The Barcode of Wildlife Project and the Regulatory and Forensic Uses of BARCODE Records

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Why aren't regulatory agencies and companies adopting DNA barcoding?

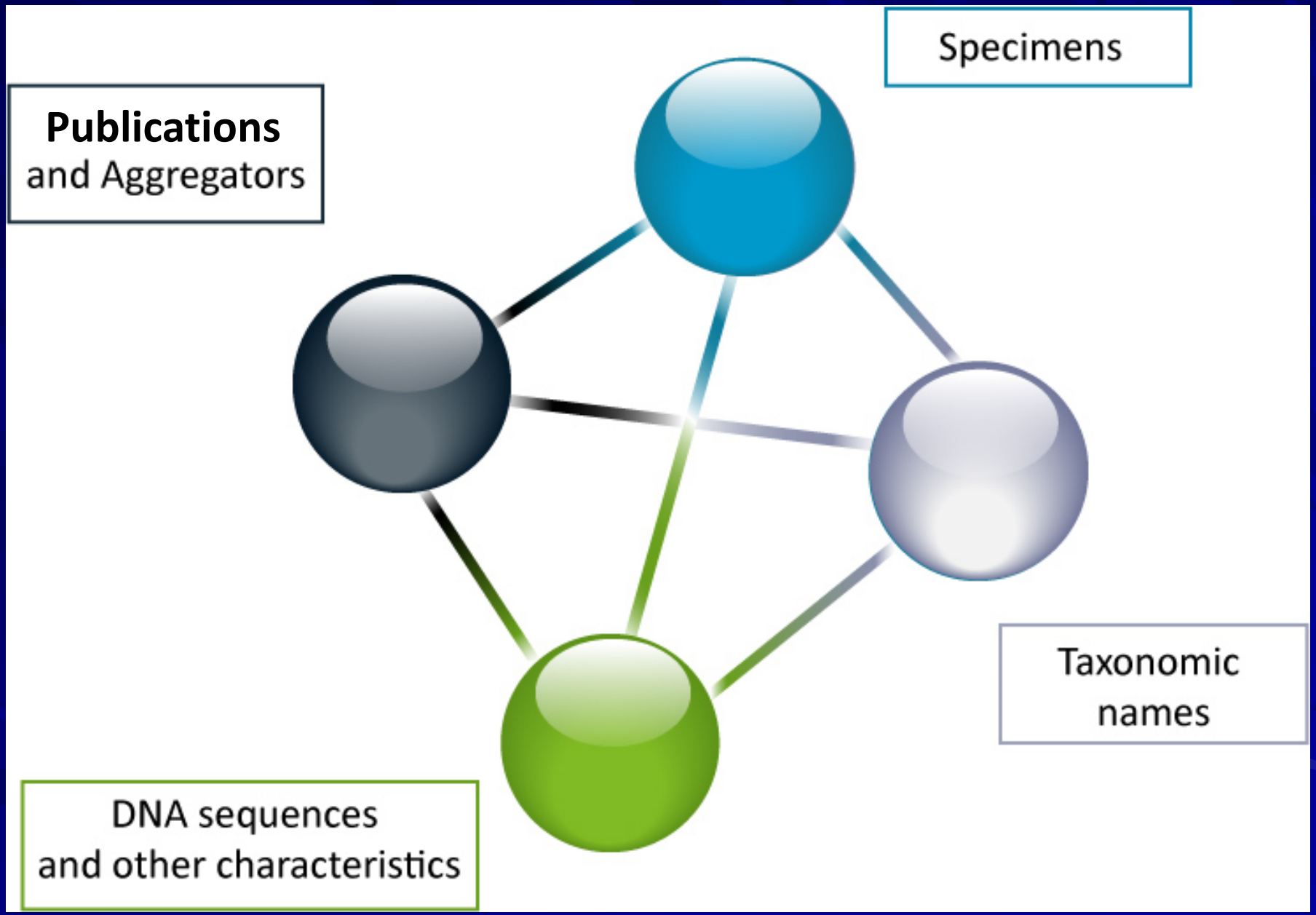
Peter Christey
Vice President at Life Technologies, San Francisco
CBOL Executive Committee Member

CBOL's Post-Adelaide Priorities

- Distribute functions, e.g., conferences
- Strengthen the BARCODE data standard
- Increase compliance with standard
- Shift to managing large projects:
  - Endangered species
  - Agricultural pests
  - Consumer protection
  - Water quality
  - Medicinal plants

BARCODE Data Standard

- A set of required elements for a reserved Keyword ('BARCODE') in GenBank
  - Ensure data longevity by archiving in GenBank
  - Enable comparisons among records from approved BARCODE gene regions
  - Ensure minimum quality of sequences
  - Enable georeferencing
  - Provide traceability to voucher specimen
  - Ensure access to raw sequencer data
  - Pave the way for regulatory and forensic use


Specimens
Publications
and Aggregators
Taxonomic
names
DNA sequences
and other characteristics

Two Communities of Practice

- **Centrifugal Barcoding**
  - One idea applied in different applications and diverse users
  - United loosely by the BARCODE data standard
  - Compliance and data release challenging
- **Centripetal Barcoding**
  - Different users converge around the a shared need and solution
  - Users demanding a stronger data standard
  - Compliance with data standards a core value

Centrifugal Barcoding

- 2+ million records in BOLD workbench
  - Large portion not yet made public
  - Uneven compliance with BARCODE standard
- Highly varied taxonomic coverage, best in insects
- Primary support from research grants
- Funding programs in several countries
- 1700+ journal articles, primarily taxonomic and ecological studies

Display Settings: GenBank

Send:

Zosterops nigrorum voucher USNM:Birds:607559 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial

GenBank: JQ176684.1

FASTA Graphics

Go to:

```
LOCUS       JQ176684                657 bp    DNA     linear   VRT 27-JAN-2012
DEFINITION  Zosterops nigrorum voucher USNM:Birds:607559 cytochrome oxidase
            subunit 1 (COI) gene, partial cds; mitochondrial.
ACCESSION   JQ176684
VERSION     JQ176684.1  GI:359285639
DBLINK      BioProject: PRJNA81595
KEYWORDS    BARCODE.
SOURCE      mitochondrion Zosterops nigrorum (yellowish white-eye)
  ORGANISM  Zosterops nigrorum
            Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi;
            Testudines + Archosauria group; Archosauria; Dinosauria;
            Saurischia; Theropoda; Coelurosauria; Aves; Neognathae;
            Passeriformes; Zosteropidae; Zosterops.
REFERENCE   1  (bases 1 to 657)
  AUTHORS   Schindel,D.E., Stoeckle,M.Y., Milensky,C., Trizna,M., Schmidt,B.,
            Gebhard,C. and Graves,G.
  TITLE     Project description: DNA barcodes of bird species in the national
            museum of natural history, smithsonian institution, USA
  JOURNAL   Zookeys 152, 87-92 (2011)
   PUBMED   22287908
REFERENCE   2  (bases 1 to 657)
  AUTHORS   Schindel,D.E., Stoeckle,M.Y., Milensky,C., Trizna,M., Schmidt,B.,
            Gebhard,C. and Graves,G.
  TITLE     Direct Submission
  JOURNAL   Submitted (29-NOV-2011) CBOL, Smithsonian National Museum of
            Natural History, 10th and Constitution Ave NW, Washington, DC
            20013, USA
FEATURES             Location/Qualifiers
     source          1..657
                     /organism="Zosterops nigrorum"
                     /organelle="mitochondrion"
                     /mol_type="genomic DNA"
                     /specimen_voucher="USNM:Birds:607559"
                     /db_xref="taxon:135985"
                     /country="Philippines: Luzon"
                     /lat_lon="17.833 N 122.017 E"
                     /collection_date="10-May-1989"
                     /PCR_primers="fwd_seq: ttctccaaccacaaagacattggcac,
                     rev_seq: acttctgggtggccaaagaatcagaa"
     gene            <1..>657
                     /gene="COI"
     CDS             <1..>657
                     /gene="COI"
                     /codon_start=1
                     /transl_table=2
                     /product="cytochrome oxidase subunit 1"
```

Change region shown

Customize view

Analyze this sequence

- Run BLAST
- Pick Primers
- Highlight Sequence Features
- Find in this Sequence

Related information

- Related Sequences
- BioProject
- Full text in PMC
- Protein
- PubMed
- Taxonomy
- Trace Archive

Recent activity

Turn Off Clear

- Zosterops nigrorum voucher USNM:Birds:607559 cytochrome o... Nucleotide
- schindel (2804) Nucleotide

See more...

Voucher specimen links constructed from Darwin Core Triplet:

http://collections.mnh.si.edu/services/resolver/birds/621682

USNM Resolver Webservice Results

The USNM Resolver Webservice is a simple XML-based webservice that was constructed to provide automatic linkages from Genbank's specimen voucher field to the Smithsonian Natural History Museum's collections database.

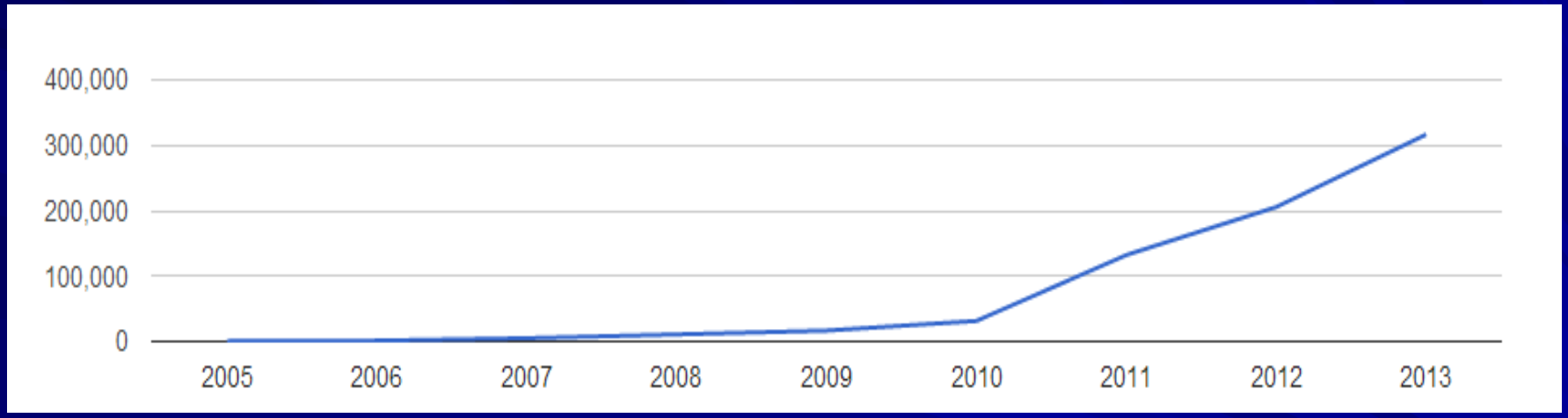
If you are reading this message, you are viewing the results on a web browser. To view and download the base XML results, select "View Page Source" on your browser.

Terms & Conditions for Use of Online Collections Databases

Results for USNM:Birds:607559

Scientifc Name:	Zosterops nigrorum aureilis
Collector:	R. Dickerman et al.
Collection Date:	
Location:	Asia, Philippines, Luzon, Cagayan
Sex:	Male
Life Stage:	
Preparation Type:	Skin: Whole
Weight:	
Globally Unique Identifier:	607559.4239705
Date Last Modified:	2013-6-1T10:07:02.000Z

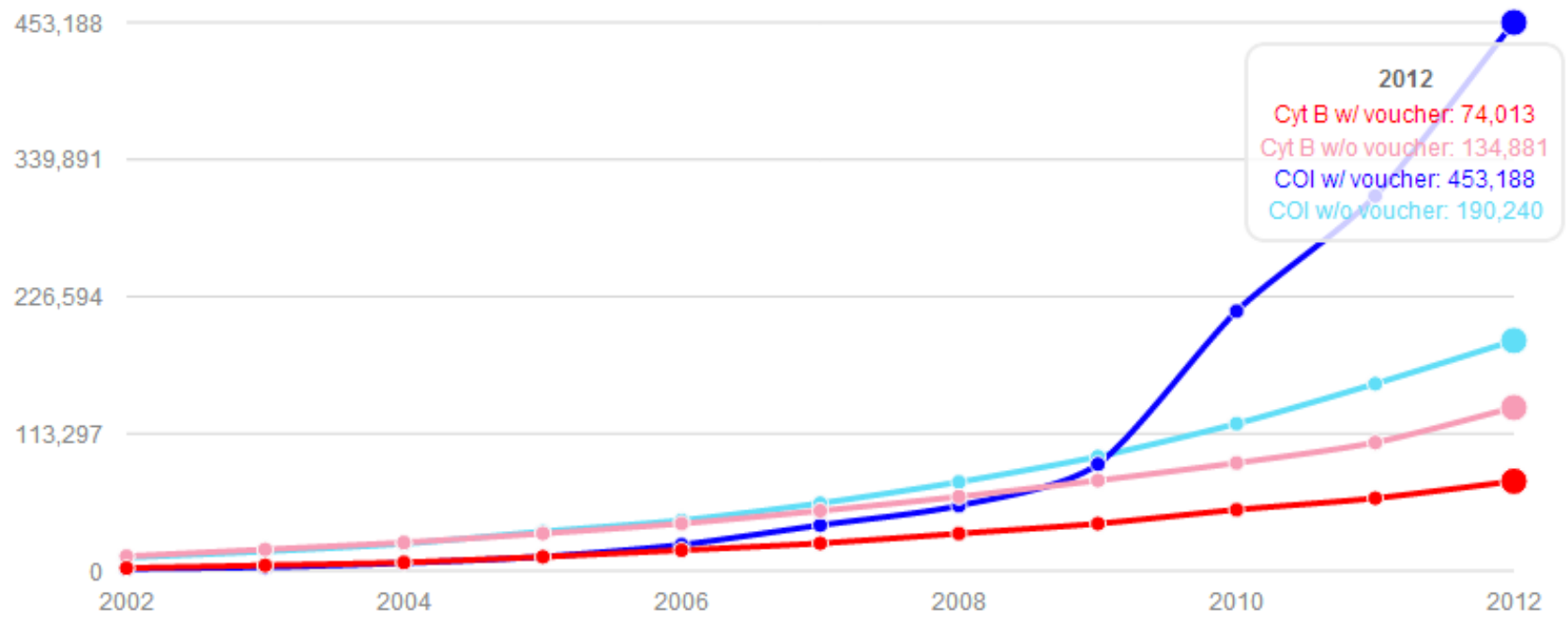
BARCODE Records in GenBank



COI and Cyt B Comparison

Label	Color
Cyt B w/ voucher	
Cyt B w/o voucher	
COI w/ voucher	
COI w/o voucher	

Cumulative Genbank Records for all Taxons



Compliance with Standard

Categories of data records	Number of GenBank records	With Voucher or Culture Collection Specimen IDs	With Latitude/ Longitude
BARCODE	347,349	347,077 (~100%)	287,058 (83%)
COI, non-BARCODE	404,606	184,351 (46%)	78,974 (20%)
All 16S	4,876,284	138,921 (3%)	461,030 (9%)
All cytb	239,796	84,784 (35%)	7,776 (3%)

Centrifugal BARCODE Data

- 2.6 million records in BOLD (50% public)
- 347,487 BARCODE records in GenBank
- 347,357 have an entry for voucherID, bio-material or culture collection
- 347,269 have Country/Ocean
- 287,058 (83%) have latitude/longitude
- 282,542 (81%) have two trace files
- 189,956 (55%) have a formatted VoucherID

Darwin Core Triplet
Structured Link to Vouchers

Institutional ID	:	Collection ID	:	Catalog ID
NHMUK	:	ENT	:	123456
personal	:	DHJanzen	:	SRNP12345

Persistent URI Pattern

- iDigBio recommendation:

```
Pattern:
http://ids.flnmh.ufl.edu/herb/abcd12345678
\_____/\_______________/\____/\__________/
   |             |           |        |
Prefix        Domain         |    Object Name
                             |
                  Collection Identifier
```

- USNM implementation:

```
http://collections.mnh.si.edu/services/resolver/resolver/birds/12345
\___/ \________________________________________________/  \___/ \____/
```

CBOL/GBIF/NCBI
Registry of Biorepositories

www.biorepositories.org

Ambiguous InstitutionIDs

AMNH	Icelandic Institute of Natural History, Akureyri Division	Akureyri	Iceland
AMNH	American Museum of Natural History	New York	USA
UNL	Universidad Autónoma de Nuevo León	Monterrey, Nuevo León	Mexico
UNL	University of Nebraska State Museum	Lincoln, Nebraska	USA
UNL	Centro de Estratigrafia e Paleobiologia da Universidade Nova de Lisboa	Monte de Caparica	Portugal
ZMK	Zoological Musem, Kristiania	Oslo	Norway
ZMK	Zoologisches Museum der Universität Kiel	Kiel	Germany
ZMK	Zoological Museum, Copenhagen	Copenhagen	Denmark

GRBio
GLOBAL
REGISTRY OF
BIODIVERSITY
REPOSITORIES

Repositories Institutional Collections Personal Collections Staff Members

The Global Registry of Biorepositories (GRBio)

Biodiversity research relies on access to specimens that have been collected over centuries and are maintained in long-term storage in reference collections around the world. The Global Registry of Biorepositories (GRBio) is a central registry of the institutions and collections that contain these specimens. GRBio is also the web portal that indexes information in the registry and provides the tools for updating, searching, and exporting this information. GRBio is moderated by a panel of data curators who check the validity of newly entered information before it is published to the portal.

GRBio has two main goals:

- To improve access to information about biorepositories, the collections and specimens they house, and the researchers and collection managers who work there; and
- To facilitate electronic linkages to this information through web services that will rely on unique identifiers assigned to biorepositories and collections.

GRBio includes four categories of data records that provide information on:

- Institutional repositories such as museums, herbaria, botanical gardens, zoos, biomedical research institutes and culture centers;
- Institutional collection records such as the bird, algal or insect collections within an institutional repository;
- Personal collections such as field samples held by a researcher before they have been accessioned into an institutional collection, or privately owned specimens held by non-researchers; and
- Staff members at institutional repositories.

Index Herbariorum records have been incorporated into GRBio and they will continue to be treated as before. The New York Botanical Garden will oversee the review of IH records and all the previous functions of IH have been incorporated into GRBio, with certain improvements.

The Globally Unique Identifiers (GUIDs) assigned to collections by BCI have also been preserved in GRBio, along with BCI's web services.

Progress

7014 Biorepository Records

471 Institutional Collection Records

18 Personal Collection Records

6899 Staff Member Records

Sponsors

The Global Registry of Biorepositories is a merger of three prior registries and an ongoing collaboration among them:

Index Herbariorum at the New York Botanical Garden

THE NEW YORK BOTANICAL GARDEN

Biodiversity Collections Index (BCI) at the Royal Botanic Garden Edinburgh

Biodiversity Collections Index

Biorepositories.org created by the Consortium for the Barcode of Life (CBOL) at the Smithsonian Institution

CONSORTIUM FOR THE BARCODE OF LIFE

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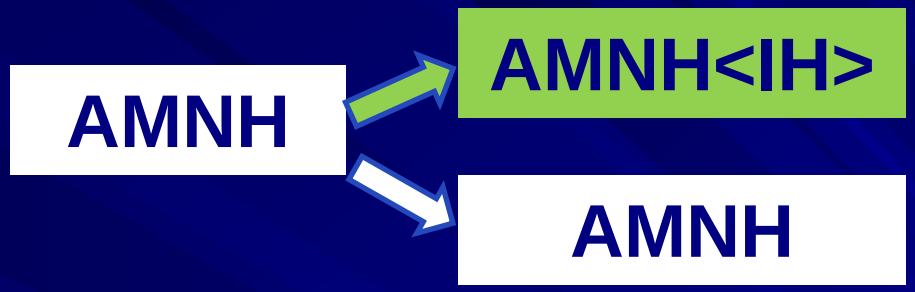
CONSORTIUM FOR THE BARCODE OF LIFE

Repositories Institutional Collections Personal Collections Staff Members



	Biorepositories.org, 2012			GRBio, 2013	
Number of Institutions	6702			7014	
Institutions w/ unique InstIDs	6036	**90.1%**		6738	**96.1%**
Insts w ambiguous InstIDs	666	9.9%		276	3.9%
Ambiguous InstIDs	**299**			**128**	
Collisions with IH	200			0	

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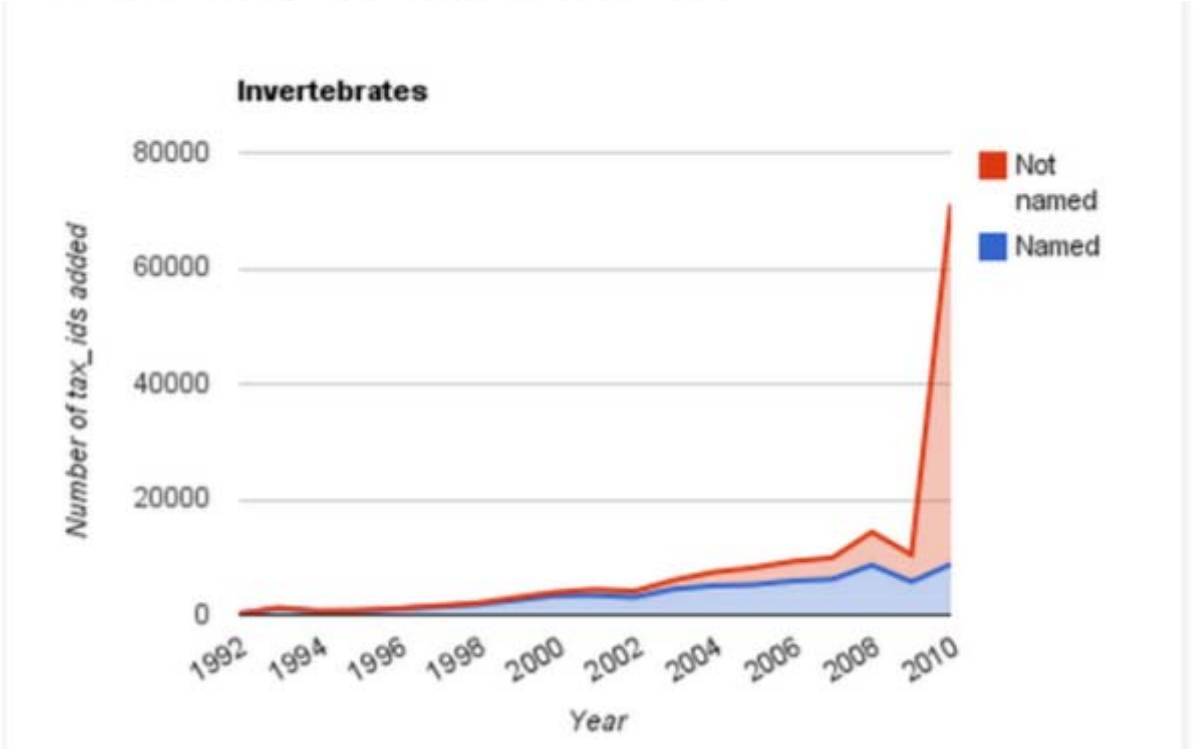
- 149,114 (43%) have "sp." in taxonomic ID

Rod Page's 'Dark Taxa':
How reliable are the identifications?

For "invertebrates" 2010 saw an explosive growth in the number of new taxa sequenced, with nearly 71,000 new taxa added to GenBank.



This coincides with a spectacular drop in the number of properly-named taxa, but even before 2010 the proportion of named invertebrate species in GenBank was in decline: in 2009 just over a half of the species added had binomials.

R. Page, iPhylo blogspot, 12 April 2011

How effective has the BARCODE data standard been in meeting expectations of regulatory agencies?

US Food & Drug Administration Reference Fish Encyclopedia

- 427 records, 389 vouchers
- Not BARCODE records in GenBank, BOLD

DNA Barcode Sequences for Seafood Species Identification: *Oncorhynchus gorbuscha*, Pink Salmon, FDA Market Name: Salmon, Pink or Humpback

Sample #FDA-245

Oncorhynchus gorbuscha, Pink Salmon, FDA Market Name: Salmon, Pink or Humpback
Authenticated: yes; vouchered: yes
FDA Market Sample, Alaska
COI DNA Sequence (barcode, FASTA format)

Additional information: NMNH[a] ID 404986

```
>FDA245_Oncorhynchus_gorbuscha
CCTCTATTTAGTATTTGGTGCCTGAGCCGGGATAGTAGGCACCGCCCTAAGCCTACTAATTCGG
GCAGAACTAAGCCAGCCAGGCGCTCTTCTAGGGAATGACCAGATCTATAACGTAATCGTTACAG
CCCATGCCTTCGTTATGATTTTCTTTATAGTCATACCAATTATAATCGGAGGCTTTGGAAACTG
ATTAATCCCCCTAATGATCGGGGCACCAGATATAGCATTTCCACGAATAAACAACATAAGCTTC
TGACTCCTACCCCCATCCTTCCTCCTCCTCCTTTCTTCATCTGGAGTCGAAGCCGGCGCTGGTA
CCGGATGGACAGTTTATCCCCCTCTAGCCGGGAACCTTGCCCACGCAGGAGCATCCGTCGACTT
GACTATCTTCTCCCTTCATTTAGCTGGAATCTCATCAATTTTAGGGGCCATTAATTTTATTACG
ACCATTATCAACATAAAACCACCGGCAATCTCTCAGTACCAAACCCCACTTTTTGTTTGAGCTG
TGCTAATCACTGCTGTACTTCTACTACTATCCCTCCCCGTTCTGGCAGCAGGTATCACTATGTT
GCTCACGGACCGAAATTTAAACACTACTTTCTTTGACCCAGCGGGGGGCGGAGATCCAATTTTA
TACCAACACCTCTTT
```

Authenticated: yes

Sample FDA-245 photograph: Full-size photograph (1280 × 960 px)

The Barcode of Wildlife Project

- Global Impact Award from Google Giving, 2012
- US$3 million to CBOL/Smithsonian, 2 years
- Management and funding by objectives
- 4 Phases:
  i. Planning, assessment, selection of priority species
  ii. Training
  iii. Testing
  iv. Implementation
- Build public awareness, political/financial support

Google+ Page and Communities

Barcode of Wildlife Project
Shared publicly - Yesterday 9:12 AM

What do conservation groups have to say about the U.S. effort to curb wildlife poaching in Africa? Darryl Fears of The Washington Post looks into the administration's and Hillary Clinton's plans to put a stop to wildlife crime.

#Africa #wildlife #wildlifecrime #endangeredspecies

U.S. anti-poaching efforts need to go

Barcode of Wildlife Project
Shared publicly - Oct 25, 2013

Although it seems incredible, a popular belief in Mexico has endangered the population of different species of turtle, because many persons believe that the eggs of these sea animals contain aphrodisiac properties.

(Original note in Spanish, you can use translate.google.com)

Fotos de la biografía - Barcode of

In their circles 52 people

National Wildlife Federation — Following

Lantum Emmanuel — Following

Andrew Consing — Following

Search

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- Favorites
- Lists

Follow Barcode of Wildlife

Full name

Email

Password

Sign up

Worldwide Trends · Change

#UNIVERSALMGJUSTINBIEBER
#babyfletcher
#LeQuieroLeerElPensamientoA
#sepotessiabbracciarequalcunoabbraccerei
#iyikidoğdunTürkiyeCumhuriyeti
Ley de Medios
Zaha and Januzaj
De Bruyne
Ne Mutlu Türküm Diyene
M&G

Barcode of Wildlife

@BoWProject

Defending Endangered Species

goo.gl/uB1D6

69 TWEETS | **44** FOLLOWING | **16** FOLLOWERS | Follow

Tweets

National Zoo @NationalZoo 22h

MT @smithsonian If you thought new species discovery #Olinguito was cute, wait 'til you see its babies! s.si.edu/1aOZ3gn via @khelgen

Retweeted by Barcode of Wildlife

Expand

Reply Retweet Favorite More

Barcode of Wildlife @BoWProject 28 Oct

@washingtonpost's Darryl Fears looks into the #US plan to put a stop to #wildlife crime in #Africa: wapo.st/1dhv04C

View summary

Reply Retweet Favorite More

Barcode of Wildlife @BoWProject 24 Oct

poaching & trafficking aren't all that's threatening #wildlife; see what #TX is doing to conserve a rare bird: bit.ly/1c0j3Pj

Expand

Reply Retweet Favorite More

World Wildlife Fund @World_Wildlife 23 Oct

There are fewer than 7,500 snow leopards left in the wild. Ambitious new plan aims to protect them wwf.to/19v6HhU

Retweeted by Barcode of Wildlife

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Barcode of Wildlife Project

Subscribed 6

Videos Discussion About

All activities

Barcode of Wildlife Project uploaded a video

The Barcode of Wildlife Project - episode 4
3 months ago • 45 views
How synonymous is animal DNA to human DNA and fingerprints? Through developing a database, much like criminal DNA databases, the Barcode of Wildlife Project will be able to...

Barcode of Wildlife Project uploaded a video

Barcode of Wildlife Project - episode 3
3 months ago • 31 views
The Barcode of Wildlife Project takes you from the wild into the fish market, revealing some issues in the retail food industry when it comes to identifying sea life.

Barcode of Wildlife Project uploaded a video

Barcode of Wildlife Project - episode 2
4 months ago • 56 views
In this video, the Barcode of Wildlife Project shows how DNA barcoding is synonymous to the universal product codes on retail items, like those in supermarkets.

Barcode of Wildlife Project uploaded a video

BWP Goals

Working with six Partner Countries:

- Demonstrate use of DNA barcode evidence in investigations, prosecutions, convictions by November 2014
- Construct a reference BARCODE library to support Partner Country priorities
  - ~2000 Priority Endangered Species
  - ~8000 closely related/look-alike species
- Partner Countries will formally adopt, implement and **sustain barcoding**

BWP Current Status

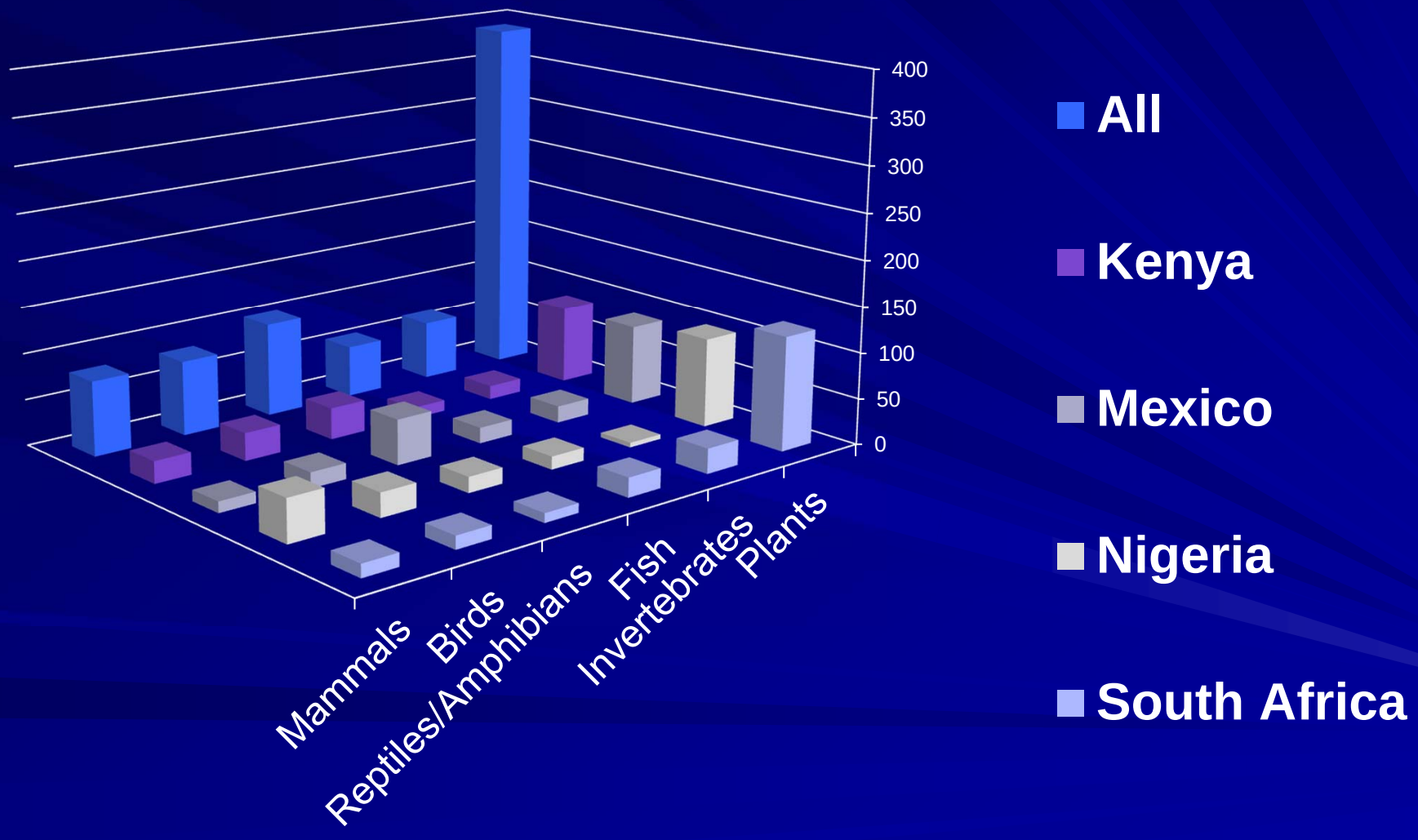
- Mexico, South Africa, Kenya, Nigeria completing Phase 1
- Partner countries in SE Asia and South America being selected
- 200 Priority Endangered Species selected
  - Heavily trafficked, hard to identify
- **National workshops on legal standards** for admissibility as courtroom evidence
  - Enforcement agencies, police, prosecutors, researchers involved, awaiting training

Fitness for Use in Courtrooms

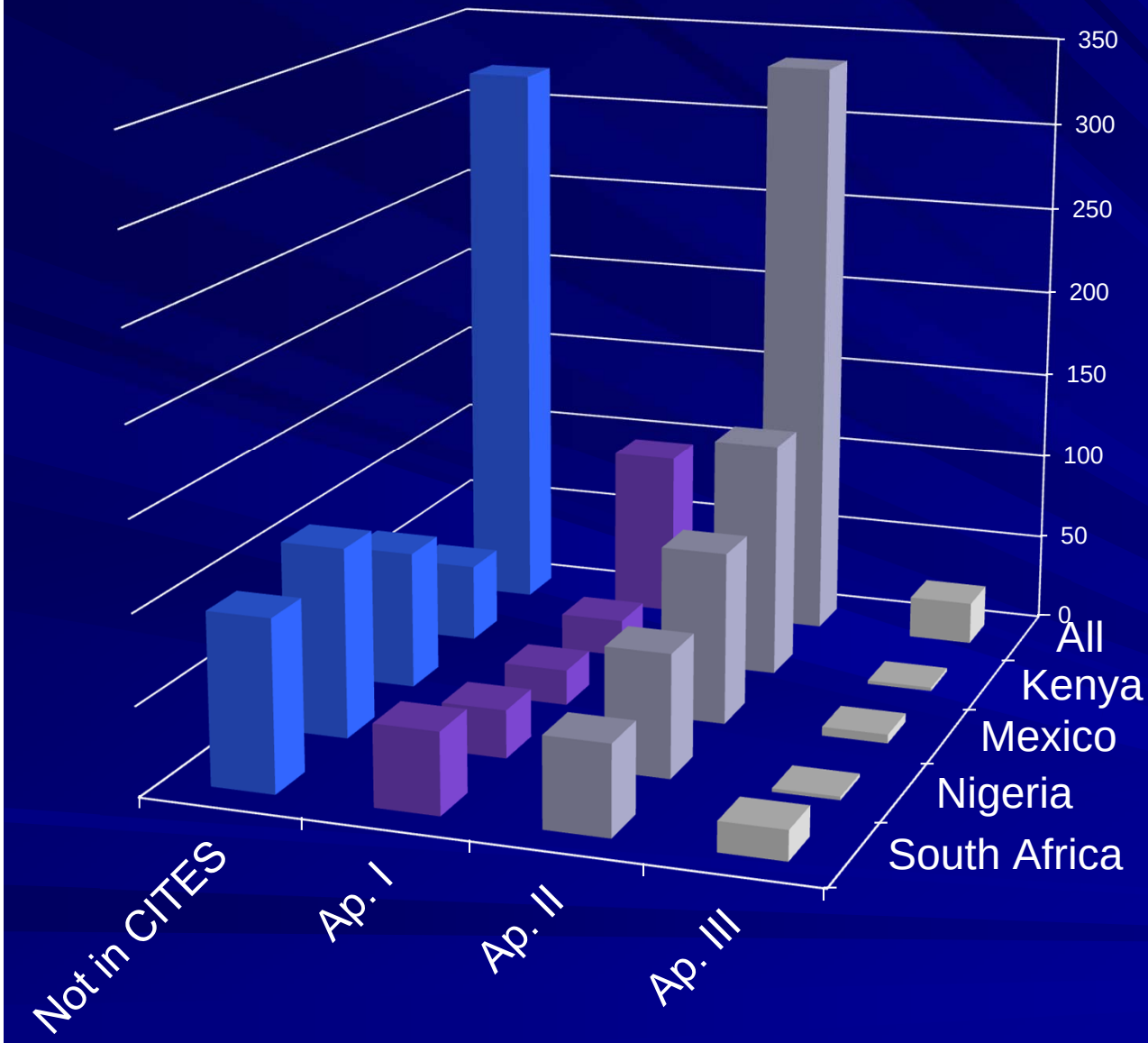
- Default mentality from Human DNA IDs
  - "Are these two items from same individual?"
  - NOT "Is this item from that species?"
- Larger sample size versus security of samples
- Barcode IDs: Statistical results or opinions?
- Chain of custody not compatible with museum/herbarium culture of openness
- No background studies of wildlife DNA by Academies, Institute of Justice, Interpol


BARCODE OF WILDLIFE PROJECT
BWP
GLOBAL IMPACT AWARDS
Project
Participation
Partner Countries
Priority Species
Meetings
Media
Documents
Donate
BarcodeofWildlife.org
Enforcing Endangered Species Laws with DNA Barcodes
Overview
Illegal poaching and international trafficking in endangered species ranks among the largest of crimes, representing tens of billions of dollars per year. On 6 December 2012, Google Giving announced the first seven Global Impact Awards, including a $3 million grant to the Smithsonian Institution for the Barcode of Wildlife Project (BWP). BWP will enable six developing partner countries to begin using DNA barcodes to identify species from minute samples in the investigation and prosecution of wildlife crimes. Learn more from:
Description
Timeline
Roadmap
Progress
Mexico, Kenya, Nigeria and South Africa have held inaugural planning meetings and formed National Project Committees
South Africa has selected 212 priority endangered species for DNA barcoding in preparation for possible prosecution cases
The first workshop on legal standards for the admissibility of DNA barcodes in law courts was held in Pretoria, South Africa on 5 June 2013
Thailand was selected as the sixth partner country in the Barcode of Wildlife Project
New Documents
BWP Project Roadmap
Endangered Species Project
Project Timeline
Google+
YouTube
Facebook
Contact

774 Priority Endangered Species: Taxonomic Distribution

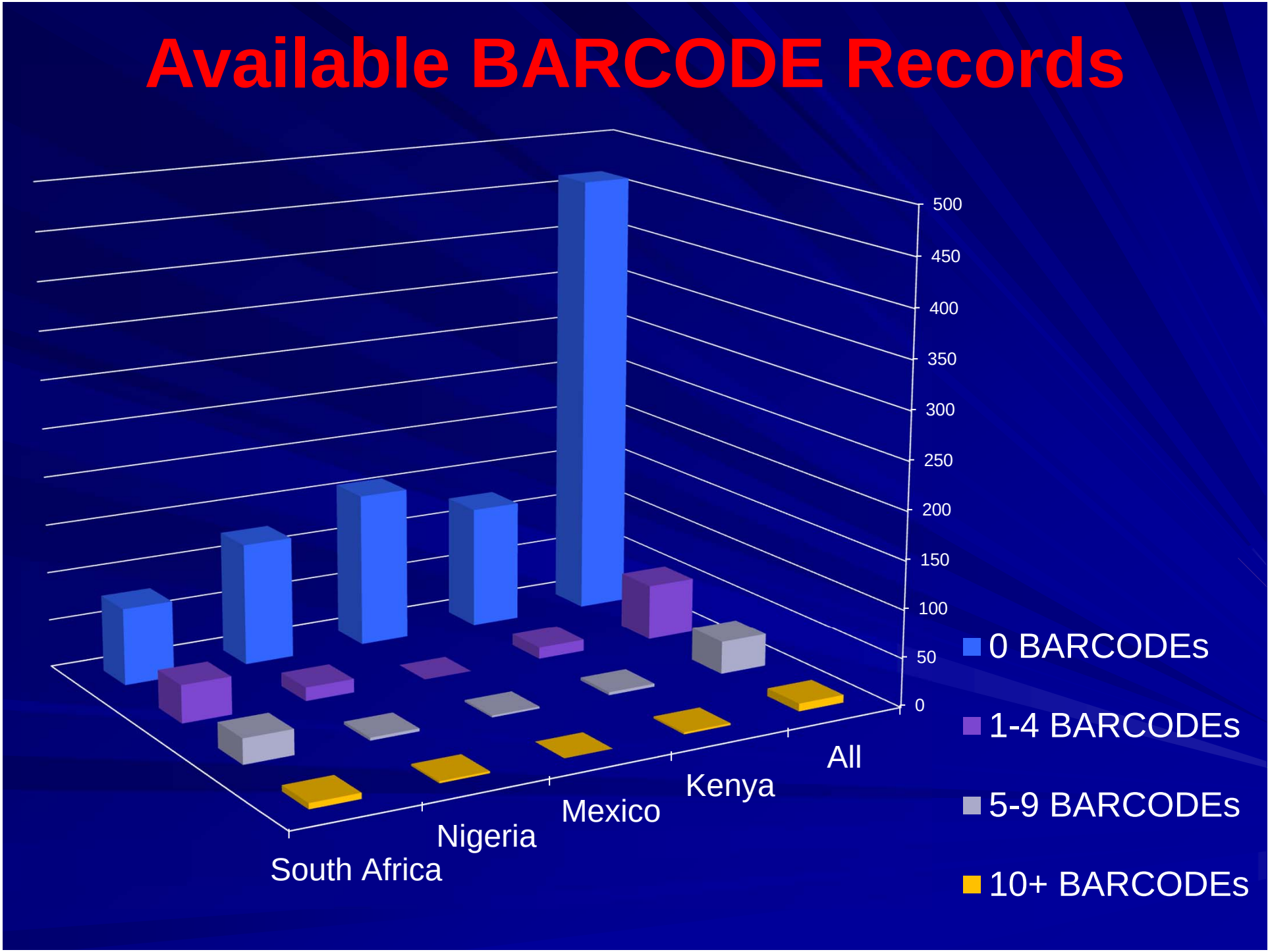


Source of Protected Status



- Not in CITES
- Ap. I
- Ap. II
- Ap. III

Available BARCODE Records



Priority Species Viewer

http://www.barcodeofwildlife.org/priority_species.html

Priority Species Viewer

You are currently viewing the dynamic priority species viewer.

Click here to view all of the pri

Abronia graminea
Family: Anguidae

Acinonyx jubatus
Cheetah
Family: Felidae

Aegypius occipitalis
White-headed Vulture
Family: Accipitridae

Acinonyx jubatus

Source: Wikimedia Commons

NCBI
NCBI Taxonomy Link: 32536

Encyclopedia of Life
EOL Link: 328680

0 official BARCODE records

GBIF

IUCN

Close

Taxonomic Reliability Data/Metadata

- Additional datafields in GenBank for BWP:
  - Name of identifier
  - Date of identification
  - Type status of voucher specimen
  - Basis of identification
  - Confidence level

Expanding the Data Standard

- Voluntary metadata on taxonomic reliability
- BARCODE Platinum:
  - Voucher handled under chain of custody
  - Analyzed in police forensic lab
- BARCODE Gold:
  - Based on a Platinum standard voucher
  - Analyzed in academic lab
- BARCODE Silver:
  - Includes all taxonomic reliability metadata

Terms of BWP Participation

- CBOL Call for Expressions of Interest from:
  - Taxonomic specialists on Priority Species
  - Biorepositories with samples
  - Field sampling programs
  - Labs with barcoding capabilities
- Terms of Participation: barcodeofwildlife.org
- Management by objectives
- Payment for deliverables

Conclusions

- The BARCODE data standard is not adequate for regulatory/forensic uses
- Additional data and metadata will be needed for regulatory applications
- Voluntary additions to the BARCODE data standard could enable regulatory/forensic use without undue burden on other users
- CBOL will be strengthening compliance with the BARCODE standard to protect the brand
- BWP is now open for business!

Questions?