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METHODS
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An Arduino-Based RFID Platform for Animal Research

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Radio Frequency Identification (RFID) technology has been broadly applied in the biological sciences to yield new insights into behavior, cognition, population biology, and distributions. RFID systems entail wireless communication between small tags that, when stimulated by an appropriate radio frequency transmission, emit a weak, short-range wireless signal that conveys a unique ID number. These tags, which often operate without a battery, can be attached to animals such that their presence at a particular location can be detected by an RFID reader. This paper describes an RFID data-logging system that can serve as the core for a wide variety of field and laboratory applications for monitoring the activities of individual animals. The core electronics are modeled on an Arduino circuit board, which is a hobbyist electronics system. Users can customize the hardware and software to accommodate their needs. We demonstrate the utility of the system with cursory descriptions of three real-world research applications. The first is a large-scale deployment that was used to examine individual breeding behaviors across four local populations of Wood Ducks. The second application employed an array of RFID-enabled bird feeders that allowed for tests of spatial cognition. Third, we describe a nest-box monitoring system that both records visits from breeding birds and administers experimental treatments, such as increasing temperature or playing audio recordings, in accordance to the presence/absence of individual birds. With these examples we do not attempt to relate details with regard to research findings; rather our intent is to demonstrate some of the possibilities enabled by our low-cost RFID system. Detailed descriptions, design files, and code are made available by means of the Open Science Framework.

Keywords: biologging, feeder, nestbox, behavior, cognition, data logging, PIT tag, bioinformatics

INTRODUCTION

The term "biologging" typically evokes a research endeavor that involves tracking the locations of animals as they move throughout a home range or embark on a migration. However, biologging can also be employed to reveal intimate details about the behavioral patterns of individual animals. Radio Frequency Identification (RFID) technology has emerged as a useful biologging tool for monitoring animal activities at specific locations, such as feeding stations, nests, and burrows (Bonter and Bridge, 2011; Dogan et al., 2016; Bandivadekar et al., 2018; Iserbyt et al., 2018). RFID is a form of short-distance communication between a reading unit and one or more transponder tags (often called Passive Integrated Transponders or PIT tags). PIT tags use energy emitted by the reader unit to emit a weak signal (either with radio waves or magnetic coupling) that contains a unique identification code. PIT tags typically do not need a battery, which allows them to function forever (in theory), and they can be sized down for use on some very small species, including insects (Sumner et al., 2007; Russell et al., 2017; Barlow et al., 2019).

RFID typically entails only short-range communication, with tag-reading ranges from over 50 cm in high power systems to a few millimeters. Hence, a fundamental limitation to the system is that tags and readers must come close to an antenna for tag reading to occur. RFID is widely used as a marking method wherein tagged animals are captured and scanned manually in a manner similar to the "microchips" used in veterinary care of pets and livestock (Thorstad et al., 2013; Anu and Canessane, 2017). However, RFID can also be effectively employed in automated, remote sensing systems, with stationary readers that record specific animal activities, such as accessing a food source or nest (e.g., Zuckerberg et al., 2009; Bonter and Bridge, 2011; Catarinucci et al., 2014b; Ibarra et al., 2015; Zenzal and Moore, 2016). More advanced systems have implemented cognitive tests in lab and field settings with experimental routines customized for individual animals (Croston et al., 2016; Morand-Ferron et al., 2016)

Although RFID systems are relatively inexpensive compared to other forms of biologging, cost can still be a barrier to the use of RFID in research, especially if large numbers of reading units are required. Another barrier is customization. Many commercial RFID systems are configured for door-entry security or applications in commerce and these systems are not likely to benefit researchers who wish to track animals. To address these issues, we have developed a low-cost, short-range RFID reader that is compatible with a popular amateur electronics platform called Arduino (Arduino LLC, Scarmagno, Italy). The device is essentially an Arduino circuit board with two RFID-reader circuits and basic data-logging infrastructure (i.e., a real time clock and memory) built into it. Like all Arduinos the microprocessor on the circuit board can be programed using the open-source Arduino programming language and the free Arduino IDE (Integrated Development Environment) software. The device is also compatible with a wide array of accessories, such as environmental sensors, motor controllers, LCD screens, and wireless communications modules, that have been designed to work with the Arduino platform.

This paper provides both a brief description of this RFID system and accounts of its implementation in three different research capacities. The first account describes a simple data logging system that was deployed on a very large scale. Second, we describe a more sophisticated system that has been used to evaluate memory and cognition in free living animals. Finally, we describe a nest box that can carry out experimental protocols based on individual birds entering and leaving. We hope that these examples inspire new uses for our RFID system. To help new users get started we have made all of our designs, and code open source, and they are freely available via the Open Science Framework (OSF) at https://osf.io/9j7ax/. This OSF project page contains files available for download as well as links to Github repositories where current firmware versions are maintained.

MATERIALS AND EQUIPMENT

In designing the RFID reader, we took an open-source design for the Arduino M0, and added two RFID circuits, a real time clock, an SD card socket, and a flash memory module for data backup (**Figure 1**). We also simplified the voltage regulation circuit and reassigned some of the inputs and outputs to accommodate the tag-reading, time-keeping, and memory functions. The RFID reader retained the high-performance Atmel SAMD microcontroller that is featured on the Arduino M0. The RFID reader can be programmed in the same manner as the Arduino M0, using the Arduino IDE; however, we had to provide a customized board definition for the IDE because of the changes made to some of the input/output pins. A board definition is a collection of code that allows for a seamless interface between a particular piece of hardware and the Arduino IDE. This board definition as well as all design files for the RFID reader are accessible via our OSF project page.

This RFID reader design follows a previous version described in Bridge and Bonter (2011). However, the new design has several key features that were not previously available. Foremost is the ability to implement custom programming using open source tools—the previous version was programmed using a proprietary language. In addition, dual RFID circuits, a superior microprocessor, increased memory capacity, more input/output pins, and the Arduino compatible hardware, set the new design apart from its predecessor.

The RFID reader has two RFID circuits or modules built in. Users can alternate between the two modules or use just one of them. Each RFID module requires an external antenna, which usually consists of a thin coil of magnet wire. In our system, a functional antenna coil generally has to be <15 cm in diameter, but beyond that antennas can vary greatly in size and shape. Users of our system can either make their own antennas or purchase them from third-party providers (e.g., Q-kits, Kingston, Ontario, store.qkits.com). Antennas can be hand wound, such that users can customize antenna size and shape to match their application. The reader is designed to work with antennas with an inductance of 1.25–1.3 mH. Hence, the process of making antennas requires

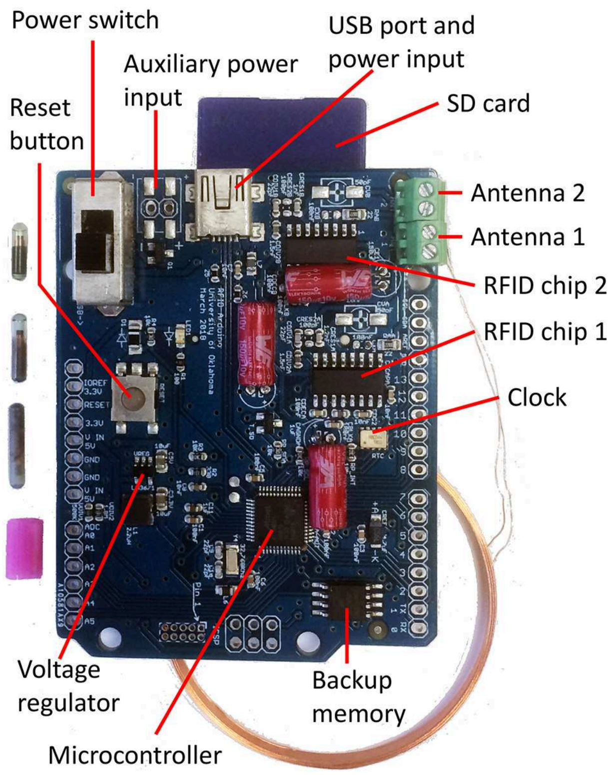

FIGURE 1 | The RFID reader circuit board with key components labeled. The dimensions of the board are those of a standard Arduino: 68.58 × 53.34 mm. Four PIT tags are shown to the left of the circuit board. The bottom tag is an RFID leg band.

inductance measurements with an LCR meter. We provide some guidelines for making antennas in our OSF project page, and users can follow our examples or use them as a starting point for their own designs. Either way, it will likely require some trial and error to find the correct number of turns for a given antenna size and shape. We plan to improve the process of antenna design with an online tool for simulating and testing RFID antennas, but this tool is still in development.

A typical deployment of an RFID reader involves polling for tags at regular intervals. In this context, "polling" means emitting a radio-frequency carrier wave at or around 125 kHz, while "listening" for a return signal from a tag. Emitting this carrier wave requires about 90 mA with a 5 V power supply, which is costly in terms of power requirements, so our firmware offers a power saving algorithm wherein a brief polling period of about 30 ms is used to determine if a tag is present. If no tag is detected, the reader can enter a "sleep" mode that uses about 100 μA for a specified period of time before polling again. If a tag is detected, then the reader will extend the duration of the polling effort to read the tag ID. The frequency and duration of reading attempts can be set by the user in a manner that balances power usage and the prospect of missing tags that enter the read range. In addition, the RFID reader can alternate polling efforts between its two RFID modules to attempt to detect tags with two different antennas.

An onboard SD-card socket is available for primary data storage. Data are typically stored in the form of a comma-delineated text file. Hence, the SD card can be removed from the circuit board and inserted into a computer or mobile device to transfer and examine data. There is also a flash memory unit (AT45DB321E) permanently attached to the circuit board, which can serve as back-up storage in case there is a problem with the SD card. This built-in memory unit has a capacity of 32 Mbits or 4,000,000 bytes, which is sufficient for recording over 200,000 data reads, assuming only a tag ID and timestamp are stored.

The circuit board can be produced in small quantities at a cost of about $30 (USD) per unit. The most expensive components are the microcontroller (∼$4), the real-time clock (∼$2), the SD card socket (∼$2), backup memory (∼$2), and the RFID front-end integrated circuit (EM4095; ∼$2). Prices can vary considerably depending on electronics components markets, and quantities purchased. The $30 production cost includes $3 for the circuit board and $10 for assembly. Full details and sources for components and assembly are available on the OSF project page.

The reader is configured to operate at 5 V. This voltage makes them compatible with a wide variety of power supplies designed for charging or maintaining cellular telephones. The circuit board may also be powered via a USB connection or a USB AC/DC converter/charger. It is possible to use higher or lower voltages, but that requires an additional power adaptor to establish a 5 V power supply for the circuit board. Solar-powered systems have been employed for some field applications, negating the need for large batteries and frequent battery changes. The OSF project page provides several suggestions for power supplies and photovoltaic systems.

As described above, the RFID reader alternates between polling for tags and "sleeping" in low-power mode. Based on measures of power usage for these two modes it is possible to calculate how long a given battery will last. For example, If there is a 1-s sleep interval between polling attempts and there is a minimal 30 ms poll time, average power use would be 2.7 mA and a 10 amp-h battery would last over 3,600 h. However, if tags are polled 5 times per second with a 30 ms polling period, then the same 10 amp-h battery would last about 730 h. These are very simplistic calculations. Another factor to consider is the power requirement of tag reads. Also, it is possible to implement a prolonged sleep period (i.e., nighttime sleep mode) to conserve power when animals are not likely to be detected. A spreadsheet calculator is provided on our OSF project page. This spreadsheet provides estimates of battery life based on RFID-polling settings, expected numbers of tag reads, implementation of prolonged sleep periods, and battery parameters.

The reader is configured to work at a frequency of 125 kHz, and it is compatible with tags that adhere to the EM41XX protocol, which includes EM4100 and EM4112. Other commonly used RFID communication protocols include the ISO11784/5 and Trovan standards. These protocols differ with regard to data encoding, the size and structure of the identification number, and the associated error checking algorithms as well as the bit rate and (in some cases) the radio frequency. Communication via these other protocols is possible with the ETAG reader but would require, at minimum, different tag reading functions in the firmware. It may also be necessary to implement hardware changes to be able to read other types of tags, especially if they work at frequencies other than 125 kHz. In addition, some forms of RFID involve active tags that often increase the read range by amplifying the transmitted signal. Thus far, our testing has only involved passive RFID tags (i.e., tags with no battery). Fortunately, compatible (EM41XX) tags are available in a wide variety of configurations including implantable glass ampoules and plastic leg bands. Tag are available through a variety of vendors. In particular, Cyntag (Cynthiana, Kentucky, USA) is a good source for glass ampoule tags, and Eccel technology LTD (Groby, Leicester, UK) is the primary supplier of small RFID leg bands. See our OSF website for more details and a list of compatible tags.

Because the RFID reader is based on the Arduino electronics format, a wide range of customization is possible both in terms of hardware and software. The reader is configured to mimic an Arduino M0. Hence it has 22 input/output pins that can serve as integration points for sensors, LCD arrays, lights, data interfaces, and motor controllers. The reader can be programmed using free, open-source Arduino IDE software. The Arduino programming language is a set of C/C++ functions, and the developers provide full documentation for programming with C in the Arduino IDE. There are also hundreds of libraries written for the Arduino IDE that provide accessibility to various hardware components (e.g., sensors, motor controllers, and LCD screens), with minimal programming requirements. The code we have developed for the RFID reader has custom routines associated with tag reading and power conservation, but it also makes use of existing Arduino libraries.

We have provided a core Arduino sketch (i.e., program) that runs the RFID reader as a simple data logger. This code can readily be configured to allow for a variety of different RFID polling strategies and sleep schedules. This code can also serve as a starting point for new sketches that incorporate new functionality. For example, some RFID tags can be programmed to store and later transmit data. Although we have not dealved into these methods, it should be possible to configure the RFID reader to program tags. This code as well as sketches for the projects described below are available through our OSF project webpage.

To demonstrate what is possible through the combination of low-cost and versatility made available by our RFID reader, we briefly describe three actual field applications that have broken new ground through the use of RFID-based biologging. These projects have employed a variety of different RFID hardware and software configurations and are not necessarily the same as the current system documented on the OSF project page. Nevertheless, the systems employed in these projects are very similar to the most current hardware version, and the current systems are equally capable of supporting these research applications. Note that it is not our intent for the descriptions below to serve as a final record of the ecological and behavioral research that was enabled by RFID technology. Rather we present these examples to illustrate a variety of scenarios where RFID is useful or even critical for addressing research questions.

METHODS

Implementation 1: Large-Scale Monitoring of Wood Duck Populations

Our first example of the utility of our RFID system is an effort to quantify brood parasitism and other breeding behaviors in Wood Ducks (*Aix sponsa*). Wood Ducks are facultative conspecific brood parasites, which means that females will sometimes lay eggs in the nest of another Wood Duck female (Bellrose et al., 1994). Researchers at the University of California (JME, TFS, ACO, BEL) deployed RFID data loggers on more than 200 Wood Duck nest boxes in the vicinity of Davis, California, with the aim of determining patterns of conspecific brood parasitism. Ultimately, this project sought to investigate life-history trade-offs and kin selection as explanations for the evolution of conspecific brood parasitism in a species with precocial offspring and a complex life history.

Each nest box in the study was equipped with a single-antenna RFID reader configured as a simple data logger. The antenna on each box encircled the entrance hole, such that a duck's tag ID would be recorded and stored each time it passed through the antenna upon entering and leaving (**Figure 2A**). The battery and circuit board were housed in a sealable plastic box affixed on the side of each nest box. This configuration allowed easy access for battery changes and data offloads. Each animal involved in the study had a pit tag implanted under the skin in the region of the back between the scapula. The research team tagged every female that used a nest box and all nestlings that hatched in the study area. Although the configuration of the RFID equipment was not remarkable, the scale of the effort was. The ongoing project has entailed monitoring hundreds of nests over the course of five field seasons, with as many as 197 systems active at once. Over three field seasons, this effort employed a total of 74 student volunteers engaged in changing batteries, offloading data, and troubleshooting. In the most recent field season, the research team designed and deployed a solar-charging system that would allow a reader to operate uninterrupted for an entire season (see **Figure 2A** and the OSF project page), greatly reducing personnel time and nest disturbance.

In total, the study followed 1,873 nest attempts by 454 breeding females (with 506 females registered at least once at a nest box). Some of these females were among the 4,128 ducklings that were tagged as part of the study. As a result of this intensive effort, up to 60% of breeding females had been tagged as ducklings in some populations, providing a unique opportunity to follow individuals of known origin, genotype, maternity, phenotype (size, physiological traits including hormones), and kinship to other females in the population throughout their entire lives. Because PIT loss rates were <1%, and battery power was not a limitation, the tags provided information for as long as there were active RFID readers and tagged birds present. During the study (now extending into its sixth year), over 1 million RFID detections have been recorded at nest boxes.

The RFID network revealed complex patterns of nest use among females (Eadie et al., in preparation). The distribution of the number of nests in which females visited and laid eggs was

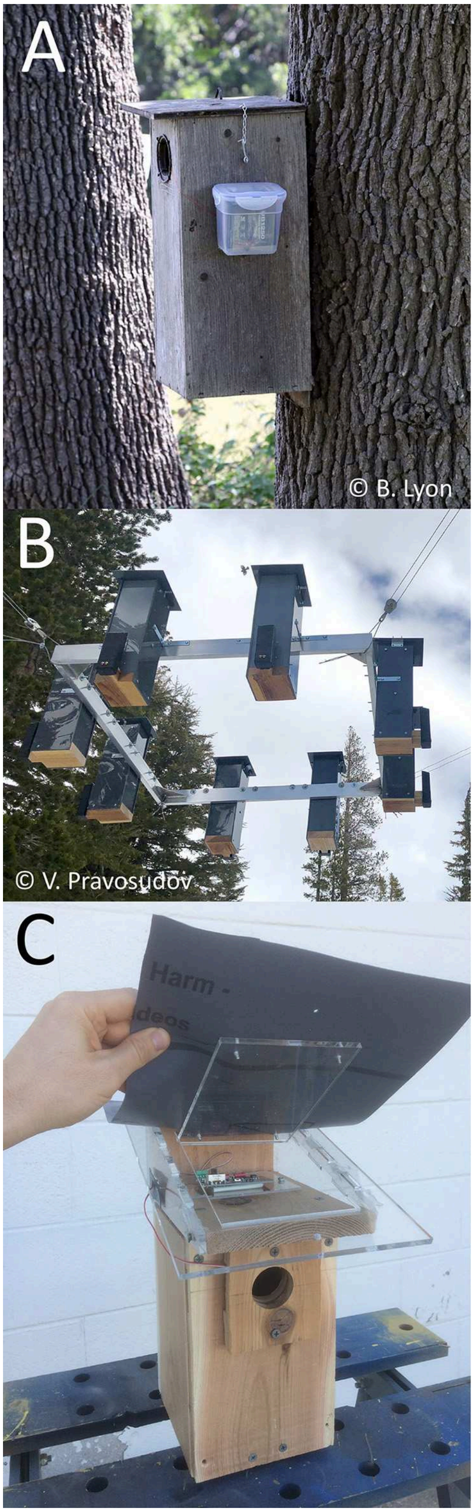

FIGURE 2 | (A) RFID-equipped Wood Duck nest box deployed near Davis California. RFID reader and battery are housed in a water-tight plastic box attached to the side of the box (painted in camouflage at other installations). The hand-wrapped and tuned antenna was coated in plasti-dip and secured around the nest entrance hole with zip ties. The antenna leads connect to the *(Continued)*

FIGURE 2 | RFID reader in the plastic box. In more recent implementations, only the RFID reader is mounted on the box in a smaller plastic case and the battery is moved to an external box on the ground and attached to a solar panel. This reduced weight and size of the box on the nest, insured constant charging of the battery via solar panel, and easier monitoring of the battery status. Also, field crews could check and download SD cards from the RFID reader without having to remove or work around the battery. **(B)** An array of eight RFID-enabled birdfeeders that can selective administer food to carry out spatial memory and cognitive tests. The array can be raised and lowered on cables such that the feeders were protected from non-avian foragers and suspended well above snow cover. Electronics and rechargeable lithium batteries are housed entirely inside the feeder. Antennas are imbedded into a wooden perch that is coated with waterproof epoxy. **(C)** A bluebird nest box capable of issuing experimental treatments (noise and temperature) in accordance with the bird or birds present in the box. The box employs two RFID reading circuits with separate antennas to better determine which birds are present. Electronics are housed in the "attic" above the nesting cavity, and they are accessed by lifting the flexible roof cover as shown. Batteries can also go in the attic or may be mounted externally.

highly non-random—some females ranged widely and visited numerous boxes (>25) while other females were faithful to only one or two boxes. There was also considerable variation in nest-box "attractiveness"—some boxes were visited by as many as 19 different females during a single breeding season, whereas other identical nest boxes nearby were never visited. Nest quality/attractiveness may be a key determinant of why multiple females lay eggs in the same nest. Preliminary social network analyses suggest that groups of females share similar nest site preferences and visit the same subset of nest boxes. Females in the same nest-visiting groups appear to be more likely to be related and from the same cohort (Stair et al., in preparation).

The results also suggest that conspecific brood parasitism is a flexible life history strategy that allows females to adjust reproductive effort to match investment with the probability of success (Lyon and Eadie, 2008, 2018). By integrating both a life-history perspective (what are females doing and why) and kinship (how does relatedness shape these tradeoffs), RFID technology coupled with population-wide genotyping has allowed for a more comprehensive understanding of the evolution and ecology of conspecific brood parasitism and its relation to other breeding systems. RFID monitoring at every nest site along with PIT-tagging all breeding females and nestlings provides a rare opportunity to follow individual females through their entire lifetime and probe more deeply into the ecological, physiological, and genetic factors that influence their intriguing breeding behavior.

Implementation 2: A Feeder Array for Assessing Spatial Memory and Cognition

A second example of the RFID system allows for tests of spatial cognition (spatial learning and memory) in Mountain Chickadees (*Poecile gambeli*) in the Sierra Nevada Mountains. These birds live year-round in mountainous habitat that can receive up to six meters of snow depth in the winter. To survive these conditions the birds must store or cache food (usually pine seeds) and then recover them days or weeks later when other food sources are unavailable in winter. Hence, a large capacity for spatial cognitive ability is key to the survival of these birds, and individual variation in spatial learning and memory ability is of great interest in understanding the ecology and evolution of these animals as well as food-caching species in general.

Researchers at the University of Nevada (VP, CB, DK, AP, and BS) have devised and implemented a means of evaluating spatial learning and memory in a population of Mountain Chickadees using arrays of RFID-enabled bird feeders that can selectively feed specific individuals. The feeders are equipped with a motorized door, controlled by the RFID circuit board, that can be lowered or raised to allow or deny access to food. An antenna is embedded in a perch positioned in front of the door such that the feeder can recognize individuals on the perch and operate the door accordingly to provide or withhold food. Movement of the door is accomplished with a rack-and-pinon gear mechanism powered by a small gear motor. A limit switch is used in conjunction with the door to indicate to the circuit board when the door had reached a fully open or fully closed position. The motor is controlled by a customized accessory circuit board that features a TB6612FNG motor controller (details on OSF project page).

The learning and memory tests employed arrays of eight RFID-enabled bird feeders that were mounted together on a square frame with two feeders on each side facing outward (see **Figure 2B** and Pitera et al., 2018). One or two of these arrays (depending on the study) were situated at high and low elevations sites. The arrays were suspended on cables stretched between trees so the feeders could be raised above the reach of bears and rodents. Prior to testing, the feeders were all configured in the open position. That is, the doors were all open such that all birds have access to clearly visible food (sunflower seeds). RFID data logging was enabled at this time such that the researchers could determine which tagged birds were visiting the feeders, but the RFID reads did not affect access to food.

After this initial acclimation period of about a week, the feeders were switched to "feed-all mode," wherein the door remained closed until any bird with a tag is detected on the perch. In "feed-all" mode, any bird with a tag will cause the feeder door to open. This training mode allowed birds to become accustomed to the movement of the door.

After another initialization period in feed-all mode, the feeders were reconfigured into "target mode" such that each bird will have access to food at only one of the eight feeders. Spatial learning and memory were assessed based on how many non-rewarding feeders a bird visited prior to visiting the assigned, rewarding feeder during each trial. A trial starts when a bird visits any feeder in the array and ends with the visit to the rewarding feeder. Birds that quickly learn and remember the location of the rewarding feeder (as evinced by progressively fewer visits to non-rewarding feeders) are deemed as having superior spatial learning and memory ability. In addition to testing spatial cognition, the system can test reversal spatial learning and memory performance by switching the rewarding feeder for each bird after the completion of the spatial learning and memory task (which usually took 4 days). The number of errors (e.g., number of non-rewarding feeders visited prior to visiting the rewarding feeder) during a 4-days reversal trial provides a measure of spatial learning and memory flexibility as a bird needs to stop visiting the feeder that provided food in the previous spatial learning and memory task and learn the location of a new rewarding feeder.

This system was applied to Mountain Chickadees living at two different elevations. The birds at high elevation (∼2,400 m) face more severe and longer winter conditions and rely more heavily on caching and recovering food for overwinter survival than do birds at lower elevation (∼1,900 m). The RFID system provided a means of testing several key hypotheses relating spatial cognitive ability to environmental conditions and fitness. First, the spatial cognitive tests revealed that the high elevation birds had better spatial learning and memory ability than did the low elevation birds (Croston et al., 2016). Second, the system revealed that individual variation in spatial learning and memory performance is associated with differences in survival in first-year, juvenile birds during their first winter, showing that spatial cognition is affected by natural selection at high elevations (Sonnenberg et al., 2019). Moreover, a series of spatial learning and memory reversal tests suggested a potential tradeoff between cognitive flexibility and spatial learning/memory (Croston et al., 2017; Tello-Ramos et al., 2018). RFID data collected in this system were also used to show that daily foraging routines in chickadees differ between elevations and among seasons. Moreover, it was apparent that spatial learning and memory performance was associated with daily foraging routines. In particular, chickadees with better spatial cognition had daily foraging routines that resembled those in milder environment and seasons, likely due to greater predictability of foraging success for these individuals (Pitera et al., 2018). Most recently the system has revealed that females allocate more reproductive effort when mated with males with better spatial learning and memory performance (Branch et al., 2019). Finally, ongoing work in this system involves analyzing chickadee social networks using RFID data to address the associations between social and cognitive phenotypes as well as the potential role of cognitive phenotype in structuring these social networks.

Implementation 3: A Nest Box Platform for Experimental Manipulation

Several studies have already employed RFID to generate a detailed activity log for birds that use artificial nest boxes (e.g., Johnson et al., 2013; Stanton et al., 2016; Zarybnicka et al., 2016; Schuett et al., 2017; Chien and Chen, 2018; Firth et al., 2018). With this third application example we describe a project that takes the next logical step forward—using RFID to orchestrate experimental treatments that can be applied individually to breeding adults and offspring. This project involved a nest box designed for Eastern Bluebirds (*Sialia sialis*) that could manipulate environmental noise and/or nestbox temperature in accordance with which birds were inside the box (**Figure 2C**). The project has progressed through several versions of the RFID-enabled nestbox, but not all features have been tested in the field (notably the temperature manipulation). Hence, the functions described here should be regarded as features that are available for a technology-driven nestbox study.

To help ensure that treatments are applied accurately to the targeted individuals, the nestbox was configured to make use of two antennas. One antenna was mounted to encircle the entrance to the nest box, and it would capture birds as they entered or exited. The second antenna was positioned inside the nest box, and it could verify the presence of a bird therein. The tag-polling strategy was conceived such that the entrance antenna was used for most of the monitoring, and polling by the internal antenna was only done periodically or when the entrance antenna had received a tag. This arrangement was necessary because birds will often rest on the nestbox entrance but not go in. Or they may fly up to the entrance but quickly fly away. The dual antenna system provided assurance that a bird had entered the nestbox.

Following the general example of Lendvai et al. (2015b), artificial noise was incorporated into the system by means of a generic serial MP3 music player module and a 1 W speaker (catalex.taobao.com). These items were purchased as a single kit for about $4. The MP3 player accesses and plays audio tracks from a mini-SD card. The microcontroller on the RFID circuit board communicated with the MP3 player via a simple serial interface. Hence, the RFID reader could control the timing, duration, and volume of playbacks. We used a transistor to control the power supply to the MP3 player, such that it could be powered down when it was not being used (see OSF project page for details). Similarly, we have configured the RFID circuit board to control the power supply to an electronic heating pad (WireKinetics Co, Ltd, Taipei, Taiwan). As with the speaker system we used a simple transistor circuit to provide current to the heating pad directly from the power supply (i.e., battery).

As part of a pilot study, a single noise emitting system was deployed on an active bluebird nest in Kent County Michigan. The system was programmed to emit noise from 05:30 to 11:00 each day when both parents were absent from the nest box. The system was successful in administering an effective treatment throughout the majority of a nesting effort. The sample size is, of course, too small to reach any conclusions, and based on this initial effort, we plan to continue this experiment in future breeding seasons. The heating pad has not yet been tested, but we plan to do so in the spring of 2019, and the results will be posted to our OSF project page.

DISCUSSION

The example studies described above do not by any means exhaust the possibilities offered by our low-cost RFID system. Given the wide array of hardware and software tools available as part of the Arduino electronics platform there are many other applications with respect to data logging and experimental manipulations that could be brought to bear in the context of RFID-based biologging. A few potential and/or recently executed applications are as follows:

- Interfacing the RFID reader with a WiFi network to get wireless RFID data transferred in real time to a data collection hub (Ramudzuli et al., 2017; Rosval, personal communication).
- Using visits to a feeding station to generate association data for use in a network model (Psorakis et al., 2015; Zonana et al., 2019).
- Evaluating and quantifying social behavior in free living animals (Aplin et al., 2013, 2014, 2015b; Zeus et al., 2017; Sabol et al., 2018).
- Administration of manipulation-based cognitive tests (Aplin et al., 2015a; Morand-Ferron et al., 2015).
- Monitoring use of food or other resources (Crates et al., 2016).
- Selective food supplementation experiments (Small et al., 2013).
- Targeted delivery of nutrients or drugs to specific animals (Schoech and Bowman, 2003).
- Environmental data logging to match conditions with behavior.
- Nest visitation patterns in colonial species (Leighton and Echeverri, 2016).
- Quantifying movement and exploratory behavior (Catarinucci et al., 2014a; Ousterhout and Semlitsch, 2014).
- Monitoring health and behavior in captive animal populations (Whitham and Miller, 2016).
- Quantifying stopover duration and behavior in migrating species (Zenzal and Moore, 2016).
- Automated Collection of body-mass and other morphometric data (Hou et al., 2015; Small, unpublished).

As new applications come online, we intend to incorporate them as modules into our OSF project page.

There is a clear taxonomic bias in the examples described in this paper; they are all applied to avian study systems. However, this bias is due to the common interests of the research teams and not limitations in the applicability of RFID systems to other taxa. PIT tags have been used in a wide variety of vertebrates and even on a few insects (Sumner et al., 2007; Bonter and Bridge, 2011; Dogan et al., 2016; Whitham and Miller, 2016). Nevertheless, their applicability to bird research should not be surprising majority of bird species require small (<1 g) tracking devices.

A major advantage to automated systems like the ones described here is the potential for continuous data recording. Traditional monitoring methods often involve sampling the activity schedules of animals—for example, a researcher may visually monitor a burrow entrance for 3 h every other day, or one may video record activity at a feeding station for 5 h every day. Although these methods may be adequate for finding averages associated with events that happen regularly, they may be inadequate for detecting rare or even infrequent events (like brood parasitism or fledging). Automated systems obviate the need for sampling such that you can capture the entirety of a breeding attempt or activity period (see Lendvai et al., 2015a).

As is the case with all research efforts that involve attaching devices to animals, researchers must make every effort to minimize the adverse effects of equipping animals with PIT tags. There are numerous studies of the effects of PIT tags on survival and behavior, with most of this research focused on fish (Keck, 1994; Low et al., 2005; Nicolaus et al., 2008; Burdick, 2011; Thorstad et al., 2013; Guimaraes et al., 2014; Ratnayake et al., 2014; Moser et al., 2017; Schlicht and Kempenaers, 2018). The vast majority of these studies conclude that the effect of implanted PIT tags is negligible. The Wood Duck research described above entailed injecting tags in both females and ducklings intrascapularly using a PIT-tag syringe, and in 6 years, no deleterious effects have been detected on females or ducklings in the wild. Related projects have involved tagging >500 ducklings in captivity and raising >200 to the fledging stage with no adverse effects on survival or health observed in the course of regular monitoring by staff veterinarians. Tag loss has been <1% (Eadie et al., in preparation).

We do not know of a systematic study that has investigated the effects of externally mounted PIT tags. In particular, external mounting would largely apply to bird species, wherein PIT tags are incorporated into leg bands (see **Figure 1**). Our own experience has indicated no problems other than those typically associated with leg bands. More specifically, the studies of Mountain Chickadees described above have involved tagging over 1,000 individuals with RFID leg bands. Thus far, there has been one instance where the leg band appeared to cause an injury. However, there are unpublished reports of apparent injuries and mortality in association with RFID leg bands (Curry, unpublished data; Morand-Ferron et al., personal communication). We advocate the use of compact RFID leg bands like those supplied by Eccel technology, which minimize the size of the tag (see **Figure 1**), and we recommend caution in selecting the appropriate band size for the species (or individual) being tagged.

It is important to note that our device is not the only RFID reader available for field biologists. Among the alternatives for low-frequency (120–150 MHz) readers, the lowest-cost options include several RFID reader modules that can simply read tag data and communicate with a computer or microcontroller. Examples include the ID-12 and ID-20 readers (ID Innovations, Canning Vale, WA, Australia), the Paralax RFID module (Parallax, Inc, Rocklin, CA, USA), the MIKROE-262 and MIKROE-1434 modules (Mikroelektronika D.O.O., Belgrade, Serbia), and the RFIDREAD-RW Module (Priority 1 Design, Melbourne, Australia). These sorts of modules range in cost from about $10–50 (USD). There are a few commercially available RFID readers that can log data in a manner similar to our ETAG reader. For example, Priority 1 Design offers the RFIDLOG circuitboard for ∼$50 (USD). A more robust, fully enclosed RFID reader/datalogger is available from Eccel Technology, Ltd (Leicester, UK), for ∼$400 (USD). For applications that require read ranges >2–3 cm, there are high power RFID readers capable of read distances of 50 cm or more. Unfortunately, readers that offer this increased read range will typically cost considerably more than the low power systems. Examples of high-power systems include Biomark readers, such as the IS1001, which costs ∼$1,500 (USD), and systems from Oregon RFID (Portland, OR, USA) with a minimum cost of just over $2,000 (USD). Although there are many RFID readers available, there are few if any standalone systems that can be programmed by the user to carry out complex protocols. Our ETAG reader offers this key feature.

To facilitate widespread collaboration and sharing of information, we have established a project within the Open Science Framework at https://osf.io/9j7ax/. This information repository contains technical information relating to the RFID system and the research applications described in this paper, and it is open to the public to view and download materials. It also allows for ongoing updates and expansion such that we can continue to add new applications and contributors into the foreseeable future. New developments on the horizon include wireless networks for real-time data delivery, and online software tools for designing antennas that will culminate in an online simulator for testing virtual antennas prior to making a physical prototype. We are also developing web-based data portal and archive for uploading, storing and managing RFID data. We hope that our RFID system and the forthcoming tools will prove to be useful resources for the biologging community, and we invite our readers to take part in our efforts to expand the utility of RFID technology for animal tracking and monitoring.

ETHICS STATEMENT

All animal research was conducted according to accepted ethical standards and with approval from the authors' respective Institutional Animal Care and Use Committees (IACUC). Specifically, the research described in this manuscript followed IACUC-approved protocols R16-010, 00603, and 20971 for the University of Oklahoma, the University of Nevada Reno, and the University of California Davis, respectively. All field efforts sought to minimize negative impacts on birds by working with animals only during favorable conditions and by following best practices for animal handling and tagging.

AUTHOR CONTRIBUTIONS

EB participated in designing and implementing the RFID systems and drafted the main text of the article. JW and AM helped design the RFID circuit board, wrote key elements of the firmware, and served as a grant PI. MP and DP designed and implemented the experimental bluebird nestbox and contributed to writing this section of the text. CH designed and implemented the solar charging system used on the bluebird boxes. CC served as a grant PI and along with TP has built data infrastructure for the RFID system and managed the OSF project page. JE, TS, AO, and BL carried out the field study of Wood Ducks, and JE and BL helped draft this section of text. CB, AP, DK, BS, and VP conceived and executed the spatial memory testing. VP and AP helped to draft this section of the text. JR served as the overall project manager and grant PI for the project, and she provided editorial assistance on the manuscript.

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