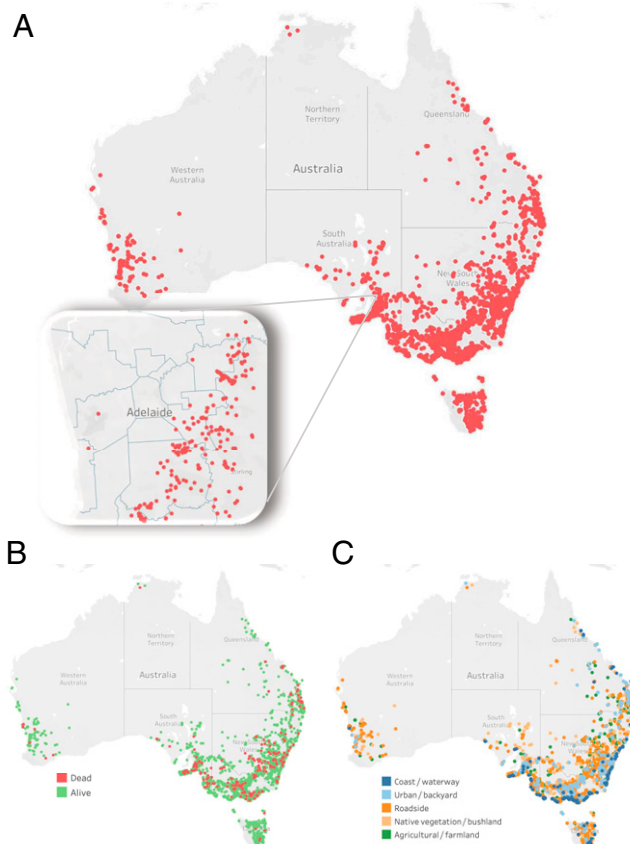


Fig. 3. Echidna sightings submitted to EchidnaCSI. (A) All sightings submitted between 4 September 2017 and 4 September 2020 are shown in red across Australia, with Adelaide highlighted as one of the major cities where echidna sightings were submitted immediately surrounding the city. (B) Sightings are colored according to whether the echidna was alive (green) or dead (red). (C) Sightings are colored according to the type of environment in which the echidna were seen in.

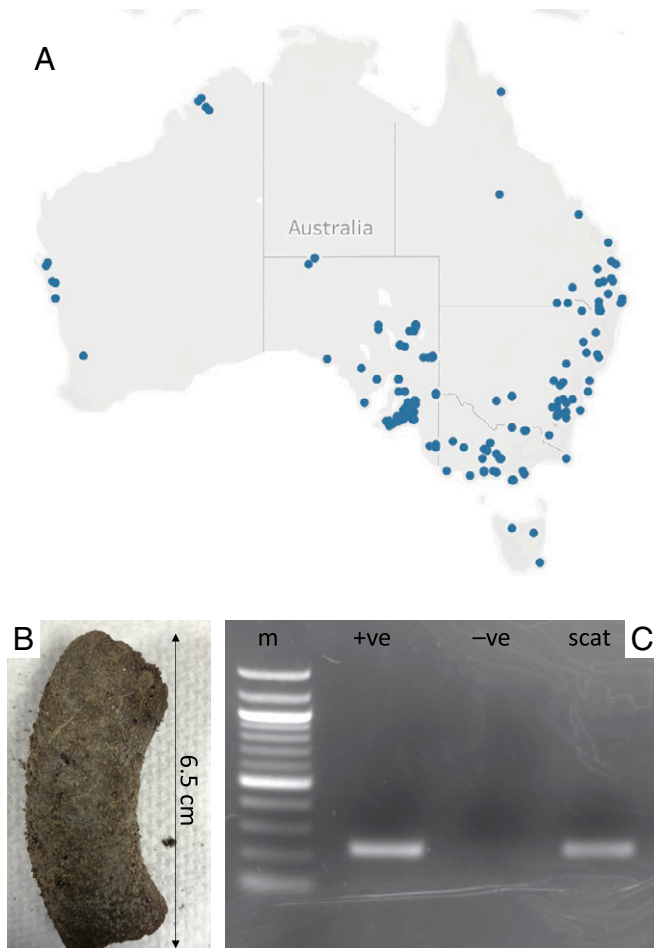


Fig. 4. Echidna scats collected by the public and validation of their use in molecular biology. (A) Locations across Australia where echidna scats were collected by the public between 4 September 2017 and 4 September 2020. (B) Photograph of an echidna scat, showing the distinct long, cylindrical shape with blunt ends and dry soil texture; color depends on the soil that the echidna was feeding in. (C) PCR of the mitochondrial D-loop region specific to echidnas (200 bp); m = 100 bp marker; +ve = positive control; -ve = negative control; and scat = DNA from echidna scat.

Queensland as well as along the east coast of Australia and throughout many regions in South Australia (Fig. 4A). Submitted samples were verified as echidna scats due to their distinct physical appearance (Fig. 4B). In order to investigate the feasibility of using these echidna scats for molecular analyses, a subset of scats ($n = 232$) underwent DNA extraction and PCR to amplify a 200-bp genomic region specific to echidnas (50) (Fig. 4C). While the low annealing temperature may lead to some unspecific binding, both approaches (morphology and PCR-positive of $n = 171$) validate the feasibility of correct scat collection through citizen science and their use for further molecular analysis.

Participant Demographics. A survey was emailed on 22 August 2019 to 5,720 participants. Survey responses were received within 3 wk from 944 participants who were engaging with EchidnaCSI. Responses were received from across Australia, clustering in major cities, and also internationally (Fig. 5A). In total, 64% of survey respondents had submitted data (either echidna sightings or scats), while 36% had not but were still engaging with the project by downloading the EchidnaCSI app, following and/or sharing posts on social media, participating in

presentations or workshops, or by having conversations with the EchidnaCSI team (via email, phone or in person). These latter participants expressed that they had not submitted data due to not seeing echidnas or scats since downloading the app (or not being able to capture a photo of an echidna) and not because they were no longer interested with the project. More females (62.5%) than males (36.6%) participate in EchidnaCSI ($P = 3.5 \times 10^{-14}$), with less than 1% preferring not to state their gender (Fig. 5B and *SI Appendix, Table S2*). EchidnaCSI participants were spread across all age groups from 18 y and older, with ~50% between 18 and 54 and ~50% aged 55 and older (Fig. 5C). In comparison to the Australian census population, the distribution of EchidnaCSI participants for all age groups were significantly different; age groups 25 to 34, 35 to 44, 75 to 84, and 85+ were underrepresented in the EchidnaCSI cohort, while age groups 45 to 54 and 55 to 64 were overrepresented ($P < 0.05$; *SI Appendix, Table S2*). In terms of education, while 55% had a Bachelor's degree or higher (Fig. 5D), 60% of participants had a maximum of year 11 or 12 high school education in science (Fig. 5E). The largest proportion of participants were fully employed (29%), followed by retirees (25%) and part-time employment (15%); students (3.5%) and unemployed individuals (2.5%) were the smallest categories. Retirees were overrepresented and part-time employment underrepresented in the EchidnaCSI participant cohort in comparison to Australian census data ($P < 0.05$; *SI Appendix, Table S2*). When asked with whom they submit data, 41% of participants contribute data on their own, 29% with a partner, 12% with children, 8% with a friend, 3% with a colleague, 2% with grandchildren, and 2% with a parent. In terms of ethnicity data collected, 82.4% self-reported as Australian, with a further 10.5% considering themselves Australian in addition to another ethnicity (e.g., Australian and European; Australian and Asian). In total, 0.8% identified as either only "Indigenous Australian or Torres Strait Islander" or "Indigenous Australian or Torres Strait Islander" in addition to another ethnicity. The remaining 5.3% identified as European, New Zealander, Asian, Indian, North American, or South American with 0.8% preferring not to answer (*SI Appendix, Table S2*). There is not sufficient census data to directly compare these results; however, the 2016 Australian census does report that 66.7% of the population list their country of birth as Australia, followed by England (3.9%), New Zealand (2.2%), China (2.2%), and India (1.9%), and 2.8% of the population identify as Indigenous Australian or Torres Strait Islander [https://quickstats.censusdata.abs.gov.au/census_services/getproduct/census/2016/quickstat/036; (51)].

Introducing Participants to Citizen Science. Survey results show that for most participants (63%), EchidnaCSI is the first citizen science project they had participated in. When comparing those who had submitted data to EchidnaCSI (submitters) to those who had not submitted any data but still engaged with the project (nonsubmitters), there was no difference between the two groups in terms of how many were actively involved in other citizen science projects (37% for both; $P = 0.95$). However, there was a larger proportion of submitters who indicated that they were more likely to be involved in citizen science in the future due to their participation in EchidnaCSI in comparison to nonsubmitters (66% and 53% respectively; $P = 4.0 \times 10^{-4}$) and a higher percentage of submitters (22%) than nonsubmitters (13%) who had joined other citizen science projects after participating in EchidnaCSI, although this was not statistically significant ($P = 0.12$). In total, 92% of submitters agree with the statement that "citizen science is worth my time," with 50% saying their views increased toward that statement since their involvement in EchidnaCSI. As for nonsubmitters, 91% agreed with the statement; however, only 32% had increased the strength of their view that citizen science was worth their time;

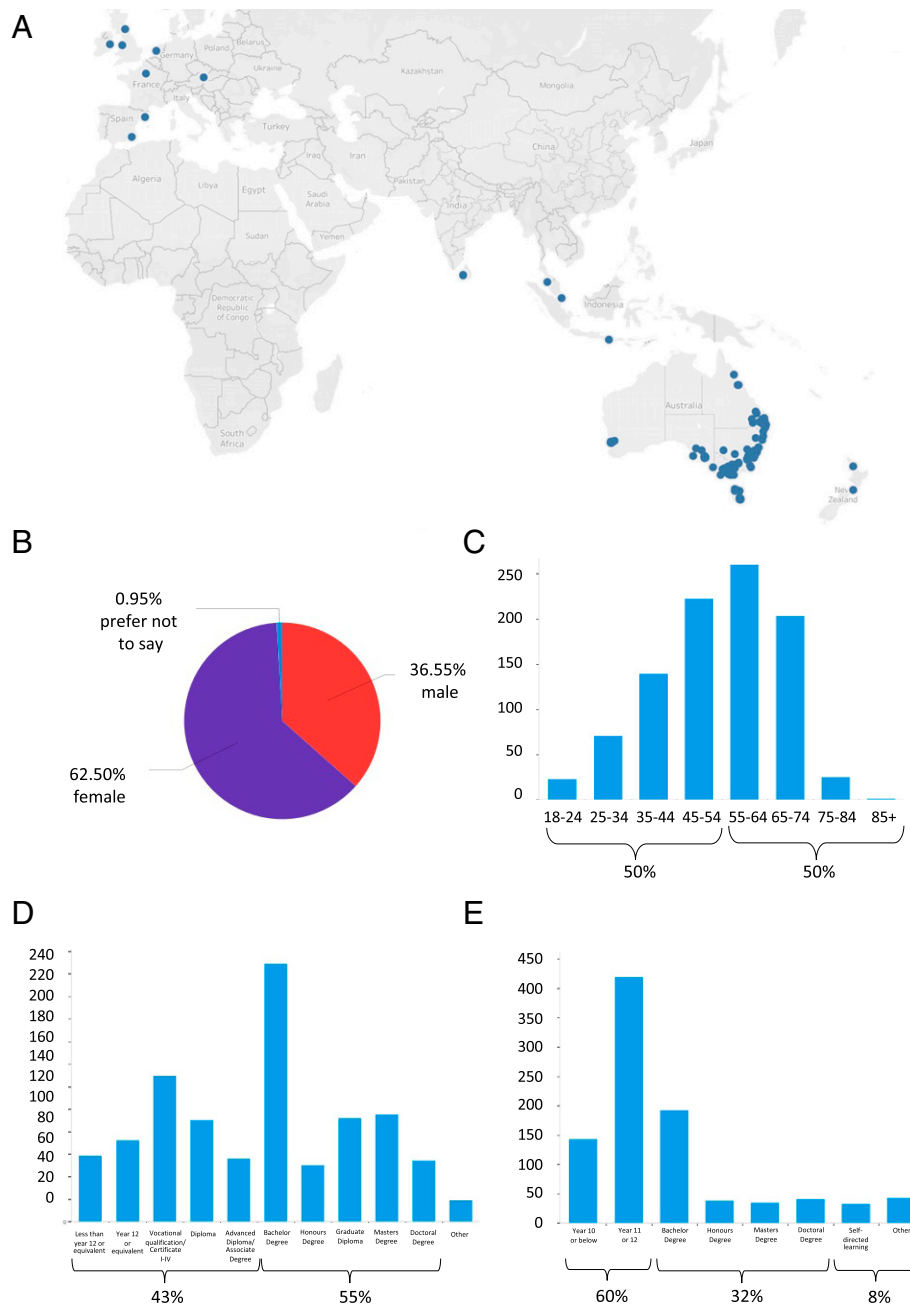


Fig. 5. Survey demographic information of EchidnaCSI participants. (A) Locations from which participants took the survey shown in blue. (B) Pie chart of gender. (C) Histogram of ages, showing the percentages of participants that were under the age of 55 and those that were 55 y and older. (D) Histogram of level of education, showing percentages of those that had an education below a Bachelor’s degree and those that had at least a Bachelor’s degree or higher. (E) Histogram of level of science education, showing percentages of those that had up to a high school–level (max year 12) science education and those that had a Bachelor’s degree or above in Science.

this change in attitude was found to be significantly different between submitters and nonsubmitters ($P = 6.1 \times 10^{-6}$) (SI Appendix, Tables S3 and S4).

Changes in Attitude and Motivation for Involvement of EchidnaCSI Participants. Next, we wanted to determine participants’ attitudes towards certain statements followed by the question about how their views had changed since engaging with EchidnaCSI. Survey results indicate that EchidnaCSI attracts participants who are passionate about echidna conservation and environmental health in general, as more than 90% agreed to the importance of these statements (SI Appendix, Table S3). However, since participating

in EchidnaCSI, a large proportion (42% of submitters and 36% of nonsubmitters) indicated that echidna conservation had become more important to them, and that their views and actions toward the health of the environment had also increased (SI Appendix, Table S4). There were no statistically significant differences between submitters and nonsubmitters ($P > 0.05$; SI Appendix, Tables S3 and S4).

The survey identified that the motivations most-greatly influencing participants were a combination of “wanting to contribute to wildlife conservation,” “liking echidnas,” “contributing to scientific research,” and “learning about echidnas.” When comparing submitters to nonsubmitters, these

four motivations were present in their top five responses at varying levels (*SI Appendix, Tables S3 and S4*). However, for the nonsubmitters, “I intend to submit data in the future” was a high motivation to continue to engage with the project, while for submitters, “the project is easy to participate in” was ranked highly. For the motivators of “wildlife conservation” and “contributing to science,” there were significant differences between the two groups, with submitters ranking these motivators as more important than nonsubmitters ($P < 0.05$; *SI Appendix, Tables S5 and S6*). The motivations that were ranked consistently the lowest included “interest in molecular biology,” “seeing recognition of my or other participants’ contributions,” and “enjoy the time spent with family and/or friends” (*SI Appendix, Tables S5 and S6*).

Discussion

EchidnaCSI Enabled High-Quality Data and Material Collection Never Before Achieved by the Scientific Community Alone. EchidnaCSI was able to achieve its main goal: to produce a large number of echidna sightings across Australia. In 3 y, EchidnaCSI has produced the equivalent of 25% of all echidna sightings in ALA, which covers more than the past 100 y of data [ALA website species page: <https://bie.ala.org.au/species/urn:lsid:biodiversity.org.au:afd.taxon:0d4c9c0c-51d3-44e0-a365-fe0f8b791c66> (52)]. Submissions require photo evidence of the echidna sighting, which ensures high-quality data additions to the ALA’s biodiversity database. Thus, this project makes a significant (and continuing) contribution that enables better assessment and understanding of echidna populations in Australia. Without a citizen science approach, which engaged thousands of members of the public, this coverage could not have been achieved. These sightings provide a considerable increase in available baseline information about echidna presence, which can now be used to monitor changes in wild echidna populations. For example, this data will be powerful in assessing the effects of the devastating 2019/2020 bushfire season in Australia (10), during which echidna distributions overlap with regions that were significantly affected, including Kangaroo Island where echidnas are already recognized as endangered. Due to the nature of the project, we receive far more echidna sightings in areas where there is higher human population density (mostly around the coastal areas of Australia), which is typical for citizen science studies (32, 53). This highlights the need for more targeted engagement strategies in rural and remote areas. Although we expected most sightings to occur close to human populated areas, we did not anticipate so many echidna sightings within or immediately surrounding all major cities in Australia. This evidence raises a number of concerns, as there is very little appropriate habitat or food sources available for echidnas in these environments and proximity to densely populated areas increases their risk of being hit by vehicles. Unlike common ring-tailed possums or koalas and in some cases even platypuses, echidnas have not previously been considered an “urban” native species, but our findings indicate that there is a need to consider echidnas when establishing policies surrounding biodiversity in cities (54, 55).

EchidnaCSI has also made a pioneering methodological contribution by successfully incorporating wildlife scat collection into a nationwide citizen science project, a strategy that has not previously been utilized at a large scale to our knowledge. This approach has resulted in a significant material collection for echidnas from geographically unique locations that would have been impossible to obtain in any other way than through community participation. Unlike other material collection projects (40, 42), the scats were collected by participants without specific training or kits. Instead, a combination of resources was provided (e.g., in-app instructions, scat identification guide in-app and on website) which, together with the distinct appearance of

echidna scats, were sufficient for the general public to successfully identify, collect, and ship scat samples. The validation of scats for their use in molecular work (as PCR-positive) opens exciting avenues for understanding more about wild echidna biology such as diet, gut health, reproductive success, and potential stressors through genomic and hormone analyses. This approach provides a model for how to incorporate scat collection and analysis for other animals into citizen science projects.

EchidnaCSI Used Effective Continent-Wide Recruitment and Engagement Strategies. EchidnaCSI represents an innovative citizen science project that has used traditional and modern approaches to obtain sightings and scat samples from the iconic echidna nationwide. The use of traditional media (e.g., radio, television, and news articles) early in recruitment was effective in reaching large audiences, which has been observed in other Australia-wide projects, for example on wombats (56). Social media became an important form of recruitment later in the project when a cohort of participants were already registered, as the majority of users first heard about the project via Facebook. In-person events were also an effective form of recruitment as seen by an increase in app downloads during National Science Week in 2018 when we held or spoke at six events over 7 d, reaching over 400 participants. Our approach of using social media as the main platform of communication also meant that we were able to engage a significant number of people who had not directly contributed data or material to EchidnaCSI (36%). We found that this cohort had similar increases in concern for echidna conservation and environmental issues after engaging with EchidnaCSI to those who had submitted data, suggesting that simply engaging with EchidnaCSI was sufficient to change attitudes and behaviors without the need to formally participate and submit echidna sightings or scat material to the project. We did, however, find that participants who submitted data or material were more likely to join other citizen science projects, likely due to having had a positive experience with EchidnaCSI. We found that reaching broad audiences through a combination of traditional and social media introduced a large cohort of the public not just to echidna conservation but to citizen science generally.

Diversity Achieved in EchidnaCSI Participant Demographics. Concerns have been raised that a lack of demographic diversity exists in many citizen science projects, as most volunteers tend to be highly educated, white adults who are 50 y or older and are often retired (57–59). Although biodiversity-type projects have no overarching gender bias (60), our survey revealed that EchidnaCSI has more female participants than male, which has only been documented in one other citizen science project that also had a conservation focus (23). Parrish et al. identified that projects focusing on membership may encourage female participants, while competitiveness would lead to more male participation (61). Although the age range in EchidnaCSI participants is diverse, those between the ages of 45 and 64 were still overrepresented in comparison to the Australian population, which has been reported for other nature-based projects (62). Due to the survey being limited to those over 18 y of age, we could not accurately gauge our engagement with younger audiences. However, as we have presented at events that were specifically aimed at primary and high school children and 11% of the survey respondents reported submitting data with their children, we expect the actual age demographics of our EchidnaCSI community to be younger than what we have been able to capture. The survey also highlighted that EchidnaCSI caters to those who are both “time poor” and “time rich,” as the largest cohort of participants are fully employed and the second largest are retirees. This is likely a key factor in our ability to have

more variety in the diversity of participants, along with the many strategies of recruitment and engagement (e.g., traditional media, social media, and in-person events). However, we would like to further improve the diversity of EchidnaCSI participants, especially for varying ethnicities (particularly indigenous and remote communities) and those without university qualifications, as well as continue to reach younger audiences. Pandya in 2012 (58) wrote a framework to engage more diverse audiences in citizen science, which includes strategies such as meeting with community members to discuss scientific questions that will mutually support the research and community goals as well as incorporating multiple sources of expertise including traditional knowledge and historical accounts in addition to rigorous scientific data. These will be particularly important when considering engaging with indigenous communities. It is likely that a project such as EchidnaCSI could resonate with indigenous Australians, as echidnas have long been a subject of cultural stories and a biological indicator for many indigenous communities as well as a food and medicine source (63). Furthermore, developing data collection and engagement strategies that do not rely on online submission and communication will also likely increase involvement from currently underrepresented groups.

Using EchidnaCSI Participants' Motivations for Future Recruitment. Details of the core motivations of current participants can be used as a powerful tool to increase recruitment and engagement. Our survey revealed, similar to other citizen science studies, that the main motivations for the participants of EchidnaCSI were 1) wildlife conservation, 2) interest in echidnas, 3) contributing to science, and 4) learning. Interestingly, unlike findings in many other conservation and biodiversity-type citizen science projects (64–66), spending time with family or friends was ranked among the lowest motivators. This may be due to the fact that echidna sightings mostly occur from opportunistic circumstances rather than planned activities due to the cryptic nature of echidnas. In other projects, participants are motivated by wanting to be recognized for their contributions (66, 67); however, for EchidnaCSI participants, recognition was a low-ranked motivator. This illustrates the importance of a survey capturing the motivation and expectations of participants to inform engagement, retention, and education approaches, which is also emphasized by Dibner and Pandya (60). Although our marketing had already focused on echidnas and their conservation (the highest motivators), incorporating more about the contributions that this research is making and opportunities to learn from participation will be important for future communications and engagement strategies, based on evidence from our survey. Parrish et al. provide an in-depth analysis of project design in relation to data quality, participant profile, and retention, which supports the experience with EchidnaCSI that a well-designed project with a dedicated participant base is very capable of generating high-quality data. While we did not quantify retention, we do know that a number of participants have engaged continuously with our project, suggesting high retention and long-term commitment to the project.

Conclusion and Future Directions. EchidnaCSI delivers a continuously growing baseline dataset for wild echidna populations Australia-wide, which could not be generated without a citizen science approach. EchidnaCSI contributes to research and public awareness, which are essential for long-term conservation of echidnas in our changing environment. Incorporation of nationwide scat collection through citizen science worked remarkably well and provides a massive value-add to the scientific outcomes of the project. In our experience, a combination of ongoing traditional and social media is key for engaging and retaining a large audience over a continental scale, although there is a need to specifically target rural, regional, and remote areas across Australia to avoid bias toward urban areas. EchidnaCSI attracted a

markedly different demographic compared to other projects, which is important to inform project-specific engagement and education strategies. In future, the sighting data from EchidnaCSI will be used to evaluate the distribution of echidnas across Australia including habitat preferences and will allow long-term population monitoring; scats will be used for molecular studies to investigate diet and gut microbiome changes; and targeted strategies will be used to engage with rural, regional, and indigenous communities across Australia where uptake has been comparably low.

Materials and Methods

Data and Sample Collection. EchidnaCSI collects data via a smartphone app using both iOS (Apple, Cupertino, CA) and Android (Google, Mountain View, CA) operating systems (*SI Appendix, Fig. S1*). Three main functions exist within the app. First, users can submit a live photo of an echidna, through which the app collects the date, time, and GPS (global positioning system) location of the photo. Second, users can submit photos of echidnas that they have previously taken on their smartphones, so long as the photo has the date, time, and location data embedded within the photo. Following the taking or selection of the photo, the app then guides the user through questions about the echidna itself, such as whether it was alive or dead, what activity it was doing, approximately how large it was, and what environment it was in. Finally, users can submit any scats that they collect, which requires them to take a photo of the scat at the time and location of collection, again to capture the related metadata associated with the collection. Participants are encouraged to collect scats if they are long, cylindrical in shape, dry in texture, and mostly composed of soil and insect exoskeletons; participants did not appear to have any apprehension to collecting echidna scats. Once the photo is taken, the app then guides the user on how to collect the scat, such as placing the scat in a plastic bag without touching it with bare hands (to avoid contamination of the scat), and then placing the scat in a freezer until ready to send to the University of Adelaide. Scats are then cataloged and stored at -80°C at the University of Adelaide. Echidna scats are dry and consist of mostly soil, they have no smell, and there has been no issues traveling through post. While there are no known pathogens in echidna scats, we do encourage participants to avoid direct contact with the scat. Users can submit data immediately in areas with mobile data or internet access; if out of range, the data can be submitted later by selecting a specific “upload” button within the app, or if the next data submission occurs within phone data range, then all previous submissions will be uploaded along with the current submission. For participants who could not or did not wish to use the smartphone app, an online submission form was created through the ALA's BioCollect platform [<https://www.ala.org.au:443/biocollect>] (68). The online form allows users to submit both sightings and scat collections by uploading a photo and self-selecting the GPS location along with answering the same sets of questions embedded within the EchidnaCSI app. Although EchidnaCSI is directly integrated with ALA, only sighting data submitted to EchidnaCSI (either through the app or through the EchidnaCSI BioCollect page) were used for data visualization in this study, not all echidna sightings recorded within ALA.

Communication and Engagement. EchidnaCSI was launched with a media release from the University of Adelaide and nationally televised interviews of lead researchers in September 2017. Following this, regular media engagements have further advertised the project. Over three years, EchidnaCSI has been the topic of >40 radio interviews, two television appearances, and >50 newspaper articles or online blog posts. Leaders of EchidnaCSI have also participated in 20 in-person talks within South Australia. A dedicated webpage was created for hosting information about EchidnaCSI, including how to use the app, what the research was aiming to achieve, and frequently asked questions. Facebook, Twitter, and Instagram accounts were created and updated at least weekly with information about the project and to share photos and videos that participants had submitted. When a participant downloads the EchidnaCSI app, there is the option to submit user contact details such as name, email address, and postcode. The users' email addresses were used to send a welcome email with links to the EchidnaCSI webpage and social media pages and to send updates about the project via an e-newsletter. Scat identification information and images are embedded in the app itself as well as on the EchidnaCSI webpage and social media channels.

Survey. The survey was designed and run through Qualtrics software (Provo, UT). A link to the survey was sent to 5,720 registered users via email and posted on all EchidnaCSI social media accounts (Facebook, Twitter, and Instagram) on 22 August 2019. The survey was active for ~3 wk, closing on the

8 September 2019, and participants were incentivized to complete the survey with the chance of winning one of five \$50 gift cards. Human ethics clearance was obtained through the University of Adelaide (HREC-2019-156). Survey participants were required to be 18 y or older to participate, and submission of the survey acted as user consent. The survey contained questions regarding their demographics and motivations; most questions were multiple choice, Likert-scale, or open answer (SI Appendix, Supplementary File 1). Data from the 2016 Australian Census were used to compare demographics from the survey to the Australian population [https://quickstats.censusdata.abs.gov.au/census_services/getproduct/census/2016/quickstat/036 (51)]. Questions relating to motivations were developed based on similar surveys in the literature, including citizen science projects Galaxy Zoo, Great Pollinator Project, iSPEX (Spectropolarimeter for Planetary Exploration), and a scoping study for potential of citizen science in marine research (23, 24, 69, 70). Pearson's Chi-Square analysis was used to determine whether any groups were significantly different for the demographic analysis and for comparing the "submitters" to "nonsubmitters" groups, with post hoc Bonferroni test to determine which groups were significant if the degrees of freedom were greater than 1.

Scat DNA Extraction and PCR. Total genomic DNA was extracted from scat samples using the Qiagen QIAamp Mini Stool Kit (Qiagen, Hilden, Germany) as per the manufacturer's protocol. The extractions took place in a Flow Cabinet Biological Safe Level 2 that was cleaned with 10% bleach (sodium hypochlorite) to reduce contamination. Approximately a third of the sample was crushed up in the presence of liquid nitrogen using a mortar and pestle, prior to adding the sample to InhibitEX Buffer, and then processed according to the manufacturer's instructions.

Samples were PCR amplified using primers designed to specifically target a unique region of the echidna mitochondrial D-loop (50). DNA was amplified with the primer pair Forward 5'-TGCATTCATCTTTATCCCATAC-3' and

Reverse 5'-TAATCTGTCAGAACCTCAATTATG-3'. Single reactions of 18.9 μ L dH₂O, 2.5 μ L 10 \times buffer, 1 μ L of 50 mM MgCl₂, 0.1 μ L IMMOBASE Polymerase (BioLine), 0.5 μ L of 10 mM dNTP (deoxynucleotide triphosphates) mix, 0.5 μ L of 10 μ M forward primer, 0.5 μ L of 10 μ M reverse primer, and 1 μ L DNA. DNA was amplified using an initial denaturation at 96 °C for 10 min, followed by 35 cycles of denaturation at 96 °C for 30 s, annealing at 50 °C for 1 min, and elongation at 72 °C for 2 min, with final adenylation for 7 min at 72 °C. To validate that the primers amplify on DNA extracted from scats, each round of PCR also contained a positive control, in which genomic DNA from echidna liver was used as well as a negative control. PCR product size (200 bp) was checked by gel electrophoresis (2.5% agarose).

Data Availability. Sighting data have been deposited in the Atlas of Living Australia Biocollect Database and is publicly available. EchidnaCSI BioCollect Database, <https://biocollect.ala.org.au/aca/project/index/8c3ae3b1-5342-40b4-9e72-e9820b7a9550> (71).

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