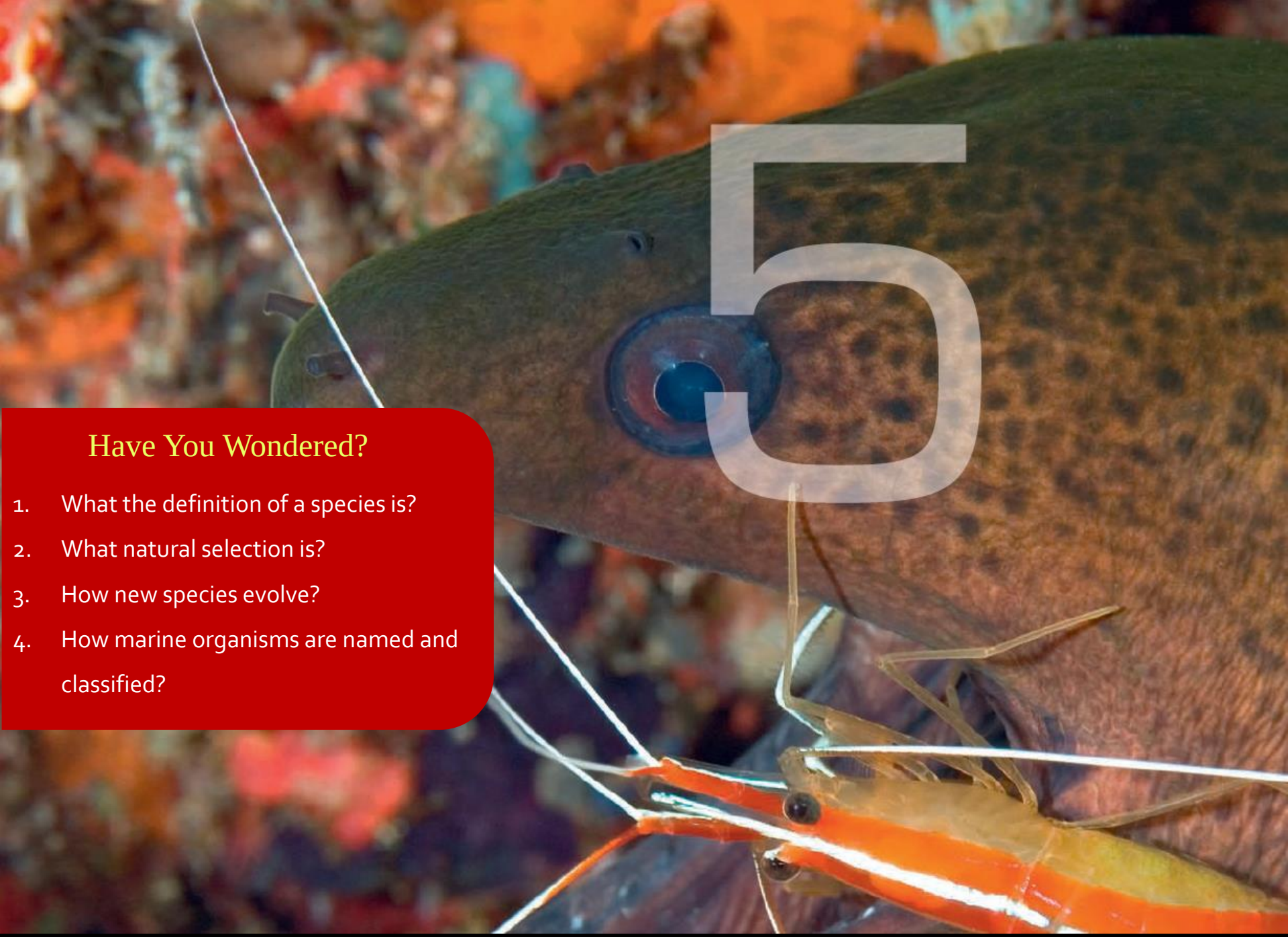


5

Have You Wondered?

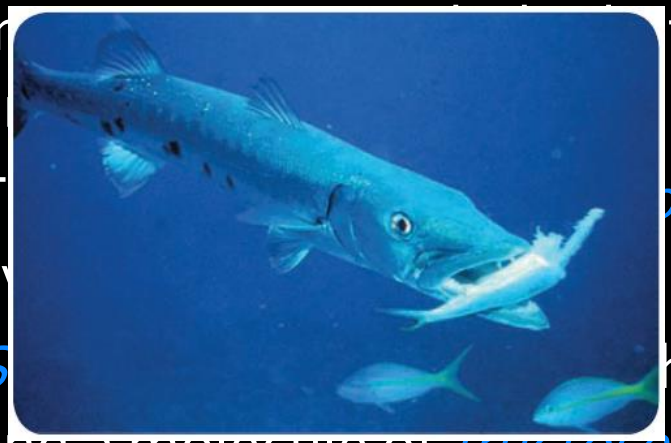
1. What the definition of a species is?
2. What natural selection is?
3. How new species evolve?
4. How marine organisms are named and classified?

Biological Concepts

- Survival and reproduction
- Living organisms are composed of chemical compounds.
- There are four major classes of macromolecules in living organisms: carbohydrates, lipids, proteins, and nucleic acids.

Carbohydrate

- The molecules known as *carbohydrates* contain the elements carbon, hydrogen, and oxygen, frequently in a ratio of 1:2:1, or CH_2O, thus the name carbohydra . The m tes in livi sugars a
- T se is the e for li
- S hat conta glucose and a molecule of *fructose*. Marine se sucrose to transport sugars within t
- *Lactose* supplies much of the energy that newborn mammals require.



Carbohydrate

- *Star*
mole
micr
ms to
store
- *Glyc*
micr
- *Cellulose* is an example of a structural polysaccharide. It is found in the cell walls of plants, seaweeds, and some microorganisms
- *Chitin* is a polymer composed of modified glucose molecules and is also strong and durable. Chitin is in the cell walls of fungi and the hard exterior skeletons of some marine animals such as crabs and lobsters

Lipids

- Lipids composed primarily of carbon and hydrogen. Marine organisms use simple *triglycerides* to store energy, to cushion vital organs, and to increase buoyancy. Homeothermic animals use *triglycerides* as insulation to trap heat.
- *Phospholipid* is the major structural component of membranes that surround cells and some of the internal components of cells.
- *Steroids* are lipids that function as chemical messengers within the bodies of animals.
- *Waxes coat* the exposed surfaces of some marine plants and seaweeds and act as a water barrier. Waxes are also found in the body coverings of some marine animals and in the ear openings of some marine mammals.

Fatty Acid Analysis

Fatty Acid Analysis

浮游动物体内的脂类物质绝大部分来源于食物(Goulden *et al*, 1990)。而海洋微藻的脂肪酸组成特征在不同门类之间具有显著的差异(李春颖等, 2008; Volkman *et al*., 1998), 可以被用来指示海洋微藻的种类组成。如甲藻中的 18:4ω3 和硅藻中的 16:1ω7 已被广泛证实可作为硅藻和甲藻的特征脂肪酸用于指示自然水体颗粒悬浮物中的硅藻和甲藻成分(Claustre *et al*, 1988/1989; Kattner *et al*, 1983; 吕淑果等, 2009)以及硅藻和甲藻脂肪酸沿食物链的传递过程(Fraser *et al*, 1989)。在摄食者中只有植食性的桡足类能大量合成 20 碳和 22 碳的脂肪酸和脂肪醇类物质(Dalsgaard *et al*, 2003)。因此, 20:1 和 22:1 脂肪酸不仅可以作为桡足类摄食浮游植物的指标, 还能作为高营养级摄食植食性桡足类的指标(Falk-Petersen *et al*, 1987)。另外, Σ15 和 Σ17 脂肪酸则可以用来指示饵料中的细菌贡献(Budge *et al*, 1998, 2001)。关于长江口海区动物

金鑫等，2012，海洋与湖沼

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PLOS ONE

Stable Isotope and Signature Fatty Acid Analyses Suggest Reef Manta Rays Feed on Demersal Zooplankton

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Abstract

Assessing the trophic role and interaction of an animal is key to understanding its general ecology and dynamics. Conventional techniques used to elucidate diet, such as stomach content analysis, are not suitable for large threatened marine species. Non-lethal sampling combined with biochemical methods provides a practical alternative for investigating the feeding ecology of these species. Stable isotope and signature fatty acid analyses of muscle tissue were used for the first time to examine assimilated diet of the reef manta ray *Manta alfredi*, and were compared with different zooplankton functional groups (i.e. near-surface zooplankton collected during manta ray feeding events and non-feeding periods, epipelagic zooplankton, demersal zooplankton and several different zooplankton taxa). Stable isotope $\delta^{15}N$ values confirmed that the reef manta ray is a secondary consumer. This species had relatively high levels of docosahexaenoic acid (DHA) indicating a flagellate-based food source in the diet, which likely reflects feeding on DHA-rich near-surface and epipelagic zooplankton. However, high levels of ω6 polyunsaturated fatty acids and slightly enriched $\delta^{13}C$ values in reef manta ray tissue suggest that they do not feed solely on pelagic zooplankton, but rather obtain part of their diet from another origin. The closest match was with demersal zooplankton, suggesting it is an important component of the reef manta ray diet. The ability to feed on demersal zooplankton is likely linked to the horizontal and vertical movement patterns of this giant planktivore. These new insights into the habitat use and feeding ecology of the reef manta ray will assist in the effective evaluation of its conservation needs.

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Competing Interests: This study was funded by the Australian Research Council Linkage Grant (LP110100712), Earthwatch Institute Australia, Sea World Research and Rescue Foundation Inc. and Sibelco Pty Ltd. Field work was supported by Casa Barry Lodge and Peri-Peri Divers in Mozambique, and Lady Elliot Island Eco Resort and Manta Lodge and Scuba Centre in Australia. This does not alter the authors' adherence to all the PLOS ONE policies on sharing data and materials.

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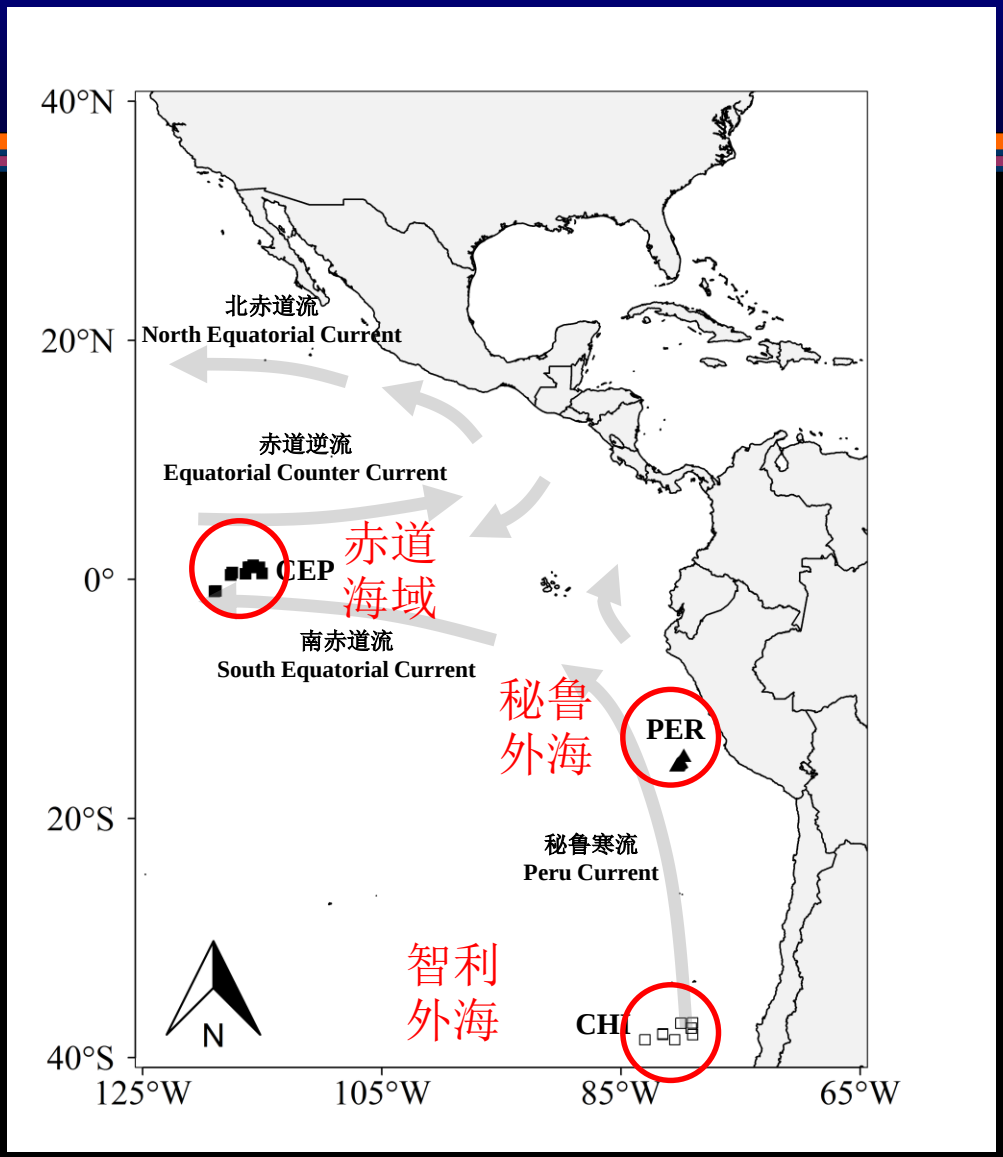
Introduction

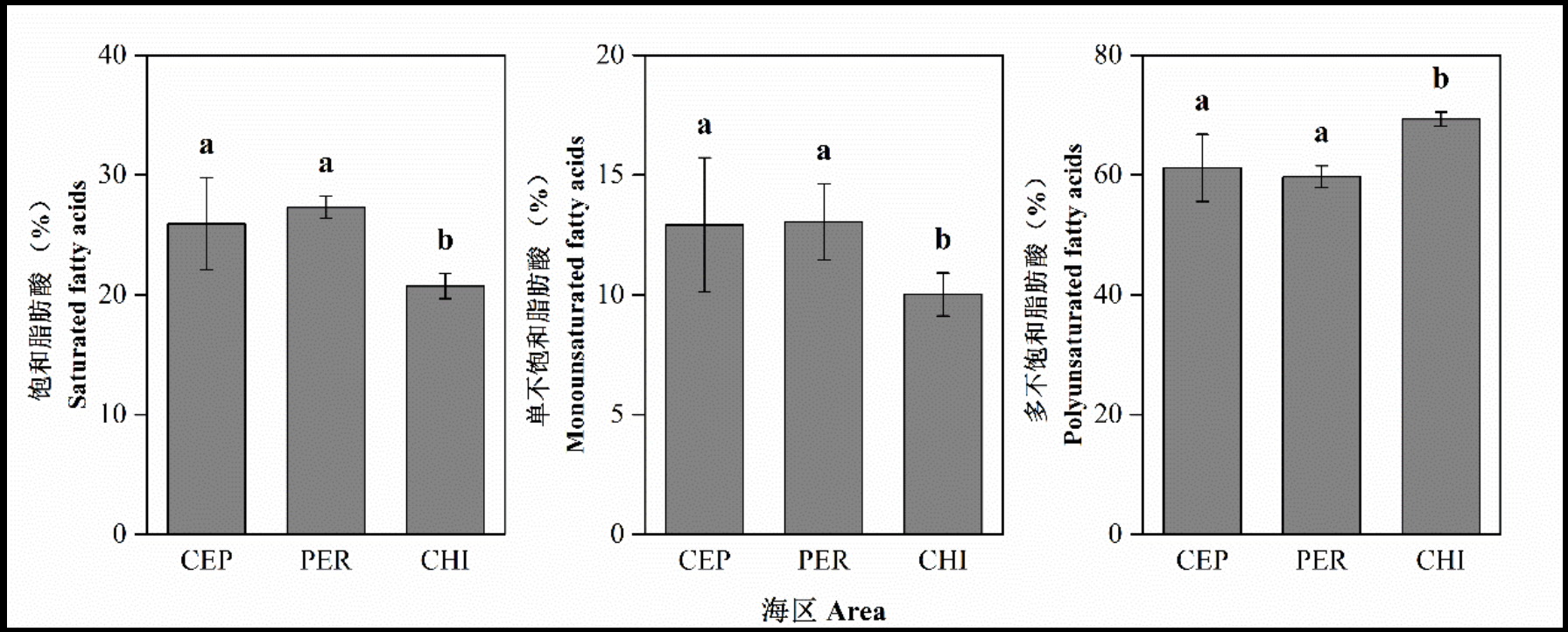
Information on the diet and trophic position of an animal can improve ecological understanding of the underlying drivers of its movements and its role within the ecosystem. Such knowledge can also support conservation plans for areas where the temporal and spatial abundance and distribution of prey are understood [1–3]. Stomach content analysis is the conventional approach used to assess a species' diet [4] and has many advantages; however, it also has several shortcomings. First, this technique only provides a 'snapshot' of recent feeding and may not accurately reflect the composition of prey items that contribute most significantly to its general diet. This technique my also not necessarily account for ontogenetic or seasonal shifts in diet nor regional variability in the diet of a species. For a comprehensive understanding of a species' diet, many specimens must be examined with samples from different seasons, locations, size classes and sexes. Sample collection therefore becomes challenging for widely distributed and wide-ranging species that may feed in numerous habitat types over large geographic areas. Second, stomach content analysis is heavily biased towards items resistant to digestion such as bones, exoskeletons, chelae and eyeballs [5]. Last, obtaining stomachs from large and threatened marine species is often difficult and killing animals for this purpose is ethically questionable.

The reef manta ray *Manta alfredi* (Krefft, 1868) is a large planktivorous elasmobranch with a circumglobal distribution in tropical and subtropical waters [6]. The species is listed as globally Vulnerable to extinction on the IUCN Red List of Threatened Species, mainly due to new or expanding targeted fisheries [7]. Many of these fisheries are considered unsustainable due to the relative small native population sizes, likely limited exchange between subpopulations and conservative life history of the species

Couturier et al., 2013, PLOS ONE

College of Marine Sciences, Shanghai Ocean University


40°N
20°N
0°
20°S
40°S
125°W
105°W
85°W
65°W
北赤道流
North Equatorial Current
赤道逆流
Equatorial Counter Current
CEP
赤道
海域
南赤道流
South Equatorial Current
秘鲁
外海
PER
秘鲁寒流
Peru Current
智利
外海
CHI
N


饱和脂肪酸 （%）
Saturated fatty acids
40
30
20
10
0
a
a
b
CEP
PER
CHI
单不饱和脂肪酸 （%）
Monounsaturated fatty acids
20
15
10
5
0
a
a
b
CEP
PER
CHI
多不饱和脂肪酸 （%）
Polyunsaturated fatty acids
80
60
40
20
0
a
a
b
CEP
PER
CHI
海区 Area

Protein

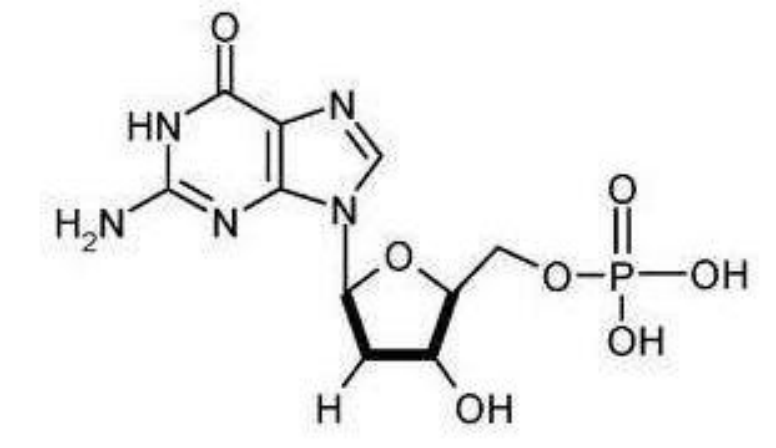
- Proteins are polymers made up of basic units called *amino acids*. Twenty different amino acids make up the various proteins found in living organisms. Within cells, individual amino acids are assembled into chains called *polypeptides*.
- The primary structural components of animals, muscles and connective tissues, are composed of protein.
- Proteins known as *enzymes* are essential for life. Enzymes are biological catalysts that speed up the rate of chemical reactions, allowing metabolism to function efficiently.
- Some proteins, such as *hemoglobin*, transport chemicals within organisms, whereas others store chemicals.

Protein and their functions

Type of Protein	Function
Enzymes	Biological catalysts that speed up the rate of chemical reactions in cells
Structural Proteins	Make up body parts of animals such as hair, skin, scales, tendons, cartilage
Contractile Proteins	Make up muscle
Messenger Proteins	Send signals from one cell to another and from organ to organ
Transport Proteins	Transport important substances such oxygen and fatty acids
Storage Proteins	Store important materials in cells, such as iron
Antibodies	Protect animals from foreign proteins and disease-causing microbes
Toxins	Help to capture prey and protect animals from predators

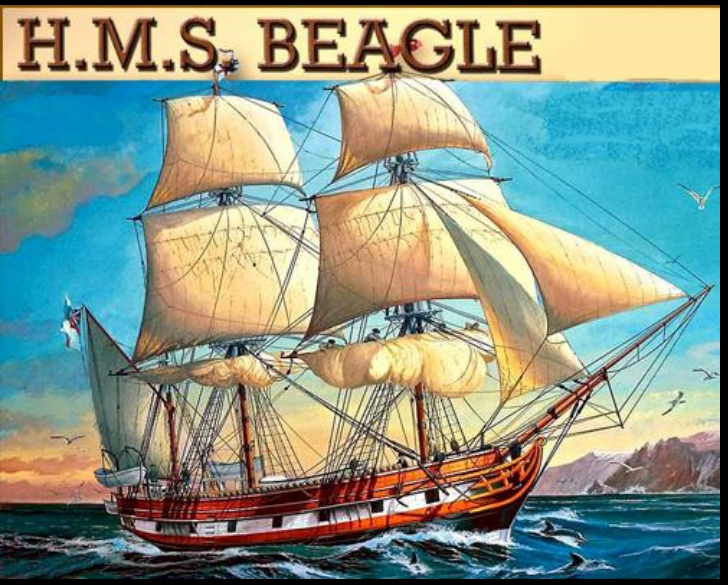
Nucleic acids

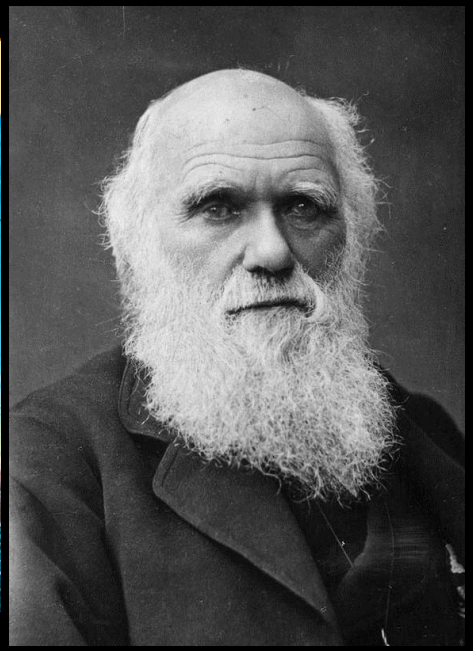
- *Nucleic acids* are polymers of molecules called nucleotides.
- Each nucleotide is composed of a five-carbon sugar, a nitrogen containing base, and a phosphate group. Two types of nucleic acids are found in living organisms: *deoxyribonucleic acid* (DNA) and *ribonucleic acid* (RNA).

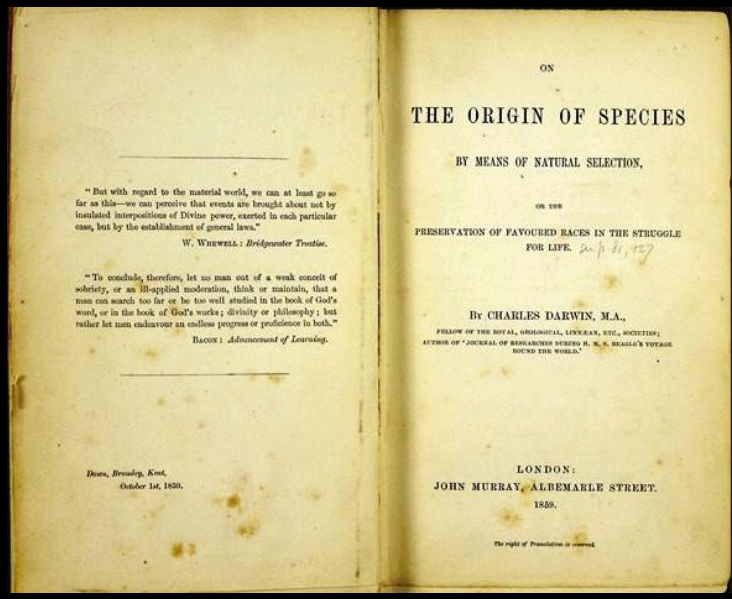


Evolution and Natural Selection

- *Natural selection* is the mechanism that explains why organisms that possess variations best suited to their particular environment exhibit a better survival rate and reproductive capacity than do less well-suited organisms.






"But with regard to the material world, we can at least go so far as this—we can perceive that events are brought about not by insulated interpositions of Divine power, exerted in each particular case, but by the establishment of general laws."
W. Whewell: *Bridgewater Treatise.*

"To conclude, therefore, let no man out of a weak conceit of sobriety, or an ill-applied moderation, think or maintain, that a man can search too far or be too well studied in the book of God's word, or in the book of God's works; divinity or philosophy; but rather let men endeavour an endless progress or proficience in both."
Bacon: *Advancement of Learning.*

Down, Bromley, Kent,
October 1st, 1859.

ON

THE ORIGIN OF SPECIES

BY MEANS OF NATURAL SELECTION,

OR THE

PRESERVATION OF FAVOURED RACES IN THE STRUGGLE FOR LIFE.

By CHARLES DARWIN, M.A.,

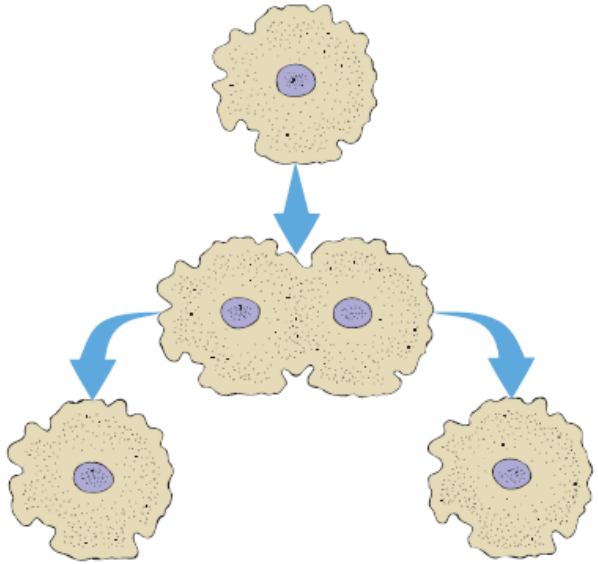
FELLOW OF THE ROYAL, GEOLOGICAL, LINNEAN, ETC., SOCIETIES;
AUTHOR OF 'JOURNAL OF RESEARCHES DURING H. M. S. BEAGLE'S VOYAGE ROUND THE WORLD.'

LONDON:
JOHN MURRAY, ALBEMARLE STREET.
1859.

Evolution

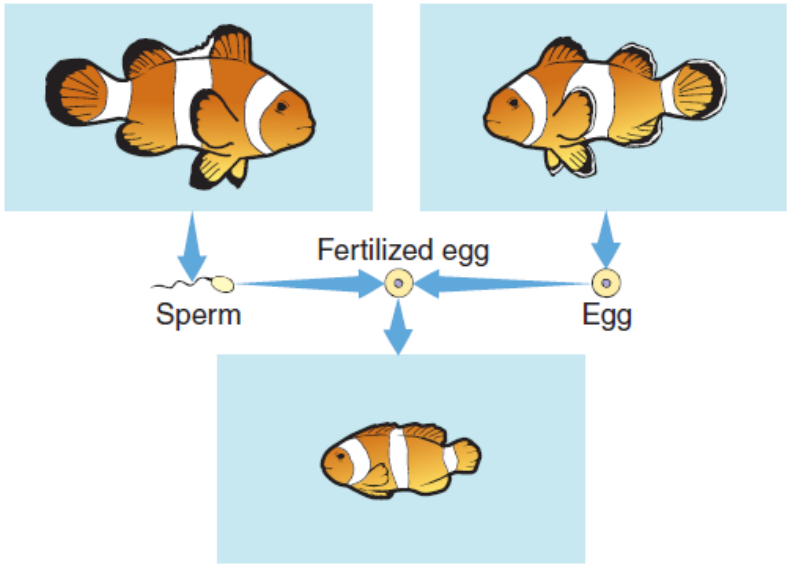
- The process of evolution operates without any ultimate goal, selecting those forms of organisms that are best able to survive and reproduce under the environmental conditions in which they live.
- Note that natural selection does not have the ability to cause variations that are better suited than others. The variations that occur are due to chance mutations. Only after the variations appear can they be affected by natural selective forces.

Evidence for Evolution



ASEXUAL REPRODUCTION

(a)

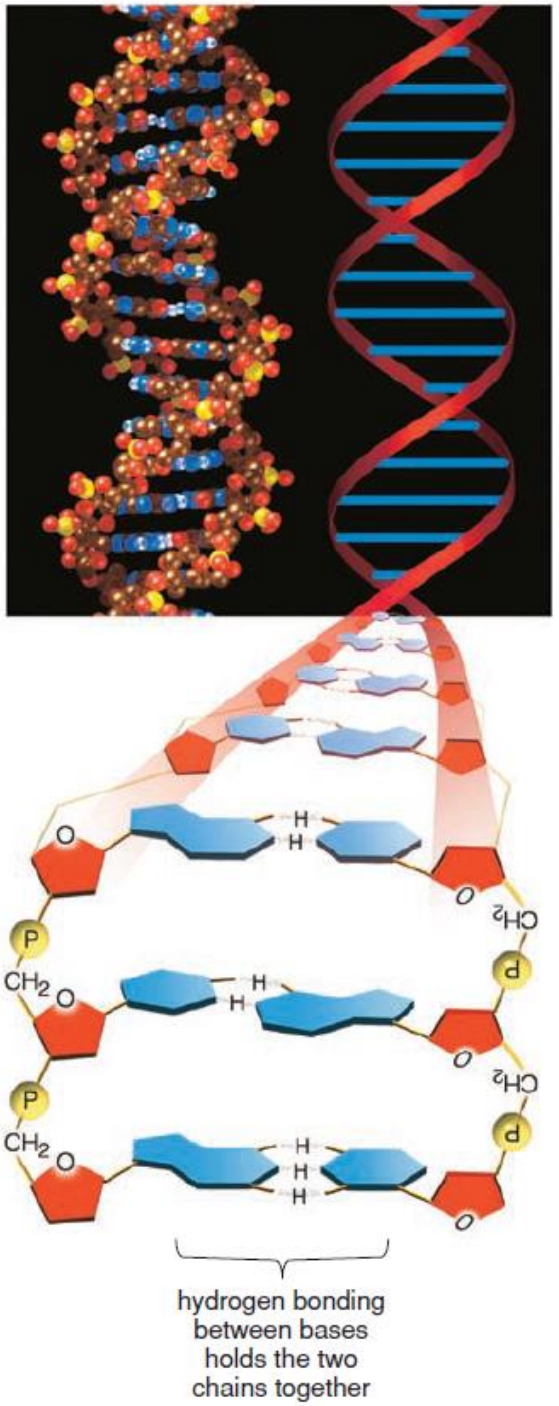


SEXUAL REPRODUCTION

(b)

DNA

- The nucleic acid DNA is composed of units called nucleotides. Each nucleotide contains the five-carbon sugar deoxyribose , a nitrogen-containing base, and a phosphate group.
- The nucleotides are linked together by the phosphate groups and form long chains. Two chains are wound together to form the characteristic double helix.



DNA function

- DNA contains an organism's genetic material, or genes, and is capable of copying itself so that the genes can be passed from one generation to the next.
- Encoded in the genes are directions for synthesizing the various proteins that an organism needs. The proteins are responsible for an organism's appearance and control its function, growth, and reproduction.
- In many cells most of the DNA is located in a specialized structure called the nucleus. In addition, smaller amounts of DNA are found in other parts of the cell such as mitochondria and chloroplasts.

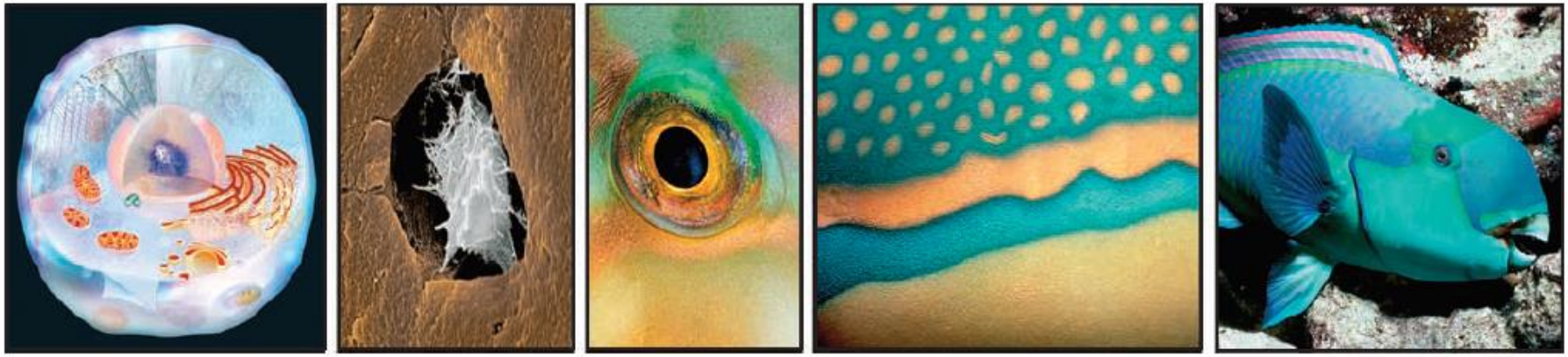
How does DNA work?

RNA

- RNA molecules are usually single-stranded and can come in different forms that have evolved for specific roles in the cell. RNA functions in protein synthesis. In this process the information carried by individual genes is copied, or transcribed, from DNA onto a molecule of RNA known as messenger RNA (mRNA). The message carried by the mRNA is decoded, or translated, by a cellular structure known as a ribosome, composed of protein and another type of RNA called ribosomal RNA (rRNA). The *ribosome* synthesizes protein by connecting the appropriate amino acids that are brought to the ribosome by a third type of RNA called transfer RNA (tRNA). The proteins produced by this process make new cell parts and enzymes that give the cells their characteristics

DNA and RNA

Protein synthesis



a cell → **b tissue** → **c organ** → **d organ system** → **e multicelled organism**

Smallest unit that can live and reproduce on its own or as part of a multicelled organism. It has an outer membrane, DNA, and other components.

Structural unit of certain types and proportions of cells interacting in some task. Many cells (*white*) made this bone tissue from their own secretions.

Structural unit of two or more tissues interacting in some task. A parrotfish eye is a sensory organ used in vision.

Organs interacting physically, chemically, or both in some task. Parrotfish skin is an integumentary system with tissue layers, organs such as glands, and other parts.

Individual made of different types of cells. Cells of most organisms, including this Red Sea parrotfish, are organized as tissues, organs, and organ systems.

Species

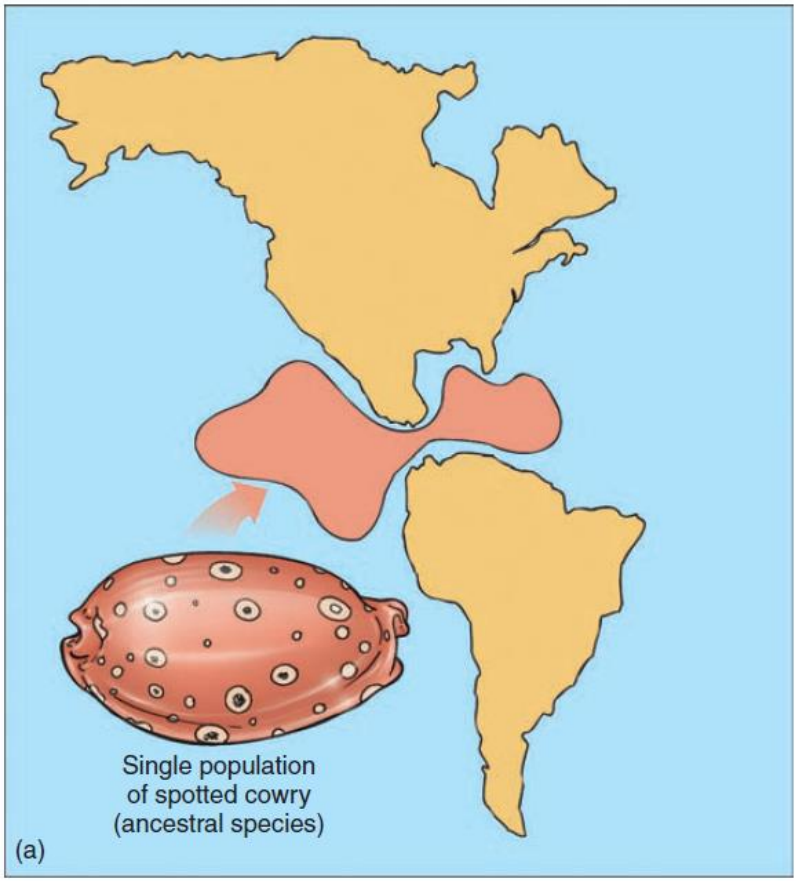
- Based on morphology

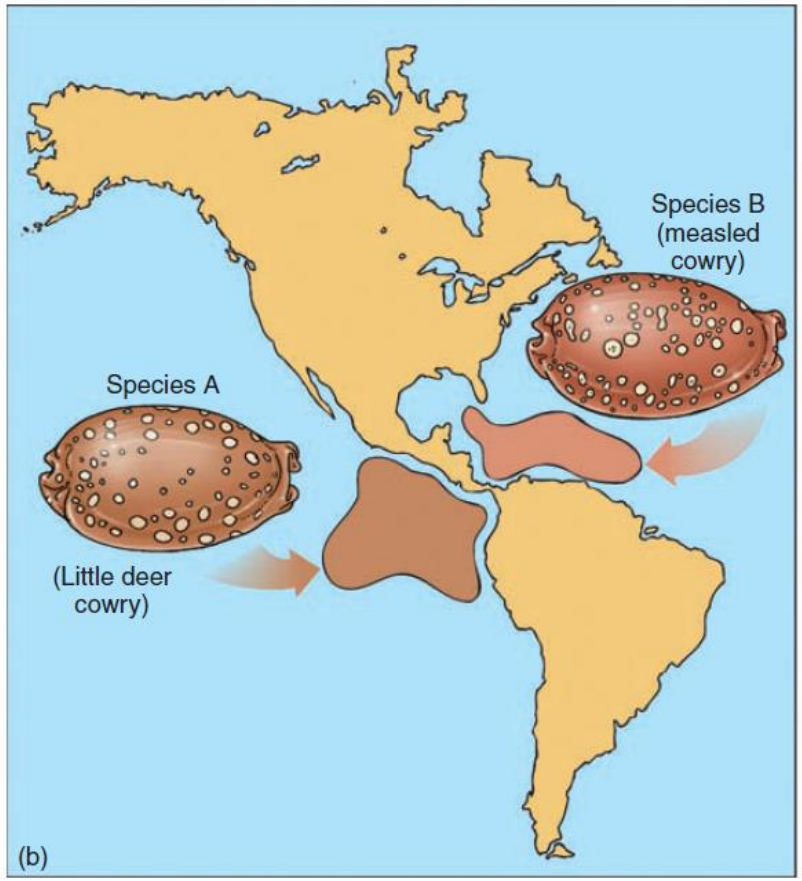


Species

- A *species* is one or more populations of potentially interbreeding organisms that are reproductively isolated from other such groups.
- *Reproductive isolation*: occurs when the members of different species are not in the same place at the same time or are physically incapable of breeding. Including habitat isolation; anatomical isolation; behavioral isolation; temporal isolation; biochemical isolation.

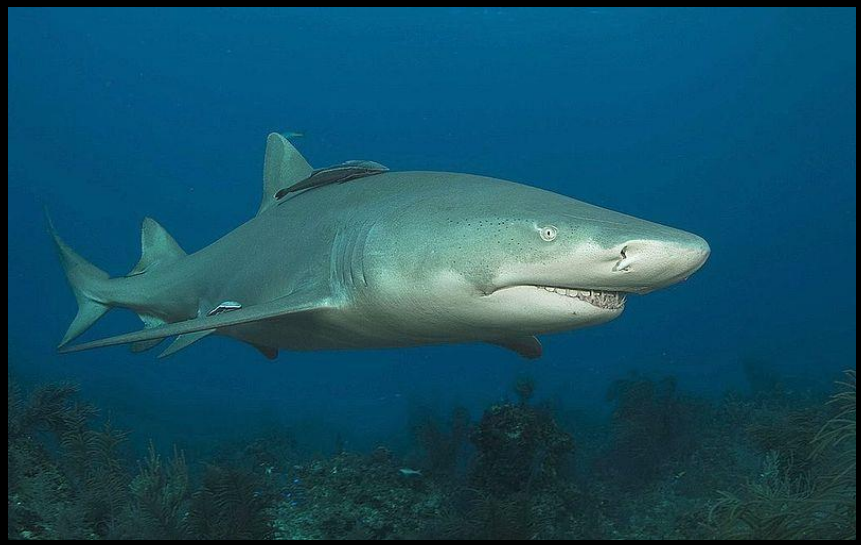
Process of Speciation





Lemon shark

- The lemon shark gets it name from its yellowish color, especially on its underside. It is related to tiger and bull sharks and can grow up to 3 m in length
- Apart from the color, you can recognize a lemon shark by its two similarly sized dorsal fins. In most shark species, the first dorsal fin is much bigger.



Lemon shark

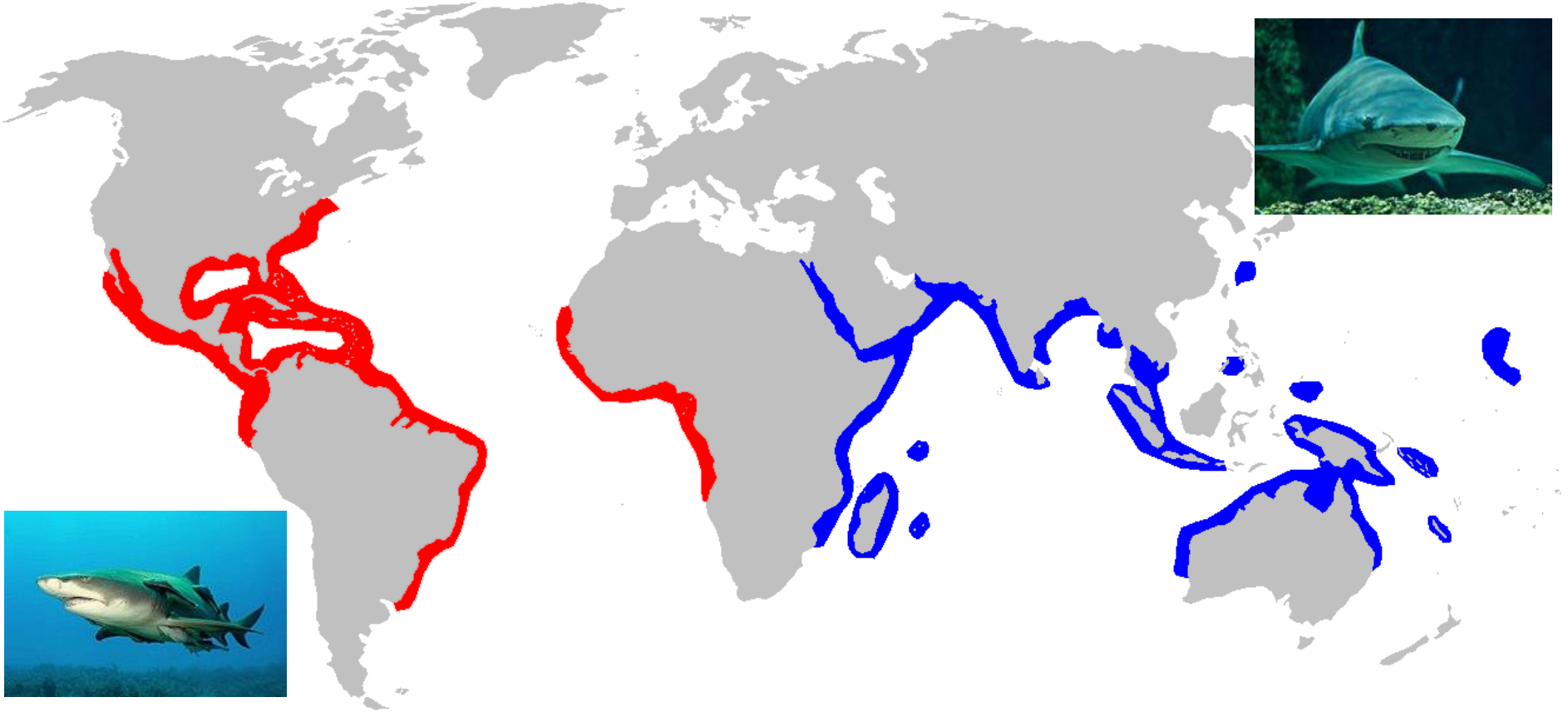
Negaprion（柠檬鲨属）

- 1. 短吻柠檬鲨 *Negaprion brevirostris* 分布于美洲和非洲西海岸
  2. 犁鳍柠檬鲨 *Negaprion acutidens*分布于印度-太平洋区，西起红海、非洲东岸，东至东南亚，北至日本，南至澳洲。中国南海、台湾南部海域也有分布。


Negaprion brevirostris

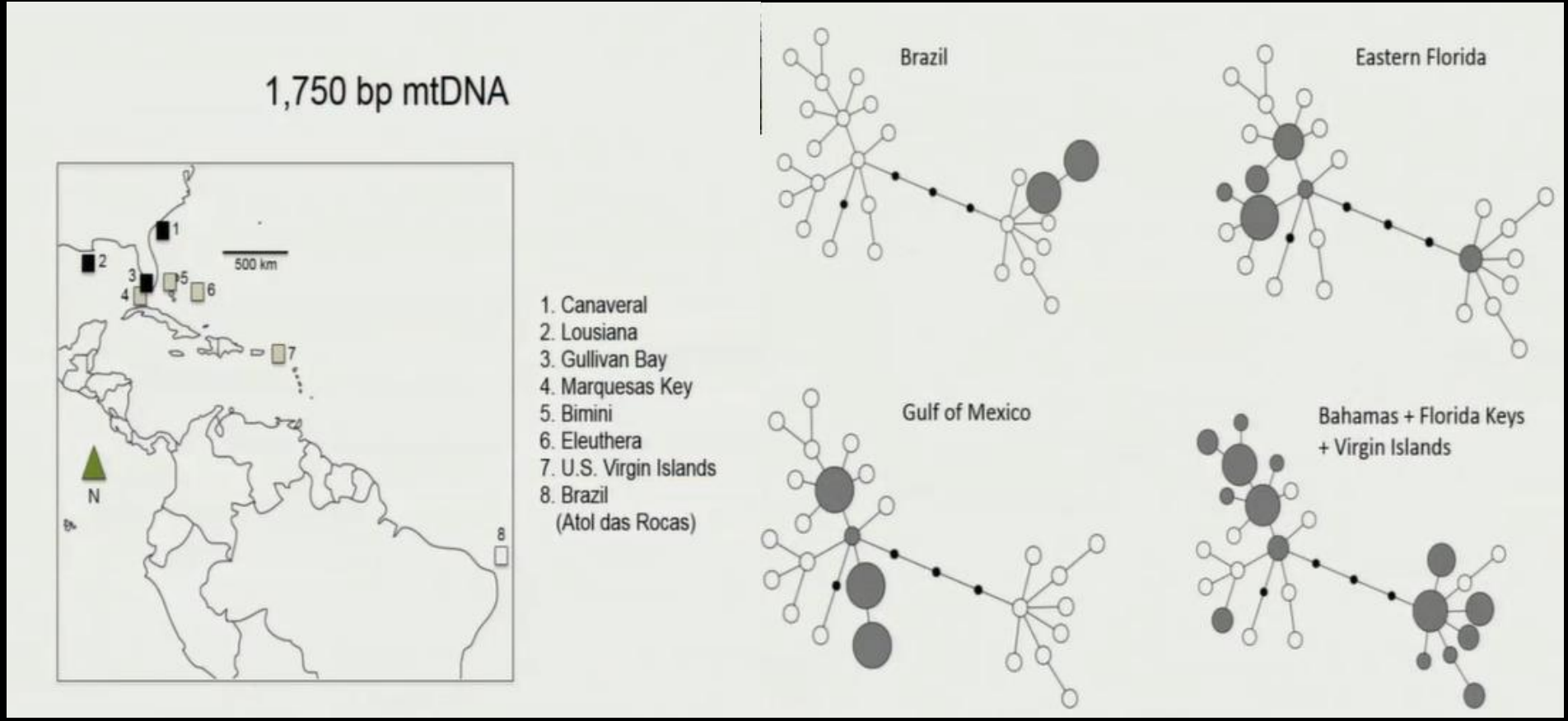

Negaprion acutidens

Negaprion（柠檬鲨属）



Lemon shark (*N. brevirostris*)

Lemon shark (*N. brevirostris*)

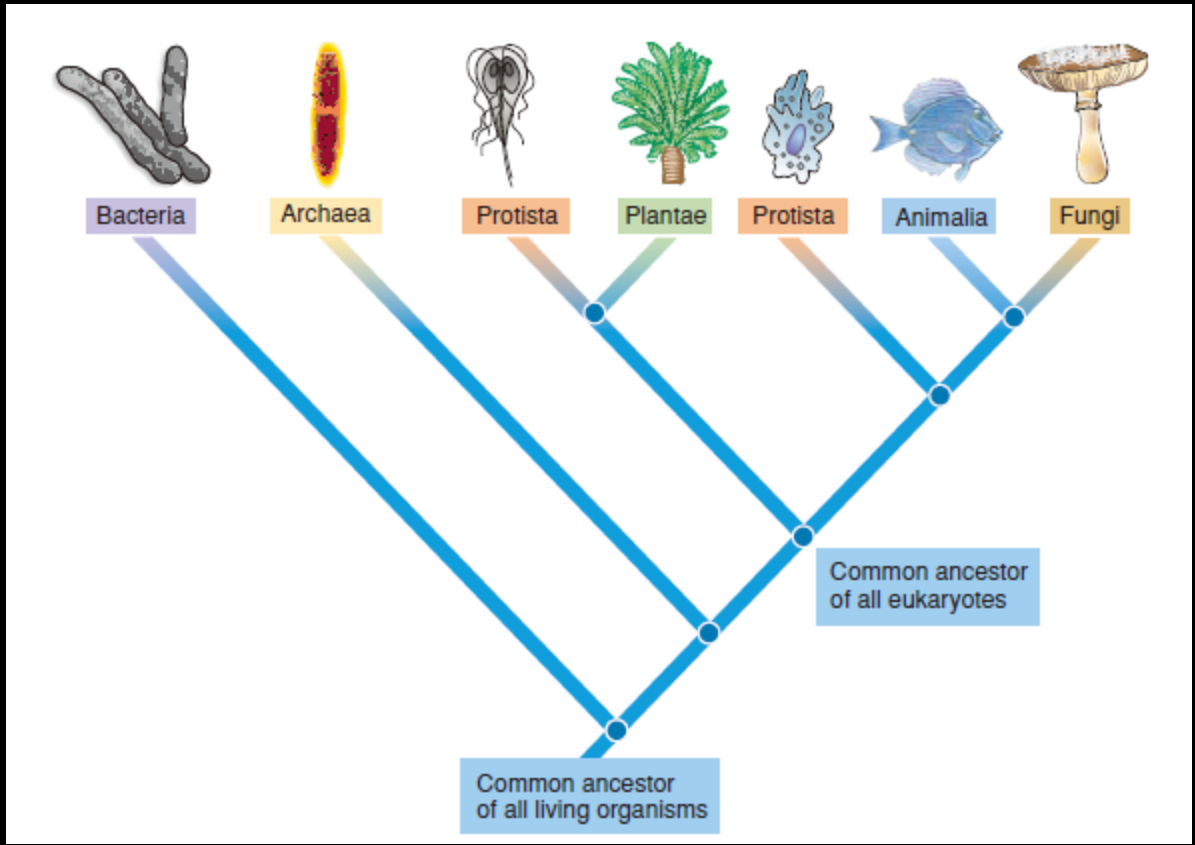


Newborn sharks

Lemon shark

Classification

- 界kingdom
- 门phylum
- 纲class
- 目order
- 科family
- 属genus
- 种species



Further reading

- Norris, R. D. 1992. So You Thought Extinction Was Forever? *Oceanus 35(4).*
- Price, P. W. 1996. *Biological Evolution. Philadelphia: Saunders* College Publishing.
- Quammen, D. 2007. A Passion for Order, *National Geographic* 211(6).
- Scheltema, R. S. 1996. Describing Diversity: Too Many New Species, Too Few Taxonomists, *Oceanus 39(1).*
- Stoeckle, M. 2003. Taxonomy, DNA, and the Bar Code of Life, *Bioscience 53(9).*

Points

If a cell lacked ribosomes, it would not be able to

A. Synthesize lipids
B. Digest food
C. Synthesize proteins
D. Form membranes
E. Signal to other cells

Darwin proposed the evolution occurs as the results of

A. Cosmic forces
B. Human intervention
C. Artificial selection
D. Natural selection
E. Inherent need

A population of potential interbreeding organisms that is reproductively isolated from other such populations defines

A. A kingdom
B. A community
C. A family
D. A genus
E. A species

If two species are in the same class, they must also be in the same

A. Family
B. Genus
C. Order
D. Phylum
E. Subspecies