



From eDNA to citizen science: emerging tools for the early detection of invasive species

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Biological invasions are a form of global change threatening biodiversity, ecosystem stability, and human health, and cost government agencies billions of dollars in remediation and eradication programs. Attempts to eradicate introduced species are most successful when detection of newly established populations occurs early in the invasion process. We review existing and emerging tools – specifically environmental DNA (eDNA), chemical approaches, remote sensing, citizen science, and agency-based monitoring – for surveillance and monitoring of invasive species. For each tool, we consider the benefits provided, examine challenges and limitations, discuss data sharing and integration, and suggest best practice implementations for the early detection of invasive species. Programs that promote public participation in large-scale biodiversity identification and monitoring (such as iNaturalist and eBird) may be the best resources for early detection. However, data from these platforms must be monitored and used by agencies that can mount appropriate response efforts. Control efforts are more likely to succeed when they are focused on early detection and prevention, thereby saving considerable time and resources.

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Biological invasions are one of many forms of anthropogenic change that threaten biodiversity, the structure and function of ecosystems, human health, and the global economy (Ehrenfeld 2010; Bradshaw *et al.* 2016). Once established in previously unoccupied areas, invasive species can be nearly impossible to eradicate. Early detection of incipient populations is therefore critical if control actions are to be both effective and affordable (Vander Zanden *et al.* 2010).

In a nutshell:

- Given the difficulty of eradicating introduced species after they have become established, scientists and resource managers need to develop best practices for early detection
- Emerging technologies, including environmental DNA, remote sensing, chemical ecology, and internet-based applications for engaging citizen scientists, can facilitate the early detection and monitoring of introduced species
- These approaches may be particularly valuable when used together, especially when coordinated with existing governmental programs for invasive species management

Surveillance for new biological invasions is being transformed by changes in how the environment is monitored and who is doing the monitoring. Advances in technologies like remote sensing and environmental DNA (eDNA), in combination with the rise of vast citizen-science initiatives enabled by the internet and smartphones, are revolutionizing our ability to detect and respond to invasions (Steenweg *et al.* 2017; Allan *et al.* 2018). However, these changes are accompanied by a host of new logistical challenges, cost considerations, and data sharing and management hurdles that must be reconciled to put these emerging tools and their discoveries to best use (Packer *et al.* 2017).

We review emerging approaches for surveillance of biological invasions and relate these technological changes to transformations being experienced by citizen-scientist and government agency monitoring programs. We focus on developments and advances in molecular biology, chemical ecology, and remote sensing, as they relate to monitoring or surveillance for the detection of new invasions. We emphasize recent advances in surveillance tools, provide examples of successful applications of these tools for the early detection of biological invasions, and identify challenges to their implementation as well as potential solutions. We then discuss recent changes to citizen science and government agency invasive species monitoring programs in response to technological advancements, including internet databases and smartphone applications for identifying species and reporting their locations. Finally, we propose opportunities for these emerging surveillance-based approaches to be used jointly to improve our ability to detect and respond to newly arrived invaders.

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eDNA

Technological advances in isolating and amplifying DNA have resulted in the rapid rise of non-invasive molecular species detection methods (Barnes and Turner 2015), including classical non-invasive genetic sampling and eDNA. Classical non-invasive genetic sampling uses biological material deposited by an organism of interest (eg feces, hair strands, urine, feathers, scent marks, eggshells, sloughed epidermal cells) without the need for handling or capturing the organism itself (Waits and Paetkau 2005). In contrast, eDNA uses genetic material extracted and identified from environmental samples, such as soil and water, where the biological material deposited by an organism is not necessarily known or identified (Taberlet *et al.* 2012). Both approaches share many general field and laboratory techniques, but we focus on eDNA, given its rapidly escalating use in surveillance for invasive species (Figure 1).

There has been a marked rise in the application of eDNA approaches to invasive species surveillance across freshwater, marine, and terrestrial ecosystems over the past decade (Jerde *et al.* 2013; Barnes and Turner 2015; Valentin *et al.* 2018). eDNA has been proposed as an effective invasive species surveillance tool because it is often more sensitive in detecting organism presence than conventional methods (Goldberg *et al.* 2016). The expanding use of eDNA for species surveillance has been facilitated by the increasing availability and affordability of techniques like quantitative or real-time polymerase chain reaction (PCR; qPCR, RT-PCR), digital droplet PCR (ddPCR), and high throughput sequencing (HTS), all of which can detect trace amounts of DNA from environmental samples. Two primary eDNA survey methods have been implemented for invasive species detection: (1) active surveys for single species and (2) passive surveys for many species using HTS methods (ie DNA metabarcoding; Deiner *et al.* 2017). Currently, single-species eDNA approaches are being used as part of an interagency monitoring strategy for invasive fish in the Great Lakes region of the US and Canada (Jerde *et al.* 2013), whereas multispecies HTS approaches are being investigated for their ability to screen ballast water for the incidental introduction of invasive species to freshwater and marine ports (Zaiko *et al.* 2015).

Users of eDNA technologies should be aware of some potential pitfalls (Roussel *et al.* 2015). Both false negatives (the failure to detect a target species when actually present) and false positives (the detection of a target species when actually absent) can occur with eDNA surveillance, just as they can for many other monitoring approaches in applied ecology (Ficetola *et al.* 2015). False negatives in eDNA can be caused by such factors as poor assay design or inhibition of PCR by common



Figure 1. A typical process of using eDNA for invasive species surveillance includes (a) collecting environmental samples and (b) capturing DNA through a method like filtering water through fine pore cellulose nitrate filters, which may be performed in the laboratory or (c) immediately under field conditions. Stored samples can then be extracted in the laboratory and analyzed using single species (eg quantitative or real-time PCR) or multispecies (eg metabarcoding) approaches.

environmental substances (eg humic acids), whereas false positives often occur as a result of field or laboratory contamination (Barnes and Turner 2015). Protocols to minimize the risk of contamination, and to screen for its incidence, are integral in using eDNA to survey for invasive species (Goldberg *et al.* 2016). Modeling methodologies like occupancy estimation with detection probabilities are also recommended to quantify and account for the rates of false negatives and false positives among eDNA samples (Dorazio and Erickson 2018). False positives may also be caused by environmental factors, including the directional flow of water and the transport of DNA by predators (Barnes and Turner 2015).

Despite these limitations, eDNA has many benefits beyond its sensitivity to the presence of rare or hard-to-detect taxa. It may be cost effective relative to many conventional approaches (Evans *et al.* 2017), and the simplicity of sample collection makes it an appealing tool for citizen scientists (Larson *et al.* 2017). However, laboratory processing of eDNA samples requires technical expertise and careful adherence to best practices in this methodology (Goldberg *et al.* 2016). Managers or researchers seeking to use eDNA to survey for invasive species can contract services from government agencies or consulting laboratories that have developed eDNA capacity. Although requiring bioinformatics expertise, HTS-based metabarcoding offers substantial capacity to survey entire communities with relatively little field sampling effort (Deiner *et al.* 2017). Managers should anticipate an escalation in the surveillance of invasive species through techniques using eDNA, given its sensitivity to species presence at low abundances, its ease of use in collecting field samples, and its applicability across a breadth of ecosystem types.



Figure 2. (a) Panel traps with chemical lures based on sex and aggregation pheromones are used to detect native and introduced wood-boring cerambycid beetles in central Illinois. In addition to presence–absence data, these traps provide information on mating phenology and conspecific attraction. (b) An assortment of pheromone traps used to detect outbreaks of forest or agricultural pests.

■ Chemical cues

Chemoreception plays an integral role in a variety of behaviors, including competition, mating, conspecific attraction, predator avoidance, predator and parasitoid defense, foraging, and alarm signaling. Organisms' dependence on chemical lures or cues in such behaviors can be exploited for attracting and detecting invasive species (Figure 2). Indeed, chemical lures have been applied for the detection, management, and control of both aquatic (Sorensen and Johnson 2016) and terrestrial (El-Sayed *et al.* 2006) invasions. In addition, the chemoreception capabilities of domestic dogs (*Canis familiaris*) and other animals have been employed for detecting introduced species that are cryptic and/or difficult to find (Engeman *et al.* 1998; Lin *et al.* 2011). Traps baited with sex pheromones or kairomones (a chemical produced by one species that modifies the behavior of a different species) can be effective for determining the presence or pattern of spread of invasive species (Hanks *et al.* 2012). Furthermore, lures may be tailored to target a single species or multiple species (Brokerhoff *et al.* 2013).

There are a number of challenges to consider regarding the use of chemical lures and other chemical-based methods for detecting invasive species. The scientific effort and monetary cost of chemical identification and production can be considerable. Consequently, knowledge gaps persist in terms of the specific compounds that most organisms use as signals to attract mates and find hosts, as well as cues associated with recruitment and migration (Brokerhoff *et al.* 2013). The use of certain chemical lures (once manufactured) could later be restricted by government regulations, given that even natural pheromones may be considered as pesticides in aquatic systems (Sorensen and Hoye 2007). There are further challenges associated with detection of molecules present at low concentrations and below the sensitivity of instrumentation. Moreover, the effective distance and duration of signals can vary considerably due to the compounds' chemical properties and their interaction with varying environmental conditions.

Moving forward, chemoreception research should prioritize characterizing attractants and synthesizing chemicals for field-based applications for actively spreading and emerging invasive species. Whenever possible, identifying chemicals that are attractive to multiple species should be a priority. Research should also determine effective distances, seasonal constraints (eg for mate attraction), and optimal spacing of lures to maximize the chances of detection. Some best practices for early detection include (1) applying multiple lures (attractive to a wide variety of invasive species) at sites vulnerable to invasion, and (2) placing lures near ports of entry, vulnerable waterways, or other areas where particular invasive species are likely to first appear.

■ Remote sensing

Remote sensing (RS) refers to various techniques that obtain information without the physical presence of a human observer (Figure 3). Many methods of RS used in ecology involve passive detection of bands of the electromagnetic (EM) spectrum, including visible light and infrared radiation (Bradley 2014). Active RS methods, such as LiDAR (light detection and ranging), emit radiation and then detect and measure its return. Although not typically considered RS techniques, methods such as acoustic monitors and camera traps also collect data remotely, without human presence, and are valuable tools in invasive species detection (Mankin *et al.* 2011; Rassati *et al.* 2016). Many RS projects utilize recent technological innovations or existing databases, such as Google Street View images (Deus *et al.* 2016), unmanned aerial vehicles (UAVs; Hung *et al.* 2014), and automated image recognition (Bradley 2014; Rassati *et al.* 2016).

The breadth of RS methods and recent growth in both remote measurement and data processing technologies have created many innovative opportunities for early detection and monitoring of invasive species (Bradley 2014). Hyperspectral RS, or the use of a larger number of narrower EM bands, distinguished native from introduced trees at the leaf and canopy level (Grobe-Stoltenberg *et al.* 2016), whereas airborne spec-

tral imagery and LiDAR successfully detected invasive species in Hawaiian rainforests (Asner *et al.* 2008). In addition, camera traps in conjunction with image recognition were used to monitor wood-boring beetle species in New Zealand (Rassati *et al.* 2016). While clearly effective for monitoring established species, there are fewer examples of successful early detections with RS. Camera traps detected the presence of gray squirrels (*Sciurus carolinensis*) in a previously unoccupied forest patch within Adda North Regional Park in Lombardy, Italy (Di Cerbo and Biancardi 2013). Space-borne data from the Satellite Pour l'Observation de la Terre (SPOT) and Moderate Resolution Imaging Spectroradiometer (MODIS) were used to construct maps of the damage to the Norway spruce (*Picea abies*) caused by an invasive scale insect (*Physokermes inopinatus*); damage to spruce stands was first reported in 2010, but satellite mapping revealed damage dating back to 2009 (Olsson *et al.* 2012). Even though these examples are promising, many RS technologies may not be effective for detecting newly established populations when abundance is low.

Major logistical challenges in implementing RS techniques include technical expertise, data resolution, storage and computational services, and cost. Some managers may lack the expertise needed to fully implement RS in their early detection operations. RS data processing, such as atmospheric and radiometric corrections, require considerable technical expertise. However, scientists in the RS community have worked to make this tool more accessible by publishing useful guides (Young *et al.* 2017), and by providing surface reflectance data that have been preprocessed and are ready to use in ecological analyses (eg Landsat Level-2 and Analysis Ready Data [ARD] imagery). Google Earth represents an alternative option that requires very little technical expertise, although its applications are fairly limited (Visser *et al.* 2014). Another logistical challenge of using RS for early detection is uncertainty in the spatial distribution of new outbreaks. Fine-scale spatial resolution is needed to detect incipient populations; however, a trade-off exists between resolution and total spatial coverage, which may constrain the effectiveness of RS as an early detection tool in many areas (Olsson *et al.* 2011). The large datasets generated by and used for RS approaches also require infrastructure for analysis and storage, although this barrier is being overcome with the onset of cloud computing and with the decreasing cost of digital storage. Google Earth Engine, for example, provides a free, large-scale computing platform with multiple petabytes of RS data available from many sources (Gorelick *et al.* 2017).

Finally, cost plays an important role in the ability to deploy various RS techniques for early detection. High spatial resolution imagery can be expensive, especially with large areas of interest. WorldView-3 (30-cm resolution), for example, costs between US\$17–58 per square kilometer, depending on image data and type, with a minimum order of 100 km² (LAND INFO 2018). High-resolution commercial satellite imagery is frequently made available for free or at substantial discounts for research and academic purposes. Multispectral imagery



Figure 3. Daniels and Larson (2020) demonstrated how remote sensing can be used to map disturbances like forest blowdowns (a) before and (b) after tornadoes or other wind storms. (c) These disturbed patches may be priority sites for monitoring biological invasions, including non-native understory plants. One-meter-resolution remote-sensing imagery in panels (a) and (b) from the National Agriculture Imagery Program.

ranging from medium to high spatial resolution is available for free from many sources, including Sentinel-2 (10 m); Landsat Thematic Mapper (TM), Enhanced Thematic Mapper Plus (ETM+), and Operational Land Imager (OLI) (30-m resolution); and the National Agricultural Imagery Program (1-m resolution). Some applications require expensive custom imagery for increased spatial, temporal, or spectral resolution, but costs of custom imagery are falling with the increasing use of UAVs (Koh and Wich 2012). Camera trapping has increased in sophistication and become more affordable (Burton *et al.* 2015) since its inception, making it a cost-effective management tool, especially when combined with image recognition software.

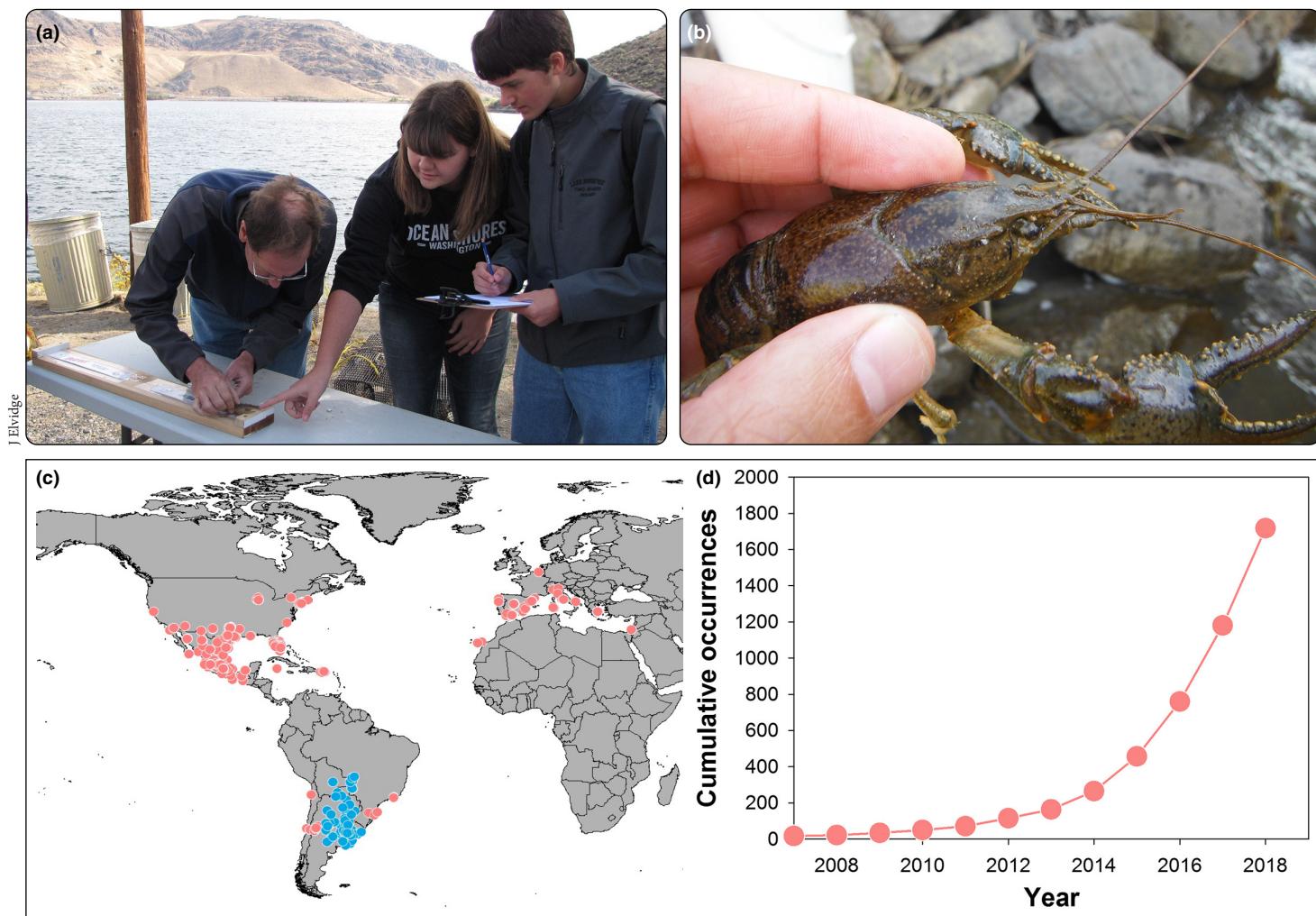


Figure 4. Coordinated by the US National Park Service, (a) The River Mile Network (WebTable 1) is a citizen-science and education program that trains educators and students to monitor for aquatic invasive species in the western US, including (b) the virile crayfish (*Faxonius virilis*). At much larger scales, smartphone applications such as iNaturalist allow individuals to report observations at any time and from any location. (c) A distribution map of the monk parakeet (*Myiopsitta monachus*) using research-grade observations from iNaturalist, representing the species' native range (blue circles) and non-native occurrence records from Africa, Europe, North America, and South America (red circles). Research-grade observations have dates, georeferenced locations, photos or sounds for identification, and two-thirds agreement among identifiers on the taxon. (d) The accumulation of *M. monachus* research-grade occurrences over time reflects the growth in biodiversity data from applications like iNaturalist. Data for panels (c) and (d) accessed from www.gbif.org on 6 Nov 2019.

■ Citizen science

Citizen science is the collection and analysis of data relating to the natural world by members of the general public, typically as a part of a collaborative project with professional scientists. Notably, to qualify as citizen science, a project must not only rely on volunteers who participate in the detection process but also include the use of any number of tools (eg collaborative databases, eDNA, or other technology). Using citizen science for the early detection of invasive species has recently become possible at large scales due to the development of collaborative technology, social media and networking, and publicly accessible databases that create opportunities for anyone to participate in ecological research (Figure 4; WebTable 1).

Citizen science can be a powerful tool for early detection of invasive species given its potential to collect high volumes of data over large areas (Silvertown 2009; Crall *et al.* 2010). Generally, there are two main types of citizen-science projects: (1) large-scale data collection that may passively record first detections or (2) place-based research that actively searches for early signs of invasion. Large-scale citizen-science data collection has successfully monitored range expansions of many non-native species. For example, data from the Christmas Bird Count tracked the range expansion of several introduced birds in the US, including the Eurasian collared dove (*Streptopelia decaocto*; Hooten and Wikle 2008). However, focused local research has also identified first detections and range expansions (Bois *et al.* 2011); examples include mapping of the invasion front of an invasive mammal in Ireland, the first

observations of invasive fish in marine protected areas in Mexico, and the discovery by pastoralists of an invasive plant in Ethiopia (Goldstein *et al.* 2014; López-Gómez *et al.* 2014; Luizza *et al.* 2016).

Although citizen science lends itself well to large-scale data collection, it also presents several challenges. One concern is access to appropriate technology (Silvertown 2009). Increasing availability of the internet and smartphones may help facilitate these types of projects but keeping up with rapidly progressing technology can prove difficult. Area-specific and up-to-date technology is of the utmost importance for proper identification of organisms and successful delineation of invasion fronts. Another concern is the quality of data produced by non-professional observers (Silvertown 2009; Crall *et al.* 2010). Projects using citizen science have noted that data can have unequal coverage of geographic areas, which is problematic when attempting to determine abundance and presence-absence (Bois *et al.* 2011). In addition, taxa and locality-specific variation in data collection can lead to sampling bias (Dickinson *et al.* 2010).

Despite these concerns, studies have repeatedly found that with limited amounts of training, non-professional observers can be nearly as effective as professionals (Crall *et al.* 2010; Bois *et al.* 2011; Gallo and Waitt 2011). Moreover, data quality for citizen-science projects is greatly improved when data collection is standardized and validated by experts, and when volunteers receive feedback on their contributions (Silvertown 2009). Social interaction, enjoying the outdoors, and frequent communication (including calls to action) are effective incentives to keep long-term citizen-science projects active, and have the added benefit of increasing interest in local conservation (Scyphers *et al.* 2015). With proper protocols and execution, citizen science can provide the personnel and large-scale data collection necessary for early invasive species detection, as well as stimulate community involvement and appreciation for scientific research.

■ Government agency monitoring

Government agencies have a major role in the surveillance of invasive species globally (Early *et al.* 2016), with monitoring or early detection programs implemented from local to federal scales. However, government agencies also conduct monitoring and research for many other purposes, including conserving biodiversity, identifying indicators of environmental quality, or implementing a census of commercially valuable habitats like forests (Marsh and Trenham 2008; Kuehne *et al.* 2017). These monitoring programs can serve as “passive surveillance” for invasive species, as was the case when divers performing a routine survey of eelgrass beds inadvertently discovered the invasive seaweed *Caulerpa taxifolia* in California (Anderson 2005).

An effective management response to the early detection of an invasive species population will require coordination and communication within and between agencies (Vander Zanden

et al. 2010). If a new invasion is discovered through passive surveillance, mechanisms must be in place to ensure that news of the discovery is communicated to managers within the same agency, at a different agency, or to different governments that may share jurisdiction over an ecosystem. Communication within and between government agencies can be improved through the digitization of data and the creation of shared databases (WebTable 1). Developing standardized methods for data collection and storage is therefore critical for improving data sharing and communication among agencies (NISC 2001; Graham *et al.* 2007; Simpson *et al.* 2009).

Finally, government agencies are primarily responsible for acting on the discovery of new biological invasions through management responses that may include containment or eradication of the population (Liebhold *et al.* 2016). Surveillance of biological invasions is only valuable if it is linked to management responses that can be rapidly mobilized to prevent invasive species establishment, spread, or impacts (Vander Zanden *et al.* 2010). We have summarized emerging technologies that may improve surveillance and early detection of biological invasions, but government agencies must be proactive in assimilating and acting on this increasing volume of information.

■ Conclusions

Global networks for the detection of invasive species are needed if prevention strategies are going to be effective (Packer *et al.* 2017). Moreover, the effectiveness of these networks may be maximized when multiple strategies are combined synergistically (Figure 5). For example, citizen science can be combined with other tools to promote the emergence of “technoecology” (the use of cutting-edge physical technology to acquire new volumes and forms of ecology data as per Allan *et al.* [2018]): camera trapping could be scaled up to a global interconnected network for early detection of invasive species (Steenweg *et al.* 2017), and the large number of associated photographs crowdsourced to help identify taxa (McShea *et al.* 2016); and RS data used to track changes in phenology can be verified on the ground with citizen-science programs, helping to merge local- and continental-scale information and increasing overall resolution (Elmore *et al.* 2016). Even technologically complex approaches (eg chemical lures, eDNA) can be combined effectively with citizen science (Larson *et al.* 2017).

One example of effective synergy across early detection methods is provided by Rullan-Silva *et al.* (2013), in which the authors proposed the development of a nationwide monitoring system that combines time-series with moderate spatial resolution but high temporal resolution (such as MODIS data) with approaches that have higher spatial resolution (such as Landsat). Coordinating public and private partners to form rapid response initiatives for early detection, to prevent establishment and spread, and to aid in eradication efforts should also be a priority. For example, the US National Early Detection and Rapid Response System for Invasive Plants is a collaboration of the US Geological Survey, the Federal Interagency

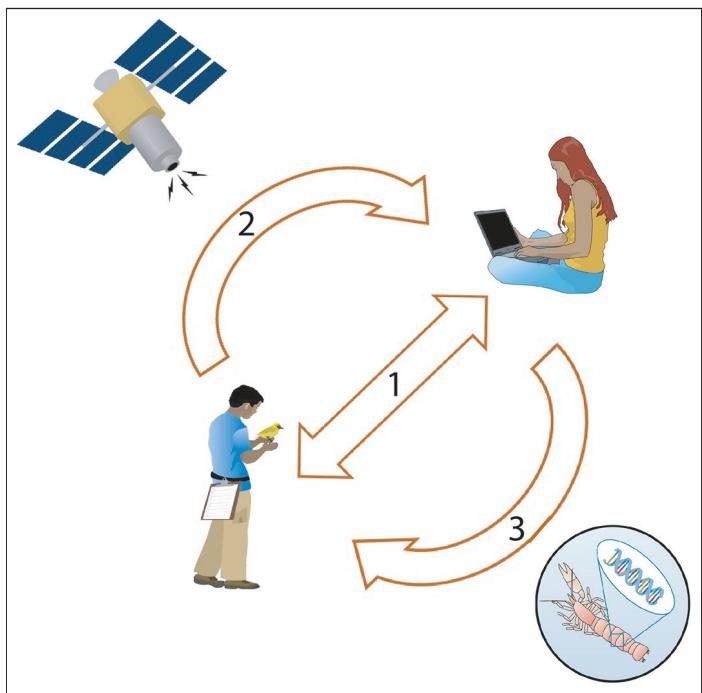


Figure 5. Some potential synergies between emerging approaches for the surveillance and early monitoring of invasive species. Government agency scientists and resource managers may recruit interested citizen scientists (1) to participate in invasive species surveillance, and share their results with these agencies through smartphone apps or online databases. Furthermore, government agencies could use remote-sensing data (2) to prioritize sites for citizen-scientist surveillance of invaders detectable by these technologies, in order to ground-truth remote observations. Finally, some emerging approaches to invasive species surveillance like trapping for invaders with chemical lures or eDNA (3) are easy to implement in the field but require technical expertise in the lab to confirm organism identifications or process samples. Citizen scientists could collect eDNA samples or run and return networks of chemical lures to government agency partners for processing and identification. Symbols from the Integration and Application Network (<https://ian.umces.edu/symbols>).

Committee for the Management of Noxious and Exotic Weeds, the Invasive Plant Atlas of New England project, and additional regional and local organizations (Westbrooks 2004).

Despite the potential benefits of the emerging technologies described above, limitations remain (Table 1). However, combining various approaches may allow for increased accuracy of detection. For example, large-scale remote-sensing technologies could be used to identify areas vulnerable to invasion, to guide more specific surveillance through chemical lures or eDNA. In the face of concerns about citizen-scientist participation, experts should validate citizen-scientist contributions whenever feasible, especially when the presence of an invasive species is indicated. A pipeline of data filtering can be established to maximize efficiency and accuracy of data analysis when images and samples are being catalogued by citizen scientists for invasive species detection. Finally, we recommend that private organizations and government agencies engage in public outreach and citizen-scientist recruitment for the most effective implementation of these technologies, and to increase public awareness and concern for the threats posed by invasive species.

Many of the detection and monitoring methods described here provide an additional benefit of being non-invasive approaches, which is ideal for systems containing imperiled species threatened by biological invaders (Barnes and Turner 2015). They can also be employed in systems where conventional surveillance methods are constrained by factors such as site accessibility or otherwise hazardous sampling conditions (Larson *et al.* 2017). In addition, each of these tools allows for increased efficiency of surveillance while at the same time reducing costs for monitoring once infrastructure (including data processing pipelines) is established. Citizen scientists, private organizations, and government agencies should partner and coordinate the use of these tools for effective early detection of invasive species. Data sharing on universal platforms will aid this effort and increase efficiency of action once invasions are detected. Ultimately, with heightened sensitivity, increased efficiency, and lower costs, early

Table 1. Several potential strengths and weaknesses of emerging methods for the surveillance of invasive species

Surveillance method	Strengths	Weaknesses
Chemical cues	Can detect single or multiple species; databases of insect attractants available; less labor-intensive than manual searches	Distance and duration of detection can be uncertain; less research and information on non-insect taxa; development and testing of new chemical attractants costly
Citizen science	Facilitated by technology (eg smartphones); many potential observers over large areas; opportunities for education and outreach	Access to technology and standardization limiting; concerns over identification accuracy for some taxa; unequal coverage of geographic areas, taxa
Environmental DNA	Can be highly sensitive to species presence; increasingly affordable, improving technology; field sampling simple and cost effective	Risk of false positives from contamination, DNA transport; standardization of best practices still needed; better developed at present for aquatic than terrestrial systems
Government agencies	Can use established monitoring programs; expertise in identification, natural history; legal mandate and associated resources	Challenges in data sharing between agencies, jurisdictions; not enough personnel for all taxa, regions; occasionally slow to adopt new technologies, approaches
Remote sensing	Broad geographic and temporal coverage; local monitoring (camera traps, acoustic); regional monitoring (aerial, satellite)	Difficult to detect new invasions early at low abundances; some imagery (eg hyperspectral) expensive; requires technical expertise (data processing, sensor operation)

detection of invasive species will become more feasible, increasing the likelihood of control or eradication.

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■ Supporting Information

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