international BARCODE OF LIFE

Barcode BULLETIN

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Vol.6, Issue 1

School Butterfly Project: Raising Environmental Awareness while Answering Pressing Questions

Applications
Seafood Barcoding Leads to Government Action

Research
Building a Comprehensive Library for Beetles

Welcome to the March 2015 issue of the iBOL Barcode Bulletin.

Only five more months left before the 6th International Barcode of Life Conference will open its doors in Guelph. Sarah Adamowicz, one of the authors in this Bulletin issue, describes the conference theme *Barcodes to Biomes* as a signal of "the ongoing expansion of our community's research agenda from studies of particular sets of species in particular places to examinations of entire biotic assemblies at local and global scales."

This issue of the Barcode Bulletin once again features a wide assortment of articles on DNA barcoding research and applications, with topics ranging from the different uses of dietary analysis to building comprehensive reference libraries for certain groups of organisms. It always amazes me how relatively easy it is for us each time to find so many interesting and exciting stories for this newsletter.

We hope you enjoy reading this first issue of 2015.

Dirk Steinke
Editor-in-chief

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News Briefs

The Academy of Finland has approved an infrastructure application for the Finnish Biodiversity Information Facility. This project, coordinated by the Finnish Museum of Natural History, will focus on the digitization of collections, with 286,000 € dedicated to the advancement of barcoding in Finland over the next two and a half years.

ACADEMY OF FINLAND

SwissBOL is pleased to announce that they have secured support for their activities, including a citizen science project, for the next two years through the Federal Office for the Environment and a private foundation.

The Biodiversity Institute of Ontario was one of nine research facilities to recently receive funding through the Canada Foundation for Innovation's Major Science Initiatives (MSI) program. The award of $2.15 million CAD, which covers infrastructure and operating costs, recognizes the national and international contributions of this "big science" enterprise.

Associate professor Smitha Hegde of St. Aloysius College, Mangalore, India, was awarded the Prof SS Bir gold medal by the Indian Fern Society for her work in pteridology. In addition, at the National Symposium "Biotechnology and Molecular Biology for Industry and the Common Man", her Ph.D. student Shaiesh Morajkar presented DNA barcoding results for the fern species *Pteris vittata* and received the Prof. Brij Mohan Johri Award for best poster.

The School Butterfly Project Malaysia

Written by: Shi-Wei JISMING-SEE, Guo-Jie BRANDON-MONG, Yu-Zun GUO, Ping-Shin LEE, Kong-Wah SING, and John-James WILSON (Museum of Zoology, University of Malaya)

Malaysia is a megadiverse country suffering from rapid biodiversity loss, yet moves to address local conservation issues are hindered by a lack of public awareness. The School Butterfly Project began in September 2014 with the goals of increasing awareness of biodiversity and instilling a sense of stewardship for local wildlife among schoolchildren. Butterflies were chosen as conservation ambassadors due to their charisma and familiarity, but also because they are good indicators of environmental conditions (see Barcode Bulletin 4(2): 9).

Citizen science takes flight while promoting outdoor activities, conservation and environmental awareness.

Five schools in five different states of Peninsular Malaysia (Semenanjung Malaysia) were approached to take part, with about 30 schoolchildren (aged 9-12 years) from each school becoming involved in the project. A sixth group included homeschooled children from the Kuala Lumpur area.

In the first stage of the project we visited each school for an interactive training day. The training sessions covered butterfly sampling, including one of the schoolchildren's favourite activities, making a homemade butterfly net, and how to take non-lethal butterfly DNA samples (legs). We also covered butterfly classification and identification using DNA barcodes. We introduced scientific thinking during the sessions, building up to the research question: "How will butterflies (in Peninsular Malaysia) be affected by global warming?"

Following the training sessions, the schoolchildren were provided with a sampling kit (forceps, tubes, marker pens and camera) and tasked with collecting butterfly samples in their schoolyards or local parks four times over the next 12 months. The first sampling day took place in October 2014 and collectively the schoolchildren sampled around 120 butterfly legs. The legs were sent to the Museum of Zoology, University of Malaya for DNA barcoding, which revealed that the schoolchildren had sampled 40 species.

"The students had sampled 40 species... an impressive start by the young citizen scientists."

There are around 1100 butterfly species known from Peninsular Malaysia, but we feel this is an impressive start by the young citizen scientists. Several of the dominant species sampled (the Striped Wanderer, *Appias olferna*, and the Tawny Coster, *Acraea terpsicore*) are relative newcomers to Peninsular Malaysia. This interesting find may help the schoolchildren generate hypotheses around the project's research question.

To the best of our knowledge, the School Butterfly Project is the first project of its kind for Malaysia and has generated positive interest from the local media. The combination of butterflies, fun outdoor activities, conservation and environmental awareness, which have intersected with the schools' curriculum (PCR, molecular biology and taxonomy) seems to have been a great success.

In the short term we look forward to the next sampling day in March 2015 and discovering if, and how, butterfly diversity changes throughout the year in the "aseasonal" tropics. In the long term we look forward to exploring ways to expand the reach of the project across the country and society. For more information and to follow the progress of the School Butterfly Project you can like our Facebook page: facebook.com/SchoolButterflyProject

Image credits: School Butterfly Project team

Image credit: Philippe Bordes

Analyzing Marine Predator Diets of the Iroise Sea

Written by: Jean-Luc JUNG (Laboratoire BioGeMME, Université de Brest) and Sami HASSANI (LEMM Océanopolis)

Deciphering complex marine food webs is one of the major topics of study in marine research. Food web analyses can detect and highlight environmental variations, whether they are caused by anthropogenic activities or otherwise, as well as contribute to fundamental ecological understanding. Conservation concerns also require detailed knowledge of predator diet, in particular when an overlap between diet and commercial resources is suspected.

The Iroise Sea is located in the northwest of France and is surrounded by the Bay of Biscay at the south and by the Celtic Sea and the English Channel at the west and north. The Iroise Sea hosts a surprisingly rich marine biodiversity, including emblematic and flagship marine mammal species. Their biodiversity is monitored primarily by a stranding network and the successful application of the DNA barcode approach (Alfonsi *et al.* 2013, DOI: 10.3897/zookeys.365.5873). Two species are particularly interesting: the harbour porpoise (*Phocoena phocoena*) and the grey seal (*Halichoerus grypus*). The grey seal is distributed in the North Atlantic temperate to sub polar waters. The Molène archipelago is its southernmost location in Europe, making this particular settlement outstanding. The harbour porpoise has clearly made a global southward shift for some years in the Northeast Atlantic, including a comeback along the French Atlantic coast starting in the 1990s.

A suspected overlap between traditional fishing and the diet of two marine predators prompted conservation concerns.

The Iroise Sea is a UNESCO Biosphere reserve because of its natural richness. This led to the creation of the first French Marine Park in 2007, named "Parc Naturel Marin d'Iroise". One of the goals of the Iroise Marine Park is to protect and maintain the rich biodiversity of the area, but also to preserve relevant and sustainable human activities, in particular, traditional fishing targeting sea bass, Atlantic pollock, European pilchard, monkfish, and crabs.

In this context, the interactions of marine mammal species with fisheries need to be identified, specifically in terms of competition for fish resources, marine mammal by-catch, and depredation.

Through a multidisciplinary project led by Iroise Marine Park that joined scientists and fishermen, we undertook a study of the diets of grey seals and harbour porpoises, using animals by-caught or stranded in the Iroise Sea. While ingested prey is conventionally identified by its hard remains (otoliths for fishes and beaks for cephalopods), the last decade has seen the use of molecular approaches that aim to identify remaining prey DNA, particularly in cases where only soft remains of prey are found in the stomach contents (e.g. when seals eat fish bodies without the head). Some of these approaches, which are highly informative but still expensive and challenging to implement, make use of NGS to identify multiple prey DNA in feces.

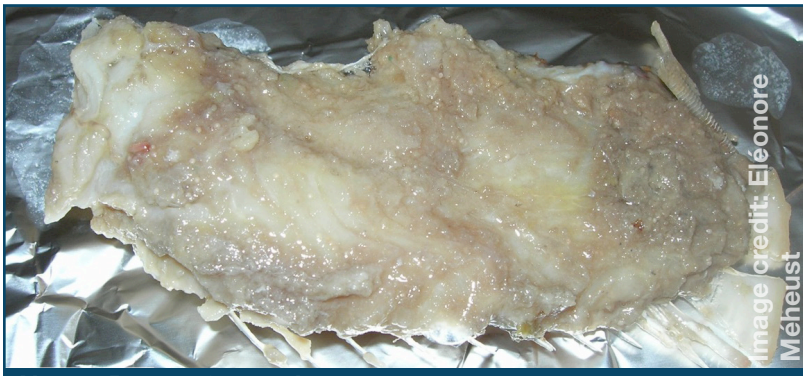
With DNA barcoding, "the number of prey item species identified per predator increased by nearly a third for the grey seal..."

We tested whether the diet analysis of grey seal and harbour porpoise could be improved by using a simple, easy to use and economic method based on DNA barcoding. We studied the stomach contents of 11 grey seals and seven harbour porpoises using two complementary methods: the classical method of observation and taxonomic identification of prey hard remains and a DNA-based identification of soft tissues. We identified nine fish species thanks to this parallel use of a classical taxonomic determination and a simple DNA barcoding approach. The number of prey item species identified per predator increased by nearly a third for the grey seal and by 21% for the harbour porpoise through the use of DNA barcoding. All of the DNA sequence data obtained are publicly accessible on BOLD in the project *Identification of diet of marine mammals [DBMM]*.

The results of our study suggest that grey seals and harbour porpoises in the Iroise sea do not seem to target the most important species for fishermen, with the exception of the sea bass identified in one grey seal. Therefore, only limited competition seems to exist for this natural resource between fishermen and grey seals in the Iroise Sea.

Once again the DNA barcoding approach has been used successfully. Because sampling the stomach contents of marine mammals that are found dead is highly opportunistic and occurs only infrequently, it is necessary that their analysis be as complete as possible. As shown by our study, the number of prey species identified is optimized by using both visual and DNA-based identification and we therefore believe that DNA barcoding should routinely be used for diet analysis of marine top predators.

This study is part of a multidisciplinary project named INPECMAM which brings together the Iroise Marine Park, Océanopolis, BioGeMME, Le Muséum National d'Histoire Naturelle, and le comité départemental des pêches du Finistère. The experimental work presented in this article was mainly performed by Eléonore Méheust. For more information about the results, see DOI: 10.1080/17451000.2014.943240



Above: Prey soft remains.

Below: Otoliths of European pollack (*Polachius polachius*), blue whiting (*Micromesistius poutassou*), and European hake (*Merluccius merluccius*).

Sixth International Barcode of Life Conference

Written by: Sarah Adamowicz, on behalf of the Operating Committee for the 6th International Barcode of Life Conference

The Scientific Organizing Committee is delighted to announce that the 6th International Barcode of Life Conference will be held at the University of Guelph, Canada, from August 18-21, 2015. This biannual Conference has grown into a major international meeting, expected to attract 500 to 600 participants from dozens of nations.

"...showcase the latest scientific achievements and socio-economic implications of work conducted by the DNA barcode research community."

The 6th Conference will build upon a rich tradition of meetings that have been held around the world: London (2005), Taipei (2007), Mexico City (2009), Adelaide (2011), and Kunming (2013). The meeting will showcase the latest scientific achievements and socio-economic implications of work conducted by the DNA barcode research community. The theme of the 6th Conference, *Barcodes to Biomes*, signals the ongoing expansion of our community's research agenda from studies of particular sets of species in particular places to examinations of entire biotic assemblies at local and global scales.

This conference will feature a shared set of experiences through morning plenary sessions as well as coffee breaks and meals, to promote discussion of breakthroughs, ideas, and proposed collaborations among all delegates. An exciting international line-up of 30 plenary speakers will share their insights in ecology, evolution, bioinformatics, systematics, global biodiversity initiatives, and conservation. This breadth of topics reflects the ongoing adoption of DNA barcoding and biodiversity genomics approaches in diverse disciplines within the biological sciences.

The research interests of the plenary speakers are now being highlighted via regular blog postings available through our conference website (dnabarcodes2015.org). Our first highlighted speaker was Nancy Knowlton. Many members of our community have been inspired by Dr. Knowlton's work on cryptic species in the sea and molecular clock calibrations employing the Isthmus of Panama. Her recent studies are exploring marine biodiversity in fragile reef ecosystems.

See the conference site for more details and for regular updates from the plenary speakers.

"...delegates will be able to focus on science and networking."

The scientific program will also feature contributed oral and poster presentations. Topics for the parallel sessions range from ecology, evolution, and systematics to socio-economic applications of DNA barcoding—such as food security, management of invasive species, and implications for governmental policy.

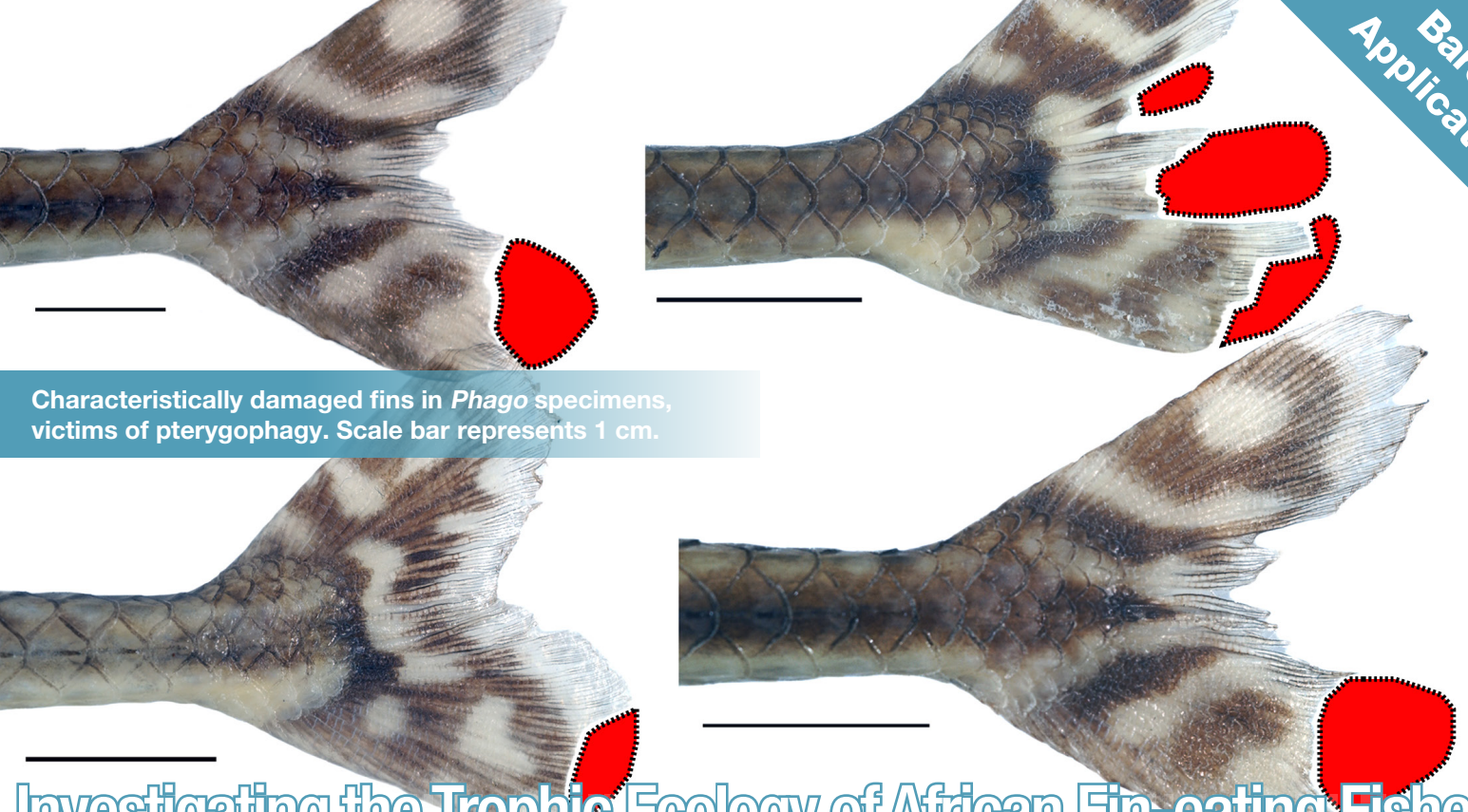
Broad international participation will be facilitated by travel awards, which are applied for at the time of abstract submission. Moreover, the conference will recognize and encourage the careers of emerging researchers by awarding prizes for both graduate students and post-doctoral fellows.

All abstracts selected for oral and poster presentation will be published in a special abstracts issue of the journal *Genome*, facilitating accessibility of research results and citations for conference participants.

The University of Guelph is one of Canada's major universities for the life sciences. The city of Guelph is easily accessible by air, as it is located just one hour drive from Pearson International Airport in Toronto. The major conference venues and accommodations are all located within a short walk of one another, ensuring that delegates will be able to focus on science and networking.

The Conference Committee has also organized several exciting pre-conference workshops as well as post-conference excursions. We hope that you will stay beyond the meeting to enjoy the natural and cultural treasures of our corner of the planet, as well as to further your networking opportunities. Please visit the conference website for further information about the scientific program as well as details regarding registration, abstract submission, travel logistics, child care options, and local activities: dnabarcodes2015.org.

We look forward to welcoming you and wish all delegates a productive and inspiring meeting.



Characteristically damaged fins in *Phago* specimens, victims of pterygophagy. Scale bar represents 1 cm.

Photo © Melanie Stiassny

Investigating the Trophic Ecology of African Fin-eating Fishes

Written by: Jairo Arroyave (American Museum of Natural History)

The freshwaters of much of sub-Saharan Africa and the Nile River basin harbor a moderately diverse group of fishes — the family Distichodontidae — whose members include four genera specialized in an unusual feeding strategy: pterygophagy, meaning that they feed primarily on fish fins. Distichodontids are one of very few lineages of teleost fishes known to exhibit this highly specialized feeding strategy.

Very little is known about the evolution and ecology of pterygophagy.

Although pterygophagy in distichodontid fishes was first documented more than 50 years ago, very little is known about the evolution and ecology of this bizarre behavior, including basic information on prey selection and diet composition. This is unfortunate because dietary information is critical for an understanding of a variety of ecological processes and can also inform conservation efforts for endangered species and/or threatened ecosystems.

In one of the very few studies investigating fin-eating behaviors in distichodontid fishes, traditional stomach content analysis revealed striking similarities in caudal-fin coloration and patterning between predators and their putative prey, which led the author to hypothesize that the striking caudal-fin patterning of pterygophagous distichodontids reflects a form of aggressive mimicry, allowing them to avoid detection by their allegedly monospecific prey.

To test this hypothesis and further investigate prey selection and diet composition in these poorly studied fish, ichthyology curator Melanie Stiassny (American Museum of Natural History) and I set up a mentoring project for a team of New York City (NYC) high school students participating in the Urban Barcode Research Program (UBRP) of Cold Spring Harbor Laboratory's DNA Learning Center (DNALC) in which DNA barcoding was used to identify prey species from fin fragments found in the stomachs of three genera of pterygophagous distichodontids from the Congo River basin (i.e., *Phago*, *Eugnathichthys*, and *Ichthyborus*).

The UBRP is a mentoring program designed to engage high school students to study biodiversity in NYC using DNA technology, in particular DNA barcoding. Projects in this program include biodiversity assessments, investigation of food mislabeling/fraud, and wildlife DNA forensics, among many others. This project, the first UBRP study investigating trophic ecology in tropical organisms, was received with enthusiasm and interest by both students and educators in the program.

DNA barcoding of fin fragments from stomachs of distichodontid fishes did "not support the hypothesis of aggressive mimicry."

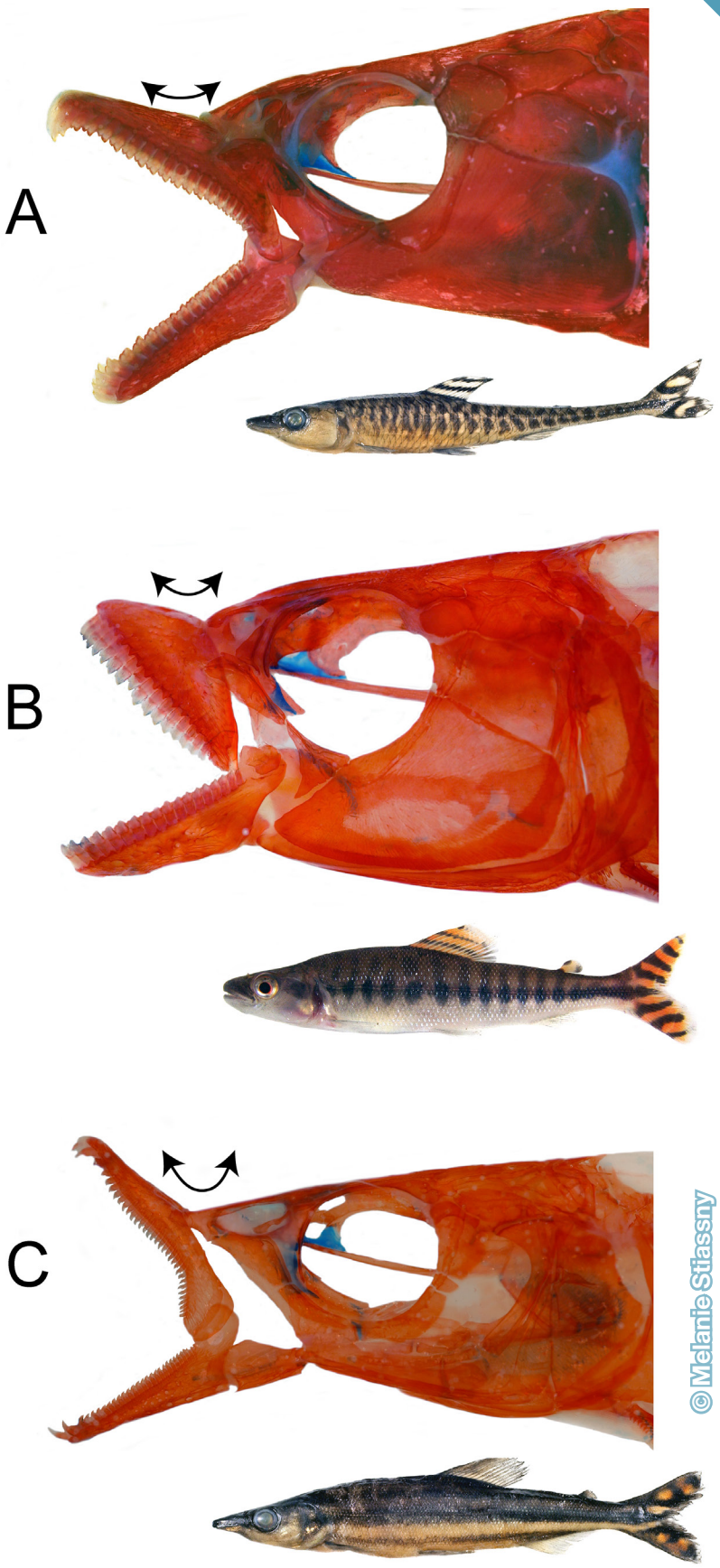
Our findings, recently published in the journal *Ecology and Evolution*, do not support the hypothesis of aggressive mimicry suggesting instead that, despite the possession of highly specialized trophic anatomies, fin-eating distichodontids are opportunistic generalists, preying on fishes from a wide phylogenetic spectrum and to the extent of engaging in cannibalism. While cannibalism in fishes is not uncommon, virtually all previously known instances involve adults eating their own offspring not parts of their own kind.

Although pterygophagous distichodontids are highly specialized in terms of how and what they eat (fins), our findings indicate that they are in fact "specialized generalists", able to snatch the fins from just about any species of fish they come across, and are therefore probably resilient to environmental change as a result—an idea supported by the discovery that even a recently introduced species, the African arowana (*Heterotis niloticus*), is among their prey.

This study demonstrates how DNA barcoding can be used to shed light on evolutionary and ecological aspects of highly specialized ectoparasitic fin-eating behaviors by enabling the identification of prey species from small pieces of fins found in fish stomachs.

For more information about the results discussed in this article, see DOI: 10.1002/ece3.1321

Variation in jaw anatomy in pterygophagous distichodontids represented in our study by the genera *Phago* (A), *Eugnathichthys* (B), and *Ichthyborus* (C).



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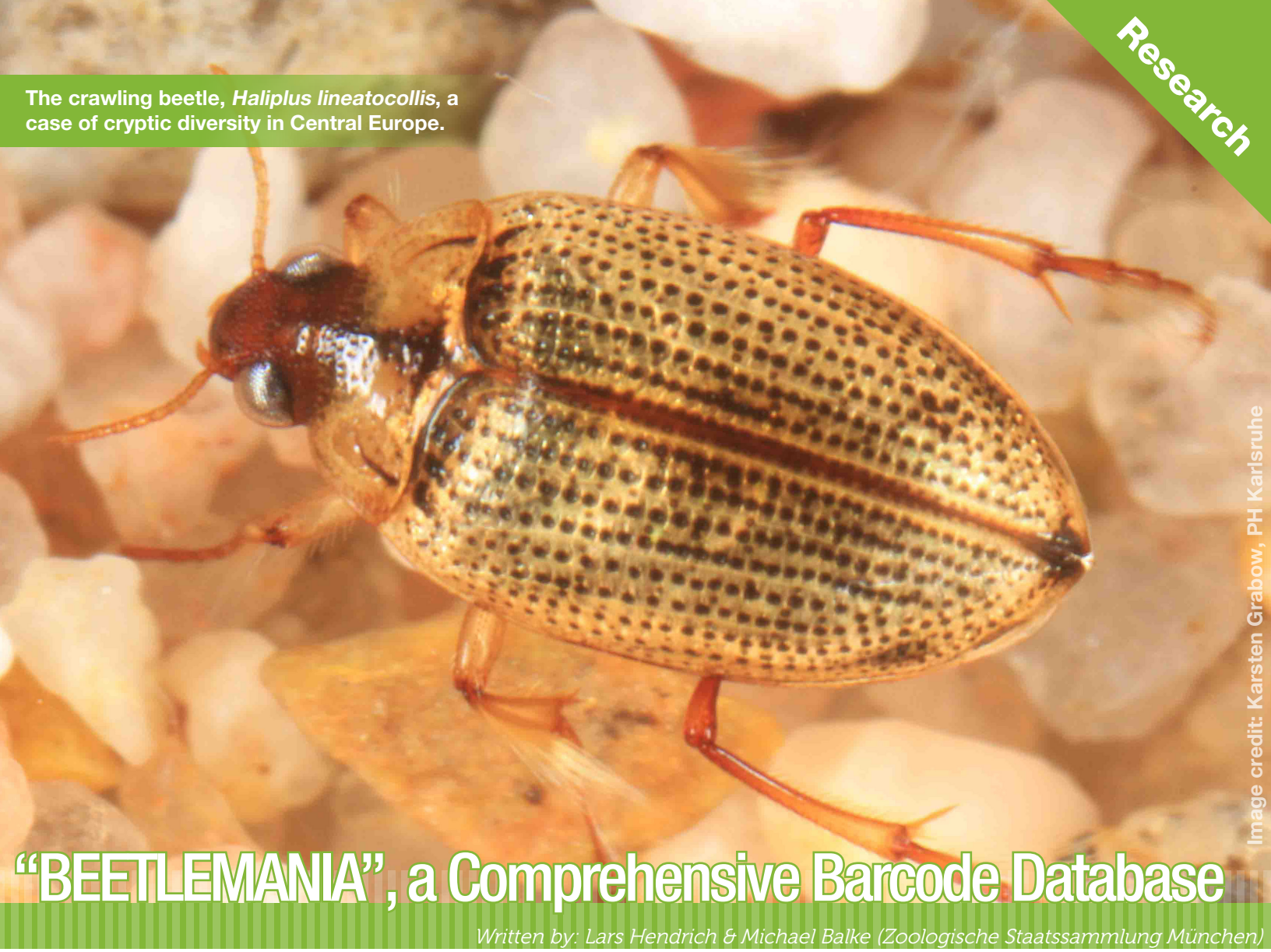


The crawling beetle, *Haliplus lineatocollis*, a case of cryptic diversity in Central Europe.

Image credit: Karsten Grabow, PH Karlsruhe

"BEETLEMANIA", a Comprehensive Barcode Database

Written by: Lars Hendrich & Michael Balke (Zoologische Staatssammlung München)

Beetles are the most diverse group of animals and crucial for ecosystem functioning. In many countries, they are intensively used for environmental impact assessments. However, even within the well-studied Central European fauna, species identification can be very difficult. A comprehensive and taxonomically well curated DNA barcode library can remedy this deficit and also link hundreds of years of traditional knowledge with modern next generation sequencing technology.

In five years, more than half a million specimens of beetles were collected by a network of taxonomists and citizen scientists from all parts of Germany using various methods (i.e. hand collecting, sweep-netting, Malaise traps, window traps and pitfall traps) which were deployed in varied aquatic and terrestrial habitats.

Of these beetles, 462,550 specimens were sorted and identified to a species level and a subset of more than 25,000 specimens was subsequently selected for DNA barcode analysis.

With 3,514 well-identified species, our study generated the world's largest Coleoptera DNA barcode reference library. It contains representatives from 97 of 103 Central European families. Approximately 42% of the Central European fauna of beetles and 53% of the species known from Germany are now represented in the publicly available reference library on BOLD.

"In five years, more than half a million specimens of beetles were collected..."

Although the majority of the specimens (92.2%) could be unambiguously identified using barcodes, 1,089 specimens (6.8%) were assigned to more than one Barcode Index Number (BIN), creating 395 BINs which need further study to ascertain if they represent cryptic species, cases of mitochondrial introgression, or simply regional variation within widespread species.

We found 409 specimens (2.6%) that shared a BIN assignment with another species, mostly involving a pair of closely allied species. Most of these taxa were separated by barcodes although sequence divergences were low. Only 155 specimens (0.97%) showed identical or overlapping clusters.

The relatively high number of 1,089 specimens representing 176 potentially overlooked species was surprising, as active beetle research in Germany goes back more than 200 years. It is also noteworthy that these tricky cases did not only involve highly diverse and taxonomically difficult groups such as the rove beetles, weevils and click beetles but also the well-studied longhorn beetles, crawling water beetles, clown beetles and scavenger beetles.

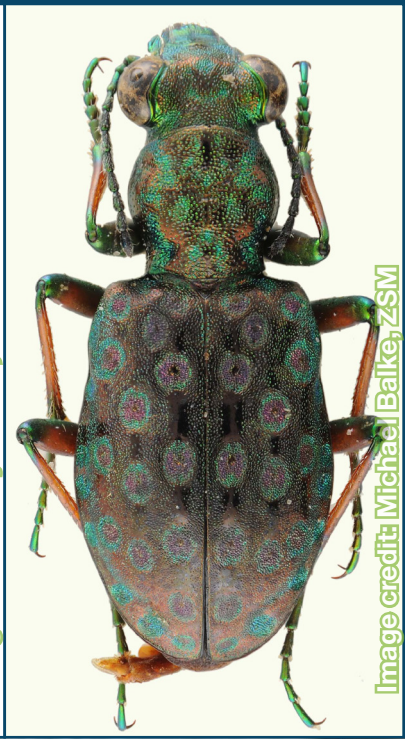
The study uncovered 176 potentially overlooked species, some of which belonged to well-studied groups.

We encourage the coleopteran scientific community to join the DNA barcoding projects on BOLD as registered users in order to participate in discussions and to comment on particular species, effectively aiding in the clarification of the status of these species and assisting in the description of possibly overlooked species.

For more information about the results discussed in this article, see DOI: 10.1111/1755-0998.12354

Below, on left: *Emus hirtus* is one of the largest rove beetles in Europe and, in Great Britain, it is called "Maid of Kent".

Right: *Elaphrus ullrichi*, a rare ground beetle that lives at the shores of natural mountain rivers.



Seafood sold in a marketplace in Florianópolis City. The species sold as Linguado (Flounder) was frequently mislabeled.

Governmental Seafood Inspection Program Employs Barcoding

Written by and images by: Daniel C. Carvalho (Pontifícia Universidade Católica de Minas Gerais)

Although DNA barcoding has revealed numerous cases of seafood mislabeling worldwide, this evidence has not yet resulted in legal action nor financial penalties for food business operators. However, a partnership between governmental agencies, a university and a biotech company enabled the implementation of a seafood inspection program based on DNA barcoding in Brazil. This program aims to detect illegal mislabeling and to penalize cases of substitutions in order to discourage seafood fraud in the Brazilian market.

The first round of seafood inspection conducted by the governmental regulatory agencies of Florianópolis, Myleus Biotechnology® (www.myleus.com) and PUC Minas University analyzed thirty fish products confiscated by officials from market places in the city of Florianópolis. Seafood was randomly chosen from fishmongers, supermarkets and restaurants and comprised of commercially important species such as cod, flounder, grouper, tuna, pink cusk-eel, hake, shark, white fish and dolphinfish. Samples were sent to Myleus Biotechnology® to be analyzed using standard DNA barcode protocols.

DNA barcoding "was able to detect eight cases (24%) of species mislabeling within highly priced species..."

DNA barcoding identified all seafood to the species level and was able to detect eight cases (24%) of species mislabeling within highly priced species such as cod, pink cusk-eel and flounder products. For instance, *Gadus morhua* cod was replaced by Ling (*Molva molva*) and Alaska pollack (*Gadus chalcogrammus*) in three supermarkets, pink cusk-eel (*Genypterus blacodes*) was substituted for Alaska pollack and flounder breaded fillet was replaced by basa (*Pangasius hypophthalmus*) in seafood dishes confiscated from restaurants. Two endangered species sold under the generic names shark and grouper were detected (*Sphyrna lewini* and *Epinephelus marginatus*).

All of the mislabeling cases detected by the governmental program of seafood inspection resulted in legal action and financial penalties for the respective restaurant, supermarket owners and fishmongers.

Governmental agencies in Brazil are now aware of the prevalence of seafood mislabeling as well as the potential of a DNA-based identification method as part of inspection programs. Currently, the NGO Proteste (http://www.proteste.org.br) is conducting a national survey in partnership with Myleus Biotechnology® in order to get a broader picture of the extent of seafood substitution in Brazil. A necessary next step to discourage any seafood substitution will be the implementation of additional regulatory programs by governmental agencies to be able to consistently penalize market fraud.

"Governmental agencies in Brazil are now aware of... the potential of a DNA-based identification method as part of inspection programs."

For more information about the results discussed in this article, see DOI: 10.1016/j.foodcont.2014.10.025

Officials from the governmental regulatory agency (PROCON) confiscating seafood at a restaurant in the City of Florianópolis.

Assassin bug (Reduviidae, which include vectors of Chagas' disease), collected and photographed in Puerto Rico.

Barcoding the Deadliest Animals on Earth

Written by: Sean Locke (Universidad de Puerto Rico, Mayagüez)

Parasites and their vectors are usually small and taxonomically challenging, and some have enormous importance to human health. This would seem like an area where DNA barcoding could lead to practical and important contributions. Indeed, just after it was proposed in 2003, the potential benefits of barcoding for medical parasitology and entomology were pointed out by Nora Besansky and colleagues at the University of Notre Dame.

However, it is not clear if this call to action was heard. While papers with barcodes from hundreds of species of birds, fish or butterflies now seem to come out almost weekly, how are we doing on parasites and vectors? Danielle Ondrejicka, I and others decided to find out.

Our first hurdle came as a surprise: no comprehensive parasite and vector checklist was available to gauge barcoding coverage. So we compiled one, 1403 species long. As of last July, barcodes existed for 597 of these species. That's 43%, which at first glance might seem like a failing grade. With more than 10,000 records added to BOLD weekly, why such slow progress in agents of disease and death?

Barcodes existed for 597 of 1403 parasite and vector species, "which at first glance might seem like a failing grade."

Simply put, building a parasite and vector barcode library is really hard. Some obstacles stem from the deep diversity among organisms with parasitic lifestyles. Human health is affected by familiar, cosmopolitan arthropods, microscopic worms, intracellular protozoans, and many others. Major players on the list lack mitochondria. Think of it this way: *18 phyla*.

The ability to collect, preserve and identify these organisms is probably institutional in scale (e.g., CDC), and beyond the capacity of an individual or even team of researchers. In that light, and particularly given the absence of an active, dedicated campaign, 43% coverage is actually not bad (we argue it's better than a random sampling of animal species). And, as we also found out, it is improving rapidly.

We reviewed 83 studies where barcoding was used to discriminate species of parasites and vectors of medical importance. Forty percent of them appeared in the 18 months preceding our search – a clear upsurge is underway, particularly in arthropod vectors.

These studies spanned the globe and applied DNA barcoding to diverse ends, including identification of blood-meal sources in vectors, descriptions of new species, and medical diagnoses. Most included some form of evaluation of the barcode as an identification tool, and barcode-based results were in agreement with authors' taxonomic conclusions in 94-95% of cases.

"We reviewed 83 studies... These studies spanned the globe and applied DNA barcoding to diverse ends..."

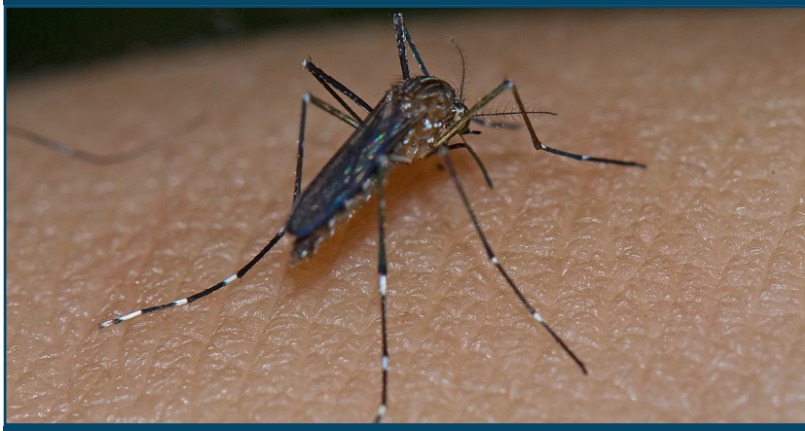
There was little or no effect of study scope (specimens or species sampled), number of other molecular markers analyzed, or whether morphological analysis was reported. Not a failing grade, in other words, but an A+.

Still, the review also revealed some problems. Most studies mentioned no vouchers, and to some, "DNA barcoding" seems to mean sequencing any convenient marker. We hope our study clarifies some of these issues, and particularly that it stimulates further work in parasites and vectors, to bring a day when specimen vouchers, images, and sequences exist for all animal species implicated in parasitic diseases of humans.

For more information about the results discussed in this article, see DOI: 10.1016/j.pt.2014.09.003

Image credits: José R. Almodóvar, Universidad de Puerto Rico, Mayagüez

Tse tse fly (*Glossina* sp., vector of sleeping sickness) collected in Tanzania by Lucy Bunkley-Williams and Ernest H. Williiams.



Above: Yellow fever mosquito (*Aedes aegypti*), dengue vector native to Africa.

Below: Louse (*Pediculus humanus*)

Image credit: Elizabeth Sears

Barcoding the Vascular Plants of Canada

Written by: Stephanie deWaard and Allison Brown (Biodiversity Institute of Ontario)

As part of the International Barcode of Life project and in collaboration with researchers across Canada, the Biodiversity Institute of Ontario (BIO) is striving to DNA barcode all Canadian vascular plant species. According to the Database of Vascular Plants of Canada (VASCAN), there are 5582 plant species recorded for Canada. All of our efforts leading up to last fall have resulted in barcode coverage of 4377 of these species (78%).

The quest to barcode the remaining 1205 species by the summer of 2015 began by first locating valuable material in herbaria across Canada. This was done using Canadensys, a digital network which provides access to published databases for several participating university, botanical garden and museum collections.

We located 606 of the 1205 species and an additional 412 of these were identified as hybrids. Many of these hybrids do not commonly occur in nature and we are choosing to omit these from the final recovery efforts.

Our most efficient strategy for gaining coverage of the last 22% of Canadian vascular plant species is to access voucher specimens housed in some of our most reputable herbaria across Canada, especially in winter when access to live specimens is limited and collecting efforts would be onerous.

"The team made impressive progress, sampling 1180 specimens and 497 species in fifteen days."

With our needed species list in hand, staff members from BIO (Connor Warne, Maryam Fatahi, and Stephanie deWaard) made two visits to the Canadian Museum of Nature's National Herbarium of Canada (CAN) in September and December 2014. CAN is a national treasure with over 575,000 vascular plant specimens, including about 2500 type specimens, and curator Jennifer Doubt and research scientist Jeff Saarela helped oversee and facilitate our efforts to barcode these remaining species.

Our first trip targeted specimens of known species represented in the collection, and processing included databasing, imaging, and tissue-sampling each of these specimens. The second trip searched non-databased herbarium material for any remaining needed species that might be present.

In total, 538 specimens and 209 species were sampled in eleven days, and 193 of these species (92%) produced a barcode for at least one plant barcode marker. The oldest herbarium specimen successfully sequenced from CAN was 139 years old, collected in 1875.

"...reached 85% barcode coverage of Canadian vascular plants as a result of recent herbaria visits."

In November 2014, three members of our Bioinventory and Collections Unit (Allison Brown, Elizabeth Sears, and Connor Warne) visited the University of British Columbia Herbarium (UBC), Beaty Biodiversity Museum. UBC is home to more than 235,000 accessioned specimens of vascular plants from around the world and contained almost 500 Canadian species that we needed to DNA barcode.

The assistant curator Linda Jennings, curators Sean Graham and Quentin Cronk, and graduate student Erin Fenneman hosted this visit and they, along with several exceptional undergraduate helpers, offered valuable insight and graciously provided their support.

The team made impressive progress, sampling 1180 specimens and 497 species in fifteen days, with 445 of these species (90%) producing a barcode for at least one plant marker. The oldest specimen successfully sequenced from this herbarium was 122 years old, collected in 1892.

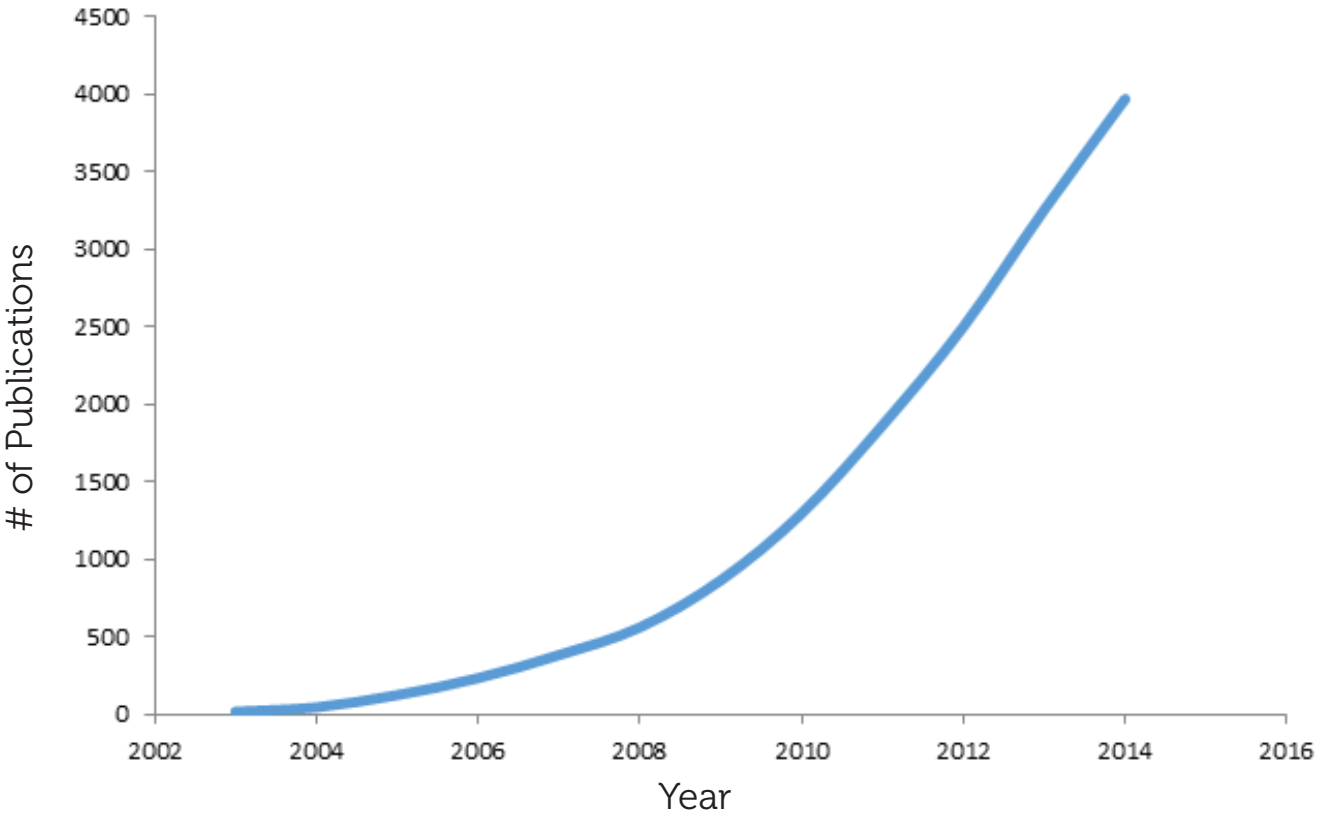
We have now barcoded 4765 vascular plants of Canada, with 388 new species added, and have reached 85% barcode coverage for this project as a result of our recent herbaria visits. There are 407 species remaining (817 species if we include hybrids). We are very close to completing our goal of barcoding all Canadian vascular plant species, but the search continues as well as a unified effort between collaborators to track down these remaining rare species over the next few months.

Image credit: Allison Brown

Research

Trends in DNA Barcoding Publications

Written by: Dirk Steinke (Biodiversity Institute of Ontario)



This graph shows the cumulative number of publications in the field of DNA barcoding from 2003 until the current day. The beginning of 2015 marked an impressive milestone; the barcoding community surpassed the 4000 paper mark, with the count now at 4015 (March 2015).

Over the years, all of these publications have amassed about 60,000 citations. The trajectory of all these measures are still showing an increasing trend. All in all, signs of a very healthy, young and growing discipline.

Data retrieved from Web of Knowledge, ISI Thompson.

Credits and Contributions

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The Barcode Bulletin owes its success to the valuable contributions of researchers and enthusiasts within the global DNA barcoding community. If you wish to contribute please contact us at bulletin@ibol.org