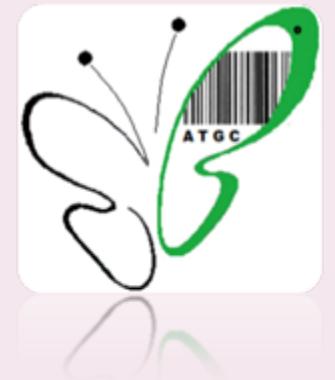


# Food Forensic using DNA Barcoding

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G. D. Khedkar Ph.D.

BOYSCAST Fellow,  
The Hebrew University of Jerusalem, Israel



**Paul Hebert Centre for DNA Barcoding  
& Biodiversity Studies,  
Dr. Babasaheb Ambedkar  
Marathwada University, POB 02,  
Aurangabad.  
E.mail- gdkhedkar@gmail.com**

# What are Food Frauds?

- when food is mislabelled and deliberately marketed, for financial gain by deceiving the consumer.
- two main types are:
  1. sale unfit and potentially harmful food,
- such as:
  - **packing and selling of beef and poultry with an unknown origin**
  - recycling of animal by-products back into the food chain
  - knowingly selling goods which are past their use by date
- the deliberate misdescription of food, whilst not necessarily unsafe,
- products substitution, eg. farmed salmon sold as wild, and Basmati rice adulterated with cheaper varieties
  - making false statements about the source of ingredients, i.e. their geographic, plant or animal origin



# Genesis behind food forensic

- Processed meat and fish products are often commercialized as stakes, frozen fillets, fins smoked, or precooked.
- The traceability for source of origin of processed food is an important issue.
- Species identification is very difficult, based on morphological characteristics.
- Mislabeling of food in markets defrauds consumers.
- Sadly, cases of fish/meat substitution do occur, generally (and unsurprisingly).



- Species identifications underpin all of biological research.
- Many species are morphologically very similar and mistakes in identification may be quite common.
- Existing morphologically-based diagnostic approaches are often both cumbersome to use and are effective only for certain life stages.
- Lacking universality standards, reorganization and more person specific.





*Nemachelius spp.*





A



B



C

(A) *Caranx ignobilis* X *C. melampygus*, (B) *Caranx ignobilis*.

(C) *Caranx melampygus*.



A



B



C

(A) *Caranx melampygus* X *C. sexfasciatus*,

(B) *Caranx melampygus*.

(C) *Caranx sexfasciatus*.

**Our publication:** PERSIS. M, CHANDRA SEKHAR REDDY. A, RAO. L.M, KHEDKAR, G. D. AND K. RAVINDER (2008): COI (Cytochrome Oxidase-I) sequence based studies of Carangid fishes of Kakinada Coast, India. **Genetics and Mol. Biol.**



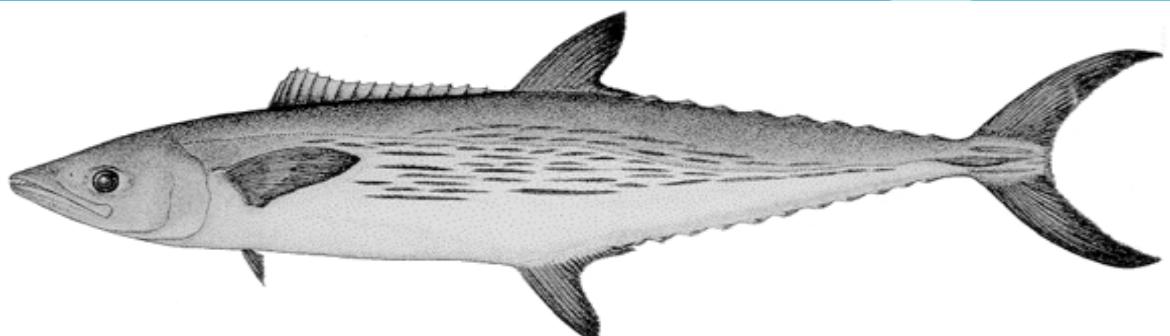
Mol Biol Rep  
DOI 10.1007/s1303-008-9375-4

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## COI (cytochrome oxidase-I) sequence based studies of Carangid fishes from Kakinada coast, India

M. Pendse • A. Chandra Sekhar Reddy •  
L. M. Rao • G. D. Khedkar • K. Ravinder •  
K. Nasaruddin





*Scomber lineolatus*  
(*Mackrel*)



*Scomber scombrus*  
(*Mackrel*)



*Scomber guttatus*  
(*Mackrel*)



*Rastrelliger kanagurta*  
(*Mackrel*)





*Anguilla bengalensis  
bengalensis*  
(Eel)



*Anguilla japonica*  
(Eel)



*Anguilla anguilla*  
(Eel)







**Restaurant owners and consumers could check fish to be sure what they are buying is what is advertised.**





**New EU Food Hygiene Legislation Regulation (EC) 178/2002 is applied from 1<sup>st</sup> January 2006 which insists for traceability requirements clearly.**

**The U.S. FDA's Center for Food Safety and Applied Nutrition recently directed the Institute of Food Technologists (IFT) to examine the current practices and technologies used to trace food products.**



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## BarCodes & Identification

[Overview](#)[Technical](#)[Implementation](#)[Support](#)[Training](#)[GS1 DataBar \(RSS\)](#)

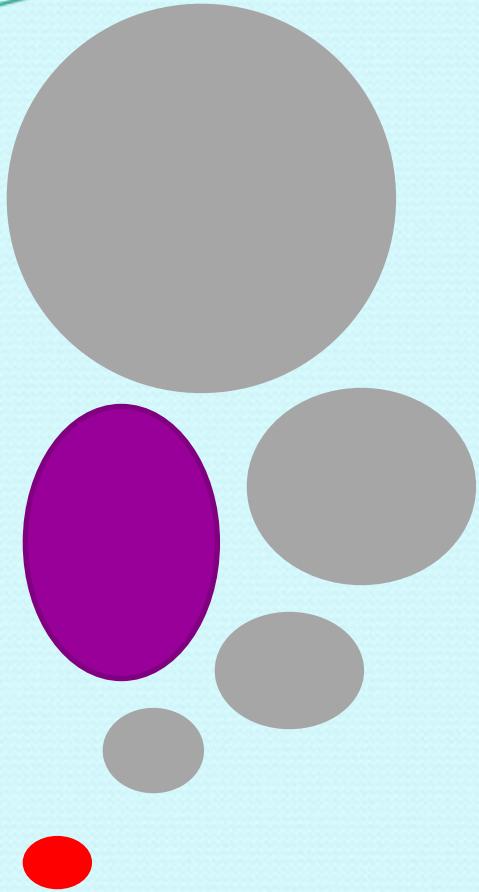
## GS1 Identification Keys (ID Keys)

There are seven GS1 Identification Keys that support the identification of items, services, locations, logistic units, returnable containers, etc. The [GS1 Company Prefix](#) assigned to a user company allow that user company to create any of the GS1 identification keys.

### Key Concepts

- GTIN - Global Trade Item Number
- GLN - Global Location Number
- SSCC – Serial Shipping Container Code
- GRAI – Global Returnable Asset Identifier
- GIAI – Global Individual Asset Identifier
- GSRN – Global Service Relation Number
- GDTI – Global Document Type Identifier
- GSIN – Global Shipment Identification Number
- GINC – Global Identification Number for Consignment





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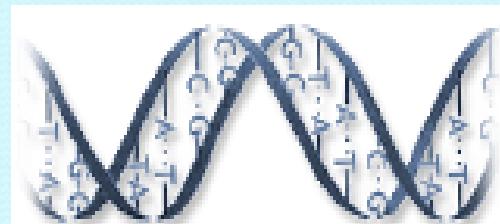
What is solution



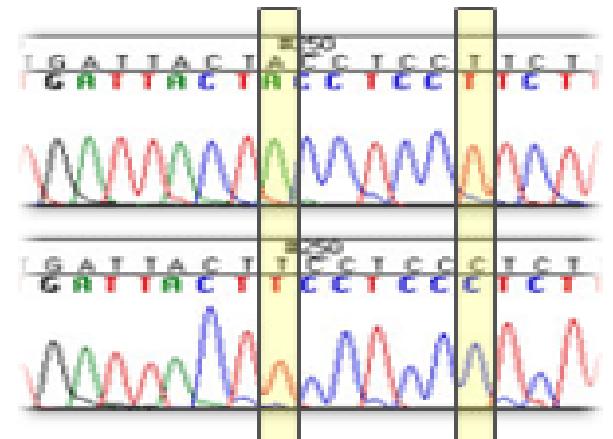
# DNA Barcoding



The UPC system can uniquely identify 100 billion individual products



DNA sequences can provide enough variation to allow species-level diagnosis



COI electropherograms provide a means of verifying key differences between species



# What is Molecular Taxonomy or DNA barcoding ??

The use of genetic markers for fish, meat or plant species identification.

A method of species identification based on DNA sequencing a single gene where very small piece or a single cell is sufficient.

The gene chosen is the mitochondrial DNA gene cytochrome oxidase I (COI).

The hypothesis is that, for COI gene, every species will have a unique sequence or a unique assemblage of closely related sequences.

This sequence is termed a 'barcode' for example:

Species A: CCTAAGCTTACGTTCC

III III III III III III

Species B: CCTAGGCTTACGTTACC

III III III III III III



# Why barcode animals with mitochondrial DNA?

Mitochondria, energy-producing organelles in plant and animal cells, have their own genome.

Ten years of research have established the utility of mitochondrial DNA sequences in differentiating among closely-related animal species.

Four properties make mitochondrial genomes suitable for identifying species:

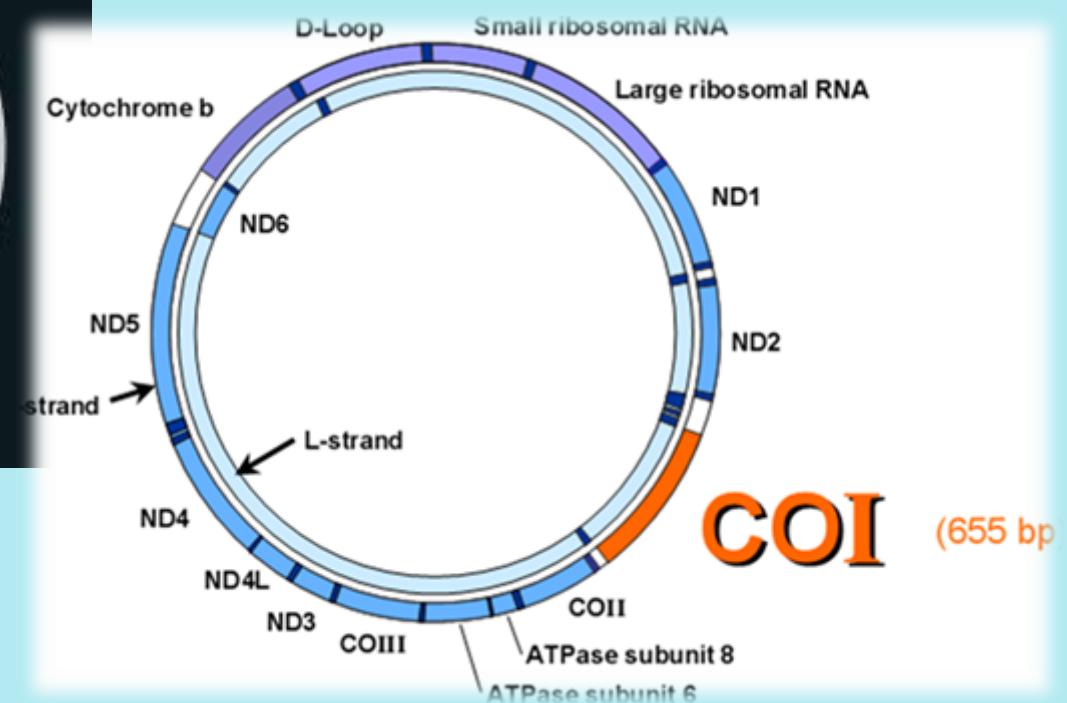
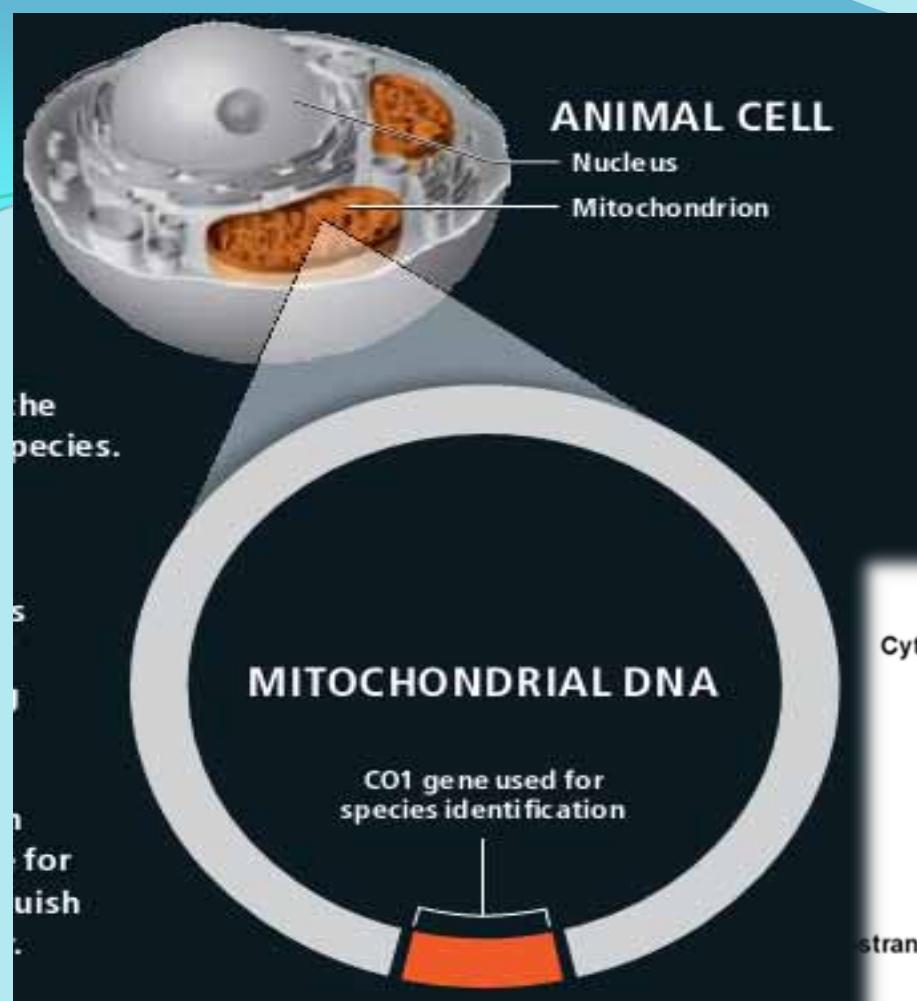
Greater differences among species,

Many copies,

Relatively few differences within species,

Introns, which are non-coding regions interspersed between coding regions of a gene, are absent from mitochondrial DNA.





## SEQUENCE RELATED DATA

Specimen Name : *Hoplostethus atlanticus*  
 Specimen Accession : BW-A304  
 Sequence ID : COAD04\_04

Translation Matrix : Vertebrate Mitochondrial Code  
 Electropherogram : [Reverse](#)  
[Forward](#)

Promoter Name :	A - Count : 150	G - Count : 120
5' Primer :	T - Count : 189	C - Count : 196
3' Primer :		

Sequence Length :	653	AT - Content :	51.8%	GC - Content :	48.2%
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Nucleotide Sequence :

```

CCTATAATCTCGTATTGCTGCCGACGGCATGGTGGCACAGCCTTAAGCTACTCATCCGAOCTGACCTTAGCCA
ACCTGGGGCGCGCTCTAAGAGACGACCAAGATTTACAATGTTATTGTACACACATGCTTTGTAATAATTTCTTTAT
ATAAATAACCAATATGATGATGTTGTTTGCGAACTGACTTGTGTTCCCTTAATGATGGAGGACGCCGATGATAGCATTTCC
TGGAAATAAATAATATGAGCTTTGACTGCTTCCCCCTTCATTCCTTCCTGCTTCTGCTGCGGAACTTGAAOCCAG
GGCGCGAAACGGTGAACAGTTAACCGCCCGCTGAGGAAGAACTGOCGGACGGGGGCTCCGTAAGAOCTAACCAT
CTTTCTTCAACATCTGGCAGGTTCTCAATTCTGGGGCCATTAACTTCATTAACACCATTAAATAATGAAGCC
TCAAGGCAATTCCCACTATCAAAACCGCCCTGTTGATGATGCTTTAAATTACAGCACTGCTCCCTCTCTATCCT
AGGAGACCCATTATACCAACACCTGTT
  
```

Amino Acid Sequence :

```

MLYVPPGAVAGRCYGTALSLIRAKLSDQPGALLCGDQIYMWIVTAHAFVVIIFPFPMPPIIGGFGRWLVPPFAMIGAPWAF
PISMMMSFWLLPPSFILLSSSGVIEAGASTGTVVPPLAGHLAMAGASVPLTIFSLNLAGVSSILGAINFTTIDHED
PPAISQTQTPFLPVWVWLITAVLILSIPVLAAGITHLTDRLNLTFFPDAGGGCDPLTQHLP
  
```

Illustrative DNA barcode :



# SPECIMEN IDENTIFICATION - Search Results

Identifications are generated by searching the BOLD database for nearest neighbors using a global alignment. Distances are calculated using the K2P model.

Length Submitted : 564 bp

Aligned : 99.63 %

Gaps : 0

Blast Sequence

## Identification Summary :

Taxonomic Level	Taxon Assignment	Probability of Placement (%)
Phylum	Chordata	100
Class	Actinopterygii	100
Order	Perciformes	100
Family	Centropomidae	100
Genus	Lates	100
Species	niloticus	100

## Distance Summary :

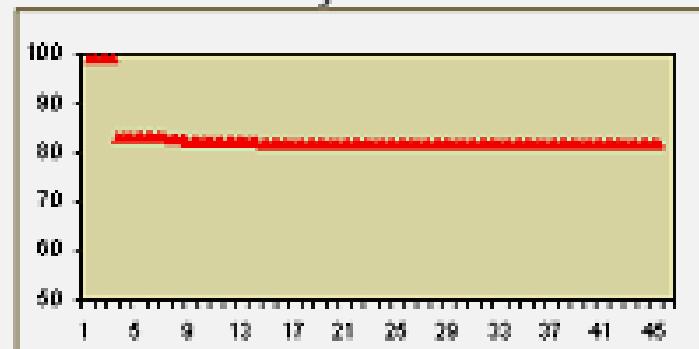


Figure is based on similarity scores of the top 50 matches

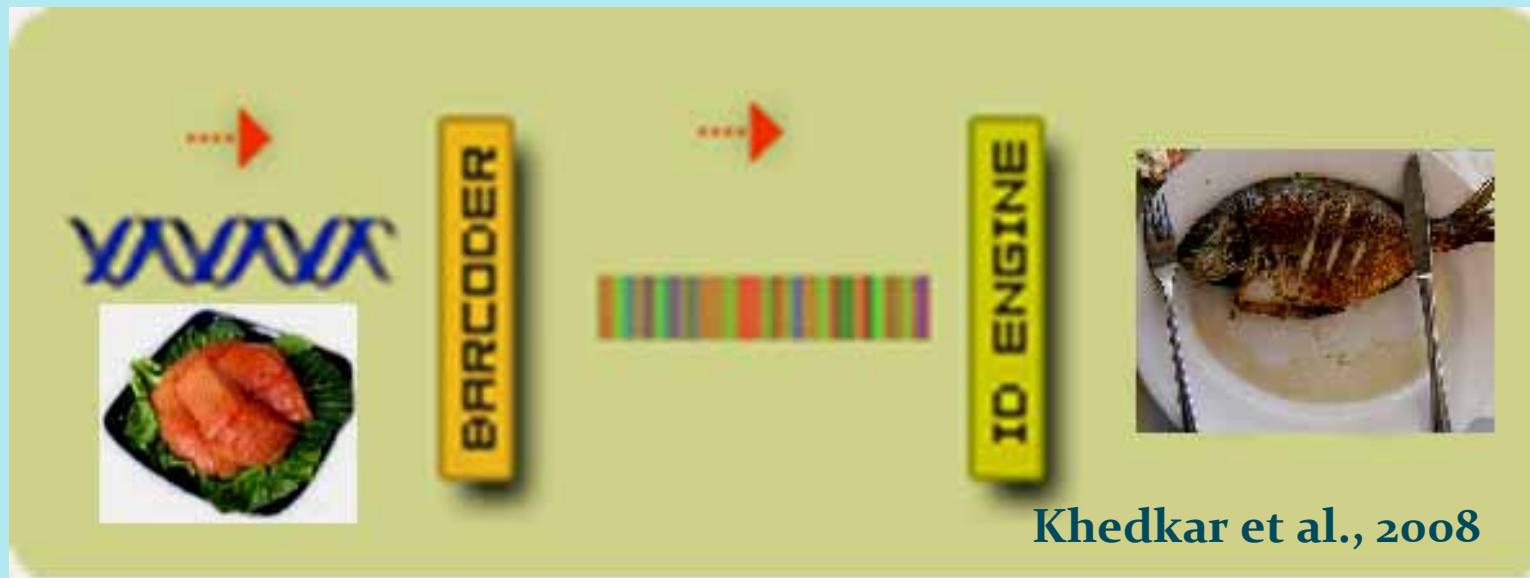
## TOP 10 Matches :

Phylum	Class	Order	Family	Genus	Species	Specimen Similarity (%)
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	niloticus	100
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	niloticus	100
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	niloticus	100
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	niloticus	100
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	niloticus	100
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	calcarifer	82.77
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	calcarifer	82.59
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	calcarifer	82.59
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	calcarifer	82.59
Chordata	Actinopterygii	Perciformes	Centropomidae	Lates	calcarifer	82.59

Generate NJ tree of 100 nearest taxa

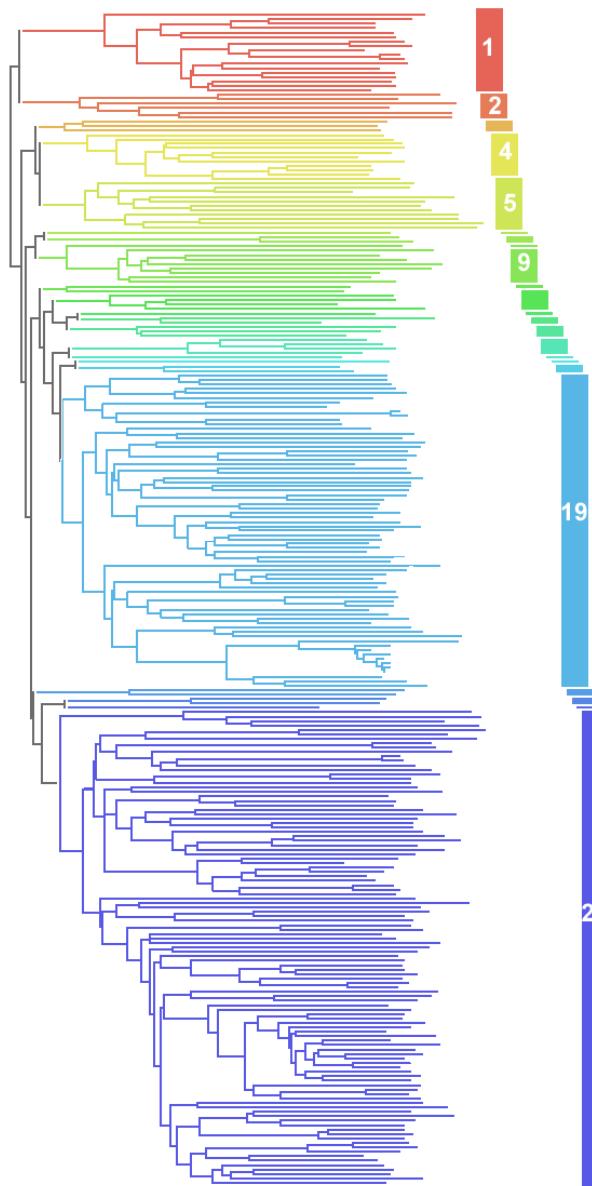
Paul Hebert Centre for DNA Barcoding & Biodiversity Studies





# Can barcodes aid understanding history of animal and plant species?

Neighbor-joining tree of COI barcodes  
for 260 species of North American birds



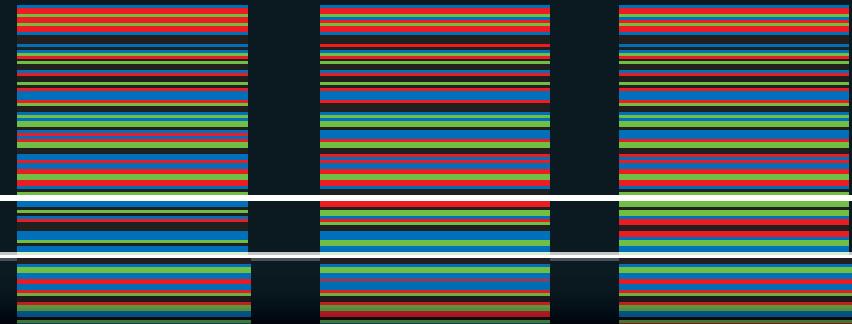
## Avian Orders

1. Anseriformes Ducks, Geese
2. Galliformes Grouse, Quail
3. Gruiformes Rails
4. Falconiformes Hawks
5. Strigiformes Owls
6. Cuculiformes Cuckoos
7. Apodiformes Hummingbirds
8. Coraciformes Kingfishers
9. Piciformes Woodpeckers
10. Pelicaniformes Pelicans, allies
11. Ciconiiformes Herons
12. Apodiformes Swifts\*
13. Gaviiformes Loons
14. Procellariiformes Petrels
15. Falconiformes Falcons\*
16. Gruiformes Cranes\*
17. Ciconiiformes Vultures\*
18. Podicipediformes Grebes
19. Charadriiformes Shorebirds
20. Columbiformes Doves
21. Caprimulgiformes Nightjars
22. Psittaformes Parrots
23. Passeriformes Passerines

\*4/260 (1.5%) of lineages differ from traditional taxonomic placements at the ordinal level



Shown here are  
300 base-pair  
segments of the CO1 gene  
for humans,  
chimpanzees  
and gorillas.



HUMAN

CHIMPANZEE

GORILLA



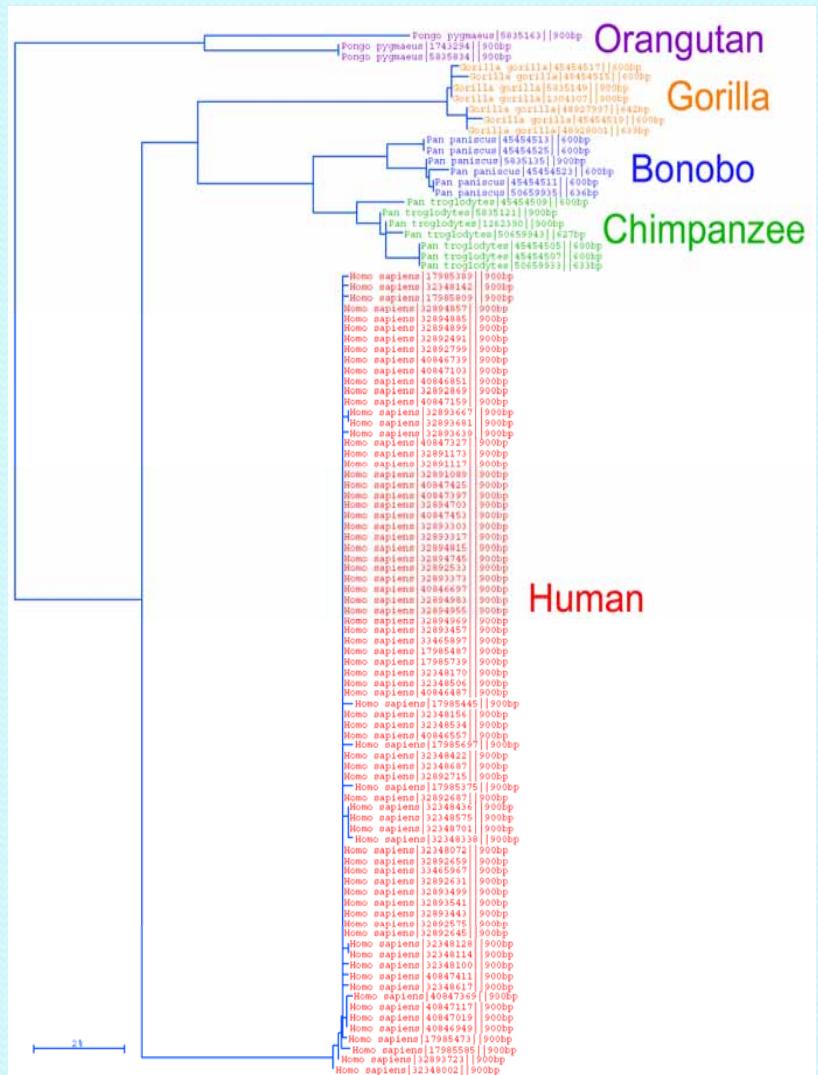
DNA code "letter"

C A T G

location of  
base-pair difference



Barcodes affirm the unity of the species *Homo sapiens*. Comparisons show we differ from one another by only 1 or 2 nucleotides out of 648, while we differ from chimpanzees at 60 locations and gorillas at 70 locations.



## UPC Codes

The Universal Product Code system developed by the industrial sector to brand retail items employs 10 options at each of 11 positions to create 100 billion alternates.

## DNA Codes

Just like UPC barcodes, the DNA sequences within each species are unique.

A run of 15 nucleotides, with 4 options at each position, creates the possibility of 1 billion codes, a hundred-fold excess over the estimated number of animal species.



FISHBOL.org: Fish Barcode of Life Initiative - Microsoft Internet Explorer

File Edit View Favorites Tools Help

Back Forward Stop Home Search Favorites Print Mail Help

Address: http://www.fishbol.org/people.php#india

pdf Go 0 PDF Links Customize Links Free Hotmail Windows Windows Marketplace Windows Media

Barcode of Life Initiative :: Partner

# FISH-BOL

## Fish Barcode of Life Initiative

Home Vision People Progress Checklists News & Updates Links

## People

Home > People

Listed below are the people involved in running FISH-BOL, both the administrative heads and the members of the various working groups. Please click on the members names to retrieve their contact information.

### Governance of FISH-BOL

The global administration of the Fish Barcode of Life Initiative is led by;

- Co-Chair: Paul Hebert
- Co-Chair: Bob Ward
- Campaign Coordinator: Robert Hanner

### Regional Working Groups

FISH-BOL has been broken into 10 regional working groups, the contacts for each group are listed below;

India Top

The administration of the Indian Regional Working Group is led by;

- Chair: Wazir Lakra

The members are;

- Achamveettil Gopalakrishnan
- Pallipuram Jayasankar
- G D Khadkar
- Wazir Lakra
- Mahender Verma

North America

**G D Khadkar** X

**Title:** Ph.D.

**Address:** Aquaculture Research Laboratory, Department of Zoology  
Dr.B.A.M. University  
Aurangabad, 431 004  
India

**Tel:** +91-240-2401197

**Fax:** +91-240-2403335

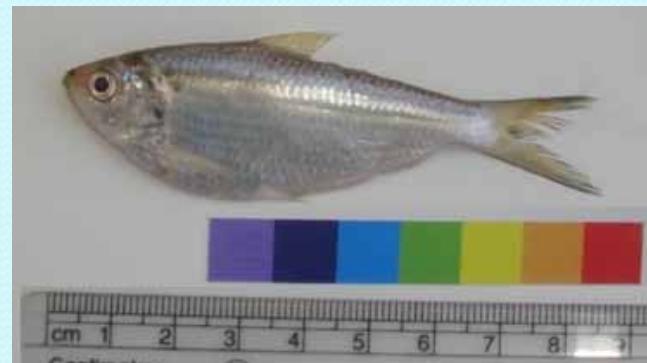
**Description:**

Internet 1:21



Fin Clip sampling





Digital record of species





Identifier :	<a href="#">Gulab D. Khedkar</a>
phylum :	Chordata
class :	Actinopterygii
order :	Perciformes
family :	Polynemidae
genus :	Eleutheronema
species :	<a href="#">Eleutheronema tetradactylum</a>
Taxonomy Note	NA

### Nucleotide Sequence

Residues: 652 CCTCTATCTAATCTTGGGGCATGAGCTGGATAGTAGGGACCGCTCTAACGCCCTATT  
Comp. A: 142 CGTGCAGAACTAAGCAGCCGGCGACTTCTAGGTGACGATCAGATTATAATGTTATCGT  
Comp. G: 133 ATGAGCTTTGACTCCTCCGCCCTTTCTTCTTCTCCTAGTCTCTCTGGGGTAGAGGC  
Comp. C: 184 TGGAGCCGGGACAGGATGAAGTGTCTACCCCCCTTAGCAGGGAACCTTGCCCACGCAGG  
Comp. T: 193 ACCATCGTGGACCTAACCAATTTCCTCCACCTAGCAGGGTATCCTCAATCCTGGGG  
Ambiguous : o CTATTAATTATTACAACTCCTGAATATAAAACCTGCCGCCCTCAATATACCAGCTTCC  
CTTATTGTCTGAGCTGTTCTAGTCACAGCGTACTGCTTCTTGTCTCTCCCCGTTCTGGC  
CGCTGGAATTACCATACTATTGACAGACCGAAACCTCAATACTGCATTCTTGACCCTGCG  
GGCGGAGGAGACCAATTCTTACCAACACCTA



# Acknowledgement

**Government of India, Ministry of Food Processing Industries**

**Genome Canada, Ontario**





Thank you

