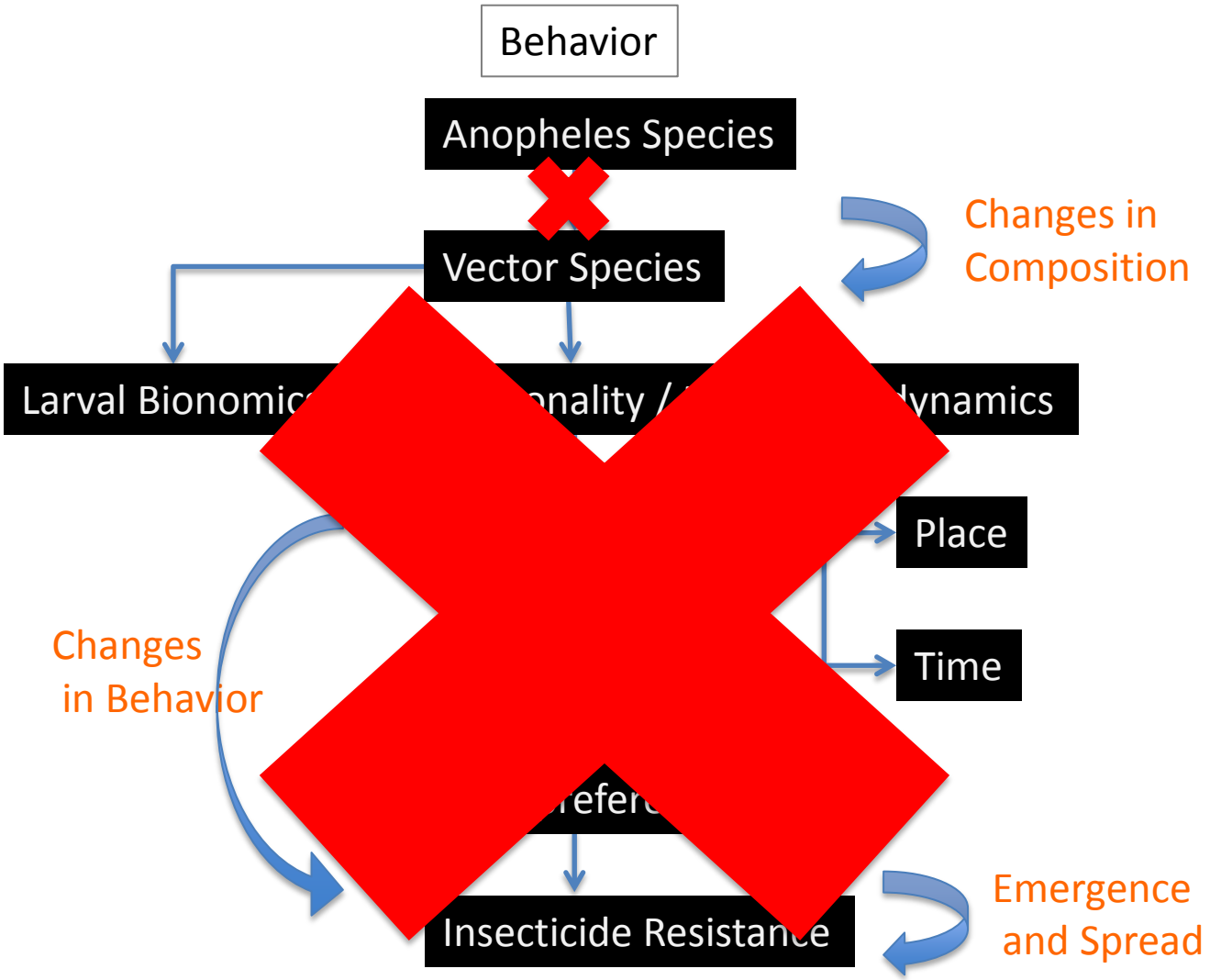
Towards better morphological and molecular identification of malaria vectors

Neil Lobo and Seth Irish

Why is Species ID important



Intervention

IRS

ITN

Larval Control

Spatial Repellants

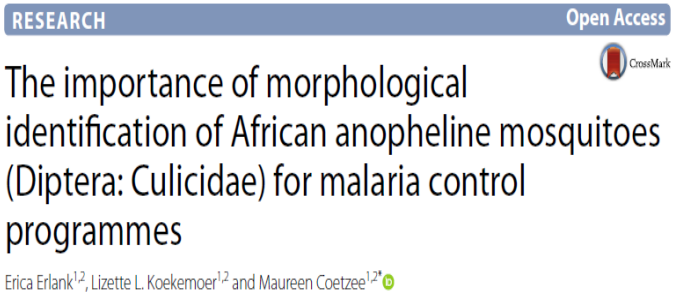
Endectocides

The problem

- Mosquitoes are difficult to identify
  - Small
  - Key characteristics can be altered (scales rubbed off) or removed (legs falling off)
  - Morphological identification keys are out of date
  - Cryptic species – molecular identification required
  - Variation seen in the field
  - Work conditions

Morphological identification aspects/challenges

- Updated keys for African *Anopheles*
  - Gillies & Coetzee (1987), supplement to Gillies & DeMeillon (1968)
  - CD-ROM by Hervy et al. (1998)
  - Lucid key, Rueda (www.wrbu.org)
- Training (and retraining) of taxonomists/entomologists
- Morphological identification is needed for some molecular assays and vice versa


RESEARCH Open Access

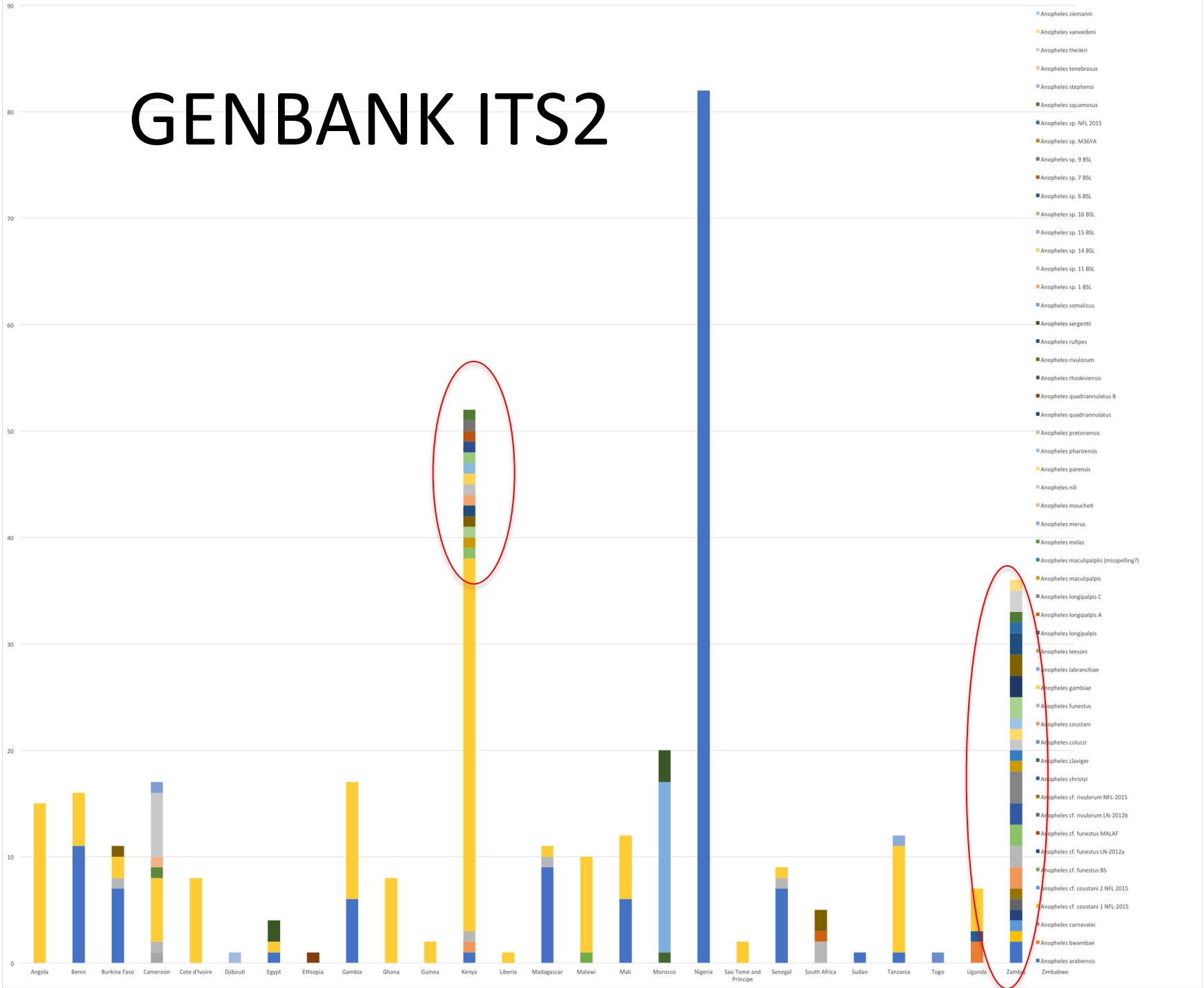
The importance of morphological identification of African anopheline mosquitoes (Diptera: Culicidae) for malaria control programmes

Erica Erlank[1,2], Lizette L. Koekemoer[1,2] and Maureen Coetzee[1,2*]

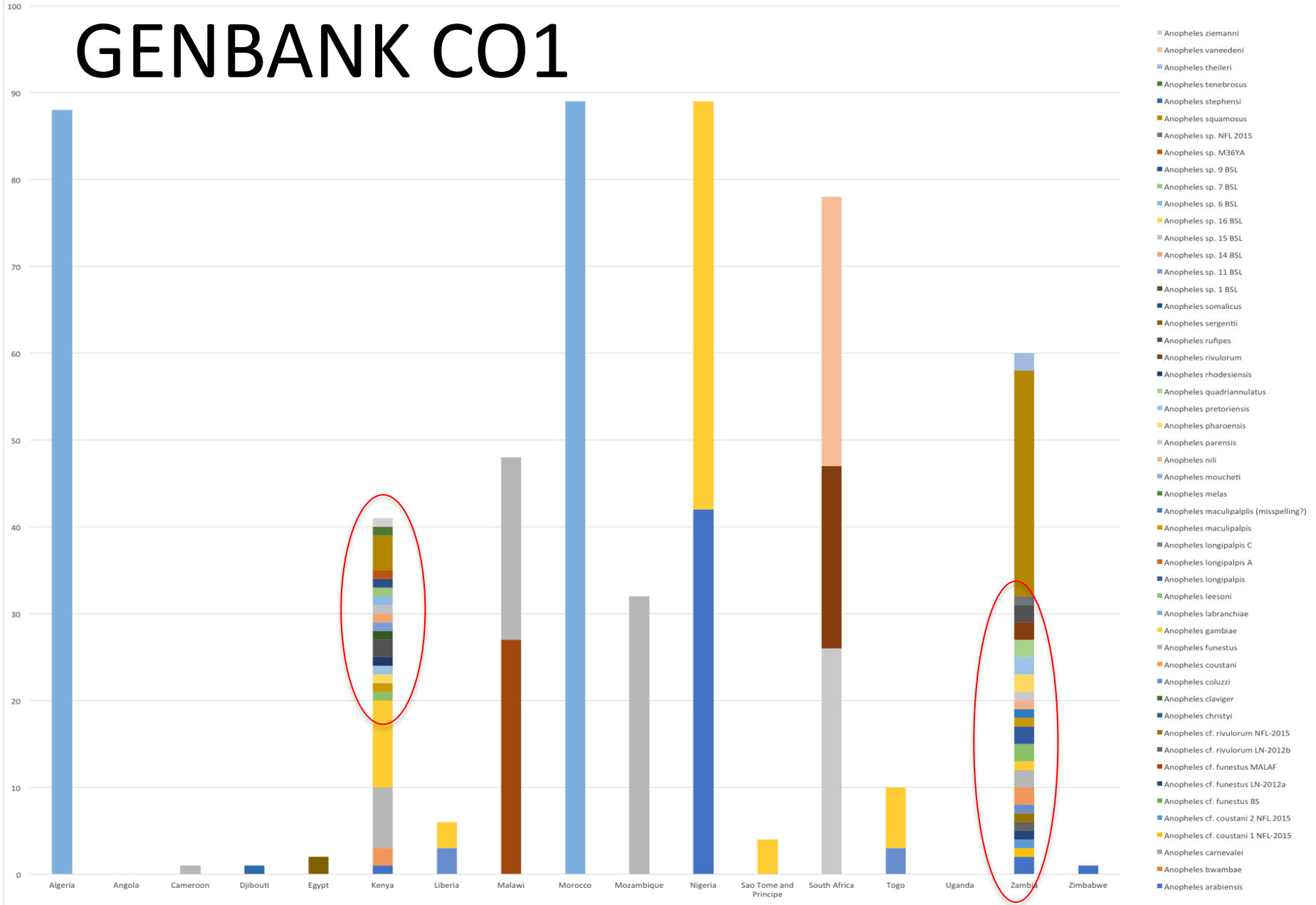
Molecular identification aspects/challenges

- PCR – identification of species complex members
  - *An. gambiae*, *An. funestus*
- Sequencing (CO1 / ITS2)
  - Identification from GenBank, BOLD
  - Relies on identifications provided by users
  - Specific, taxonomy
  - Cost / capacity
  - Ideal to link morphology with molecular data
- Ideally link morphology to molecular data

GENBANK ITS2



GENBANK CO1


100
90
80
70
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Algeria
Angola
Cameroon
Djibouti
Egypt
Kenya
Liberia
Malawi
Morocco
Mozambique
Nigeria
Sao Tome and Principe
South Africa
Togo
Uganda
Zambia
Zimbabwe
Anopheles ziemanni
Anopheles vaneedeni
Anopheles theileri
Anopheles tenebrosus
Anopheles stephensi
Anopheles squamosus
Anopheles sp. NFL 2015
Anopheles sp. M36YA
Anopheles sp. 9 BSL
Anopheles sp. 7 BSL
Anopheles sp. 6 BSL
Anopheles sp. 16 BSL
Anopheles sp. 15 BSL
Anopheles sp. 14 BSL
Anopheles sp. 11 BSL
Anopheles sp. 1 BSL
Anopheles somalicus
Anopheles sergentii
Anopheles rufipes
Anopheles rivulorum
Anopheles rhodesiensis
Anopheles quadriannulatus
Anopheles pretoriensis
Anopheles pharoensis
Anopheles parensis
Anopheles nili
Anopheles moucheti
Anopheles melas
Anopheles maculipalplis (misspelling?)
Anopheles maculipalpis
Anopheles longipalpis C
Anopheles longipalpis A
Anopheles longipalpis
Anopheles leesoni
Anopheles labranchiae
Anopheles gambiae
Anopheles funestus
Anopheles coustani
Anopheles coluzzi
Anopheles claviger
Anopheles christyi
Anopheles cf. rivulorum NFL-2015
Anopheles cf. rivulorum LN-2012b
Anopheles cf. funestus MALAF
Anopheles cf. funestus LN-2012a
Anopheles cf. funestus BS
Anopheles cf. coustani 2 NFL 2015
Anopheles cf. coustani 1 NFL-2015
Anopheles carnevalei
Anopheles bwambae
Anopheles arabiensis

Molecular data

- Supports morphological data
- Can corrects morphological data, QA
- Enable distinguishing of cryptic species, new/novel species
- Enable niche partitioning of entomological transmission dynamics with behavioral associations

- Eg.
  - *An. gambiae* complex in Kenya (*An. gambiae* s.s., *An. arabiensis*)
  - *An. cinereus* in Gondor
  - Mystery *Anopheles* in Kenya highlands
  - What's going on with *An. coustani* complex?
  - Novel species in many sites

Recommendations

- Improve morphological identification skills of entomologists
  - Trainings (and re-trainings) where many relevant species are available for study
  - Maintenance of entomological collections
  - Updated Keys (Maureen)

- Collection and deposition of mosquito specimens in museums
  - Ideally, linked larvae-exuviae-adults, and if possible, multiple progeny from a single female to create a series (some of which can be used for DNA work)

- Creation of a list for which we have “good” association of species with sequence data (and specimen in museum)
  - Testing DNA from newer species in museums
  - Collection of fresh specimens

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Resources

WRBU identification keys:
http://www.wrbu.org/aors/aors_Keys.html

GenBank:
https://www.ncbi.nlm.nih.gov/genbank/

Barcode of Life Database: www.boldsystems.org