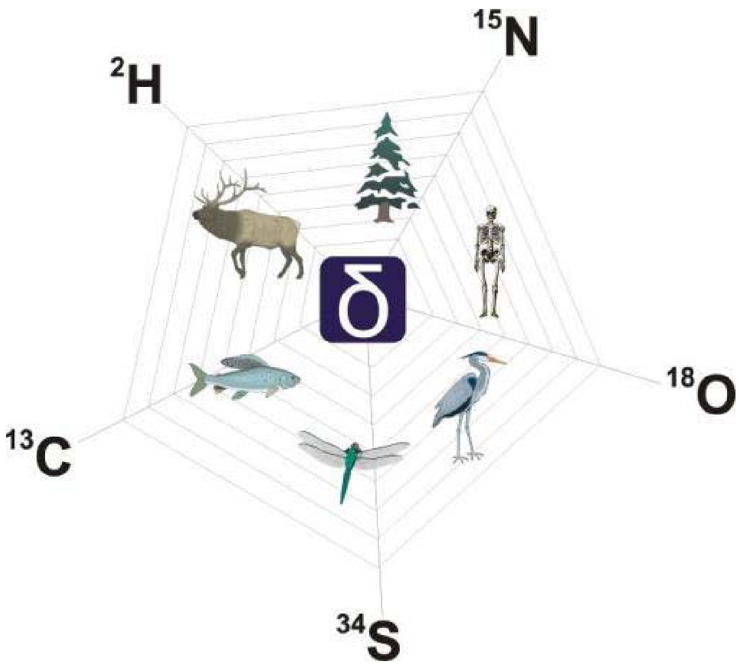
An overview of stable isotope applications to ecological and environmental studies

3rd Stable Isotope Course in Ecology and Environmental Sciences

Manuela G. Forero



ThermoFisher SCIENTIFIC

Estación Biológica Doñana CSIC

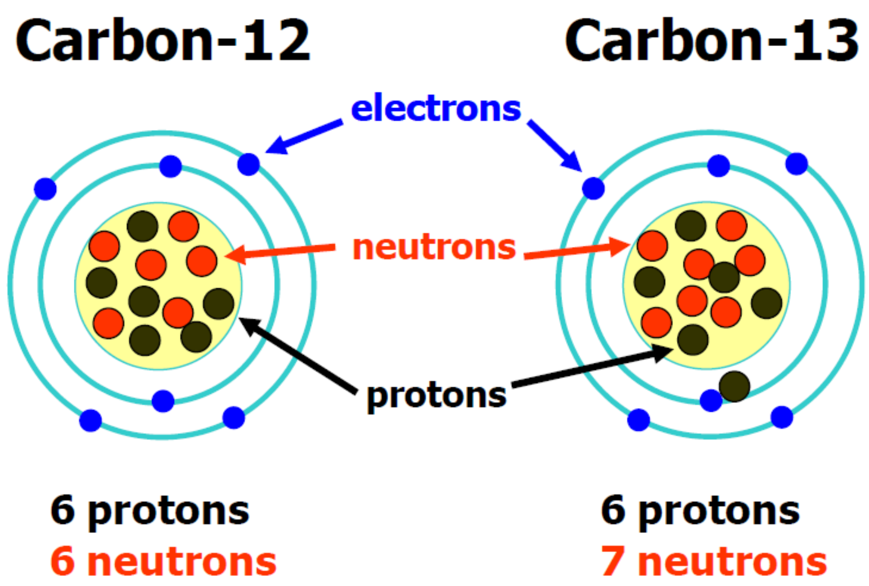
LIE EBD Laboratorio de Isótopos Estables Estación Biológica de Doñana

http://www.ebd.csic.es/lie/index.html

Stable Isotopes ***integrate ecological proccess***, ***indicate the presence and magnitute of them***, ***record*** the response of biological systems to environmental change and ***trace*** fundamental ecological processes

What is an isotope?

Frederick Soddy (1877-1956) originated the word isotope from the Greek words ***isos*** meaning "**equal**" (as in chemically non-separable) and ***topos*** meaning "**place**" or of the same place in the periodic table.



*"One of two or more atoms having the same atomic number (same protons) but different numbers of neutrons and therefore different **mass numbers**"*

All elements have more than one isotope

Periodic Table of the Elements

Main groups — Transition metals — Main groups

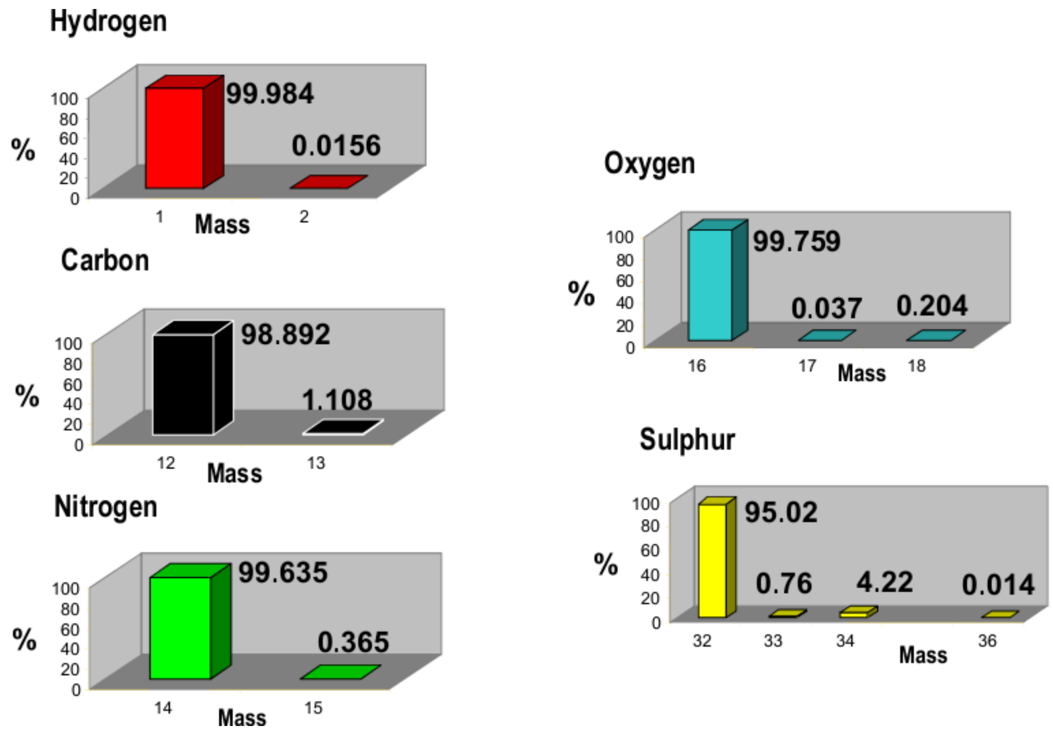
1 1A	2 2A	3 3B	4 4B	5 5B	6 6B	7 7B	8 8B	9 8B	10 8B	11 1B	12 2B	13 3A	14 4A	15 5A	16 6A	17 7A	18 8A
1 H 1.00794																	2 He 4.00260
3 Li 6.941	4 Be 9.01218											5 B 10.81	6 C 12.011	7 N 14.0067	8 O 15.9994	9 F 18.998403	10 Ne 20.1797
11 Na 22.98977	12 Mg 24.305											13 Al 26.98154	14 Si 28.0855	15 P 30.97376	16 S 32.066	17 Cl 35.453	18 Ar 39.948
19 K 39.0983	20 Ca 40.078	21 Sc 44.9559	22 Ti 47.88	23 V 50.9415	24 Cr 51.996	25 Mn 54.9380	26 Fe 55.847	27 Co 58.9332	28 Ni 58.69	29 Cu 63.546	30 Zn 65.39	31 Ga 69.72	32 Ge 72.61	33 As 74.9216	34 Se 78.96	35 Br 79.904	36 Kr 83.80
37 Rb 85.4678	38 Sr 87.62	39 Y 88.9059	40 Zr 91.224	41 Nb 92.9064	42 Mo 95.94	43 Tc (98)	44 Ru 101.07	45 Rh 102.9055	46 Pd 106.42	47 Ag 107.8682	48 Cd 112.41	49 In 114.82	50 Sn 118.710	51 Sb 121.757	52 Te 127.60	53 I 126.9045	54 Xe 131.29
55 Cs 132.9054	56 Ba 137.33	57 *La 138.9055	72 Hf 178.49	73 Ta 180.9479	74 W 183.85	75 Re 186.207	76 Os 190.2	77 Ir 192.22	78 Pt 195.08	79 Au 196.9665	80 Hg 200.59	81 Tl 204.383	82 Pb 207.2	83 Bi 208.9804	84 Po (209)	85 At (210)	86 Rn (222)
87 Fr (223)	88 Ra 226.0254	89 †Ac 227.0278	104 Rf (261)	105 Db (262)	106 Sg (266)	107 Bh (264)	108 Hs (269)	109 Mt (268)	110 (271)	111 (272)	112 (277)		114 (289)		116 (289)		118 (293)

*Lanthanide series	58 Ce 140.12	59 Pr 140.9077	60 Nd 144.24	61 Pm (145)	62 Sm 150.36	63 Eu 151.96	64 Gd 157.25	65 Tb 158.9254	66 Dy 162.50	67 Ho 164.9304	68 Er 167.26	69 Tm 168.9342	70 Yb 173.04	71 Lu 174.967
†Actinide series	90 Th 232.0381	91 Pa 231.0359	92 U 238.0289	93 Np 237.048	94 Pu (244)	95 Am (243)	96 Cm (247)	97 Bk (247)	98 Cf (251)	99 Es (252)	100 Fm (257)	101 Md (258)	102 No (259)	103 Lr (262)

What is an **isotope**: abundance?

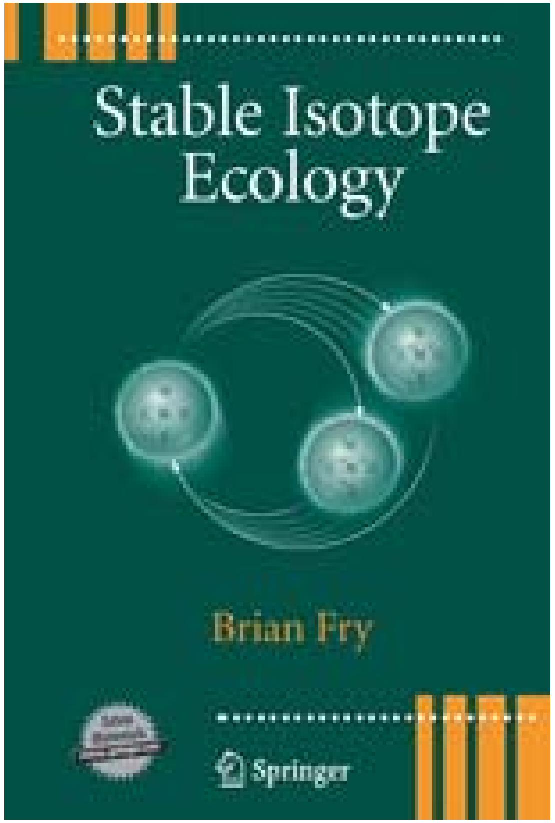
Isotope	Mass	Abundance
n	1.008665	
H	1.007825	99.9844%
D	2.0140	0.0156
^{12}C	12.0000	98.89
^{13}C	13.003355	1.11
^{14}N	14.003074	99.64
^{15}N	15.000108	0.36
^{16}O	15.994915	99.763
^{17}O	16.999131	0.0375
^{18}O	17.999160	0.1995
^{32}S	31.972070	95.02
^{33}S	32.971456	0.75
^{34}S	33.967866	4.21
^{36}S	35.967080	0.02

- Isotopes of the same element have different mass
- Lighter isotopes more common

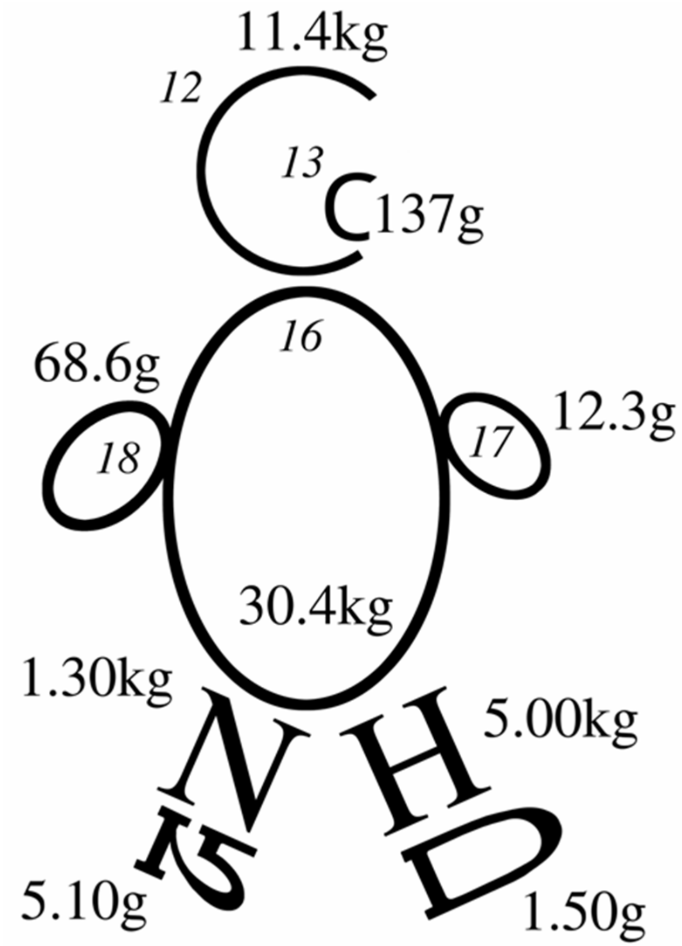


What is an **isotope**: abundance?

- Stable Isotopes are not hazardous to human hearth, in fact are natural parts of each one of us

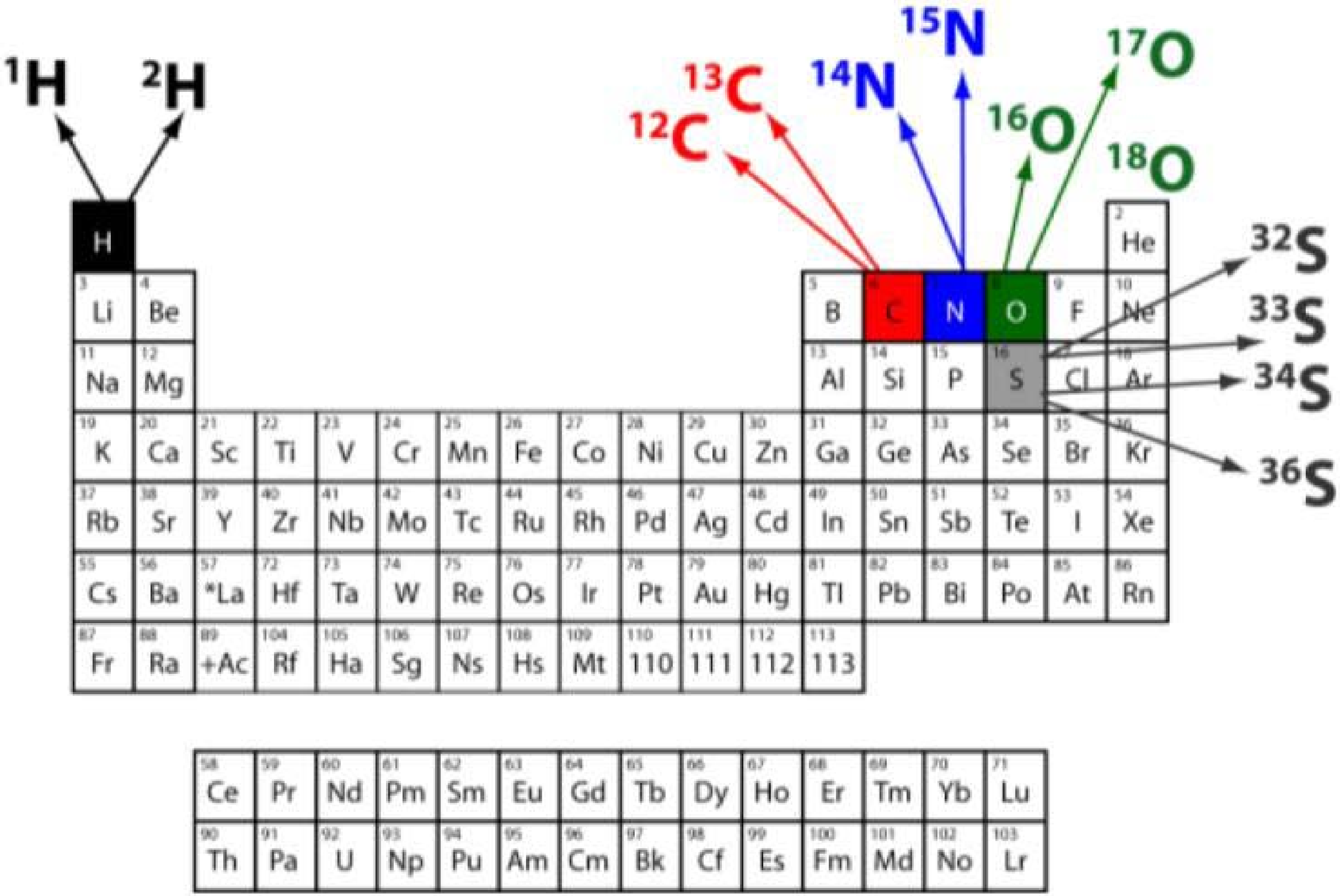


Fry 2006, *Stable Isot Ecol.*



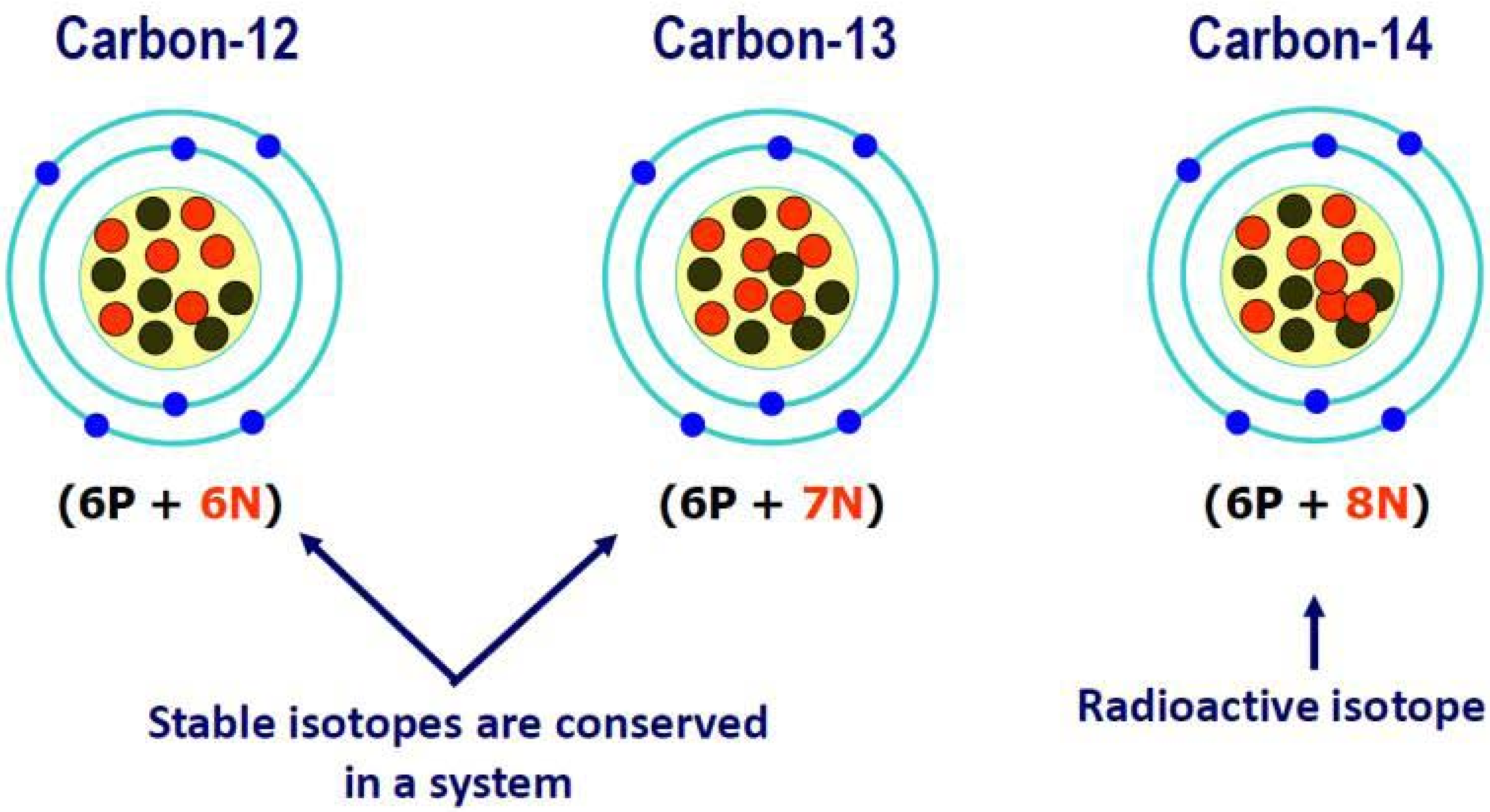
But only 283 isotopes are stable (~10% of known isotopes)

Five elements and 13 stable isotopes

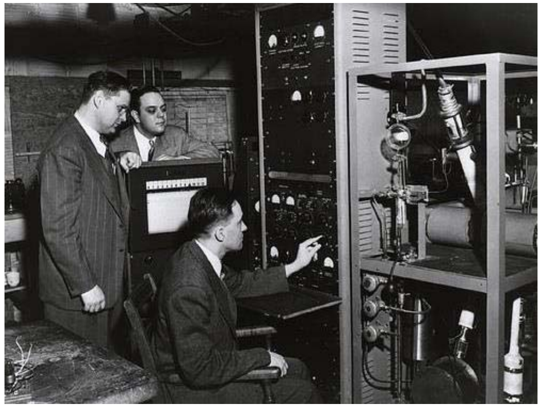


What is an stable isotope?

- Some isotopes are stable: safe isotopes that do not decay and unlike the readioactive isotopes human heal



Stable Isotope Analyses, SIA in the lab



1950's

1960's

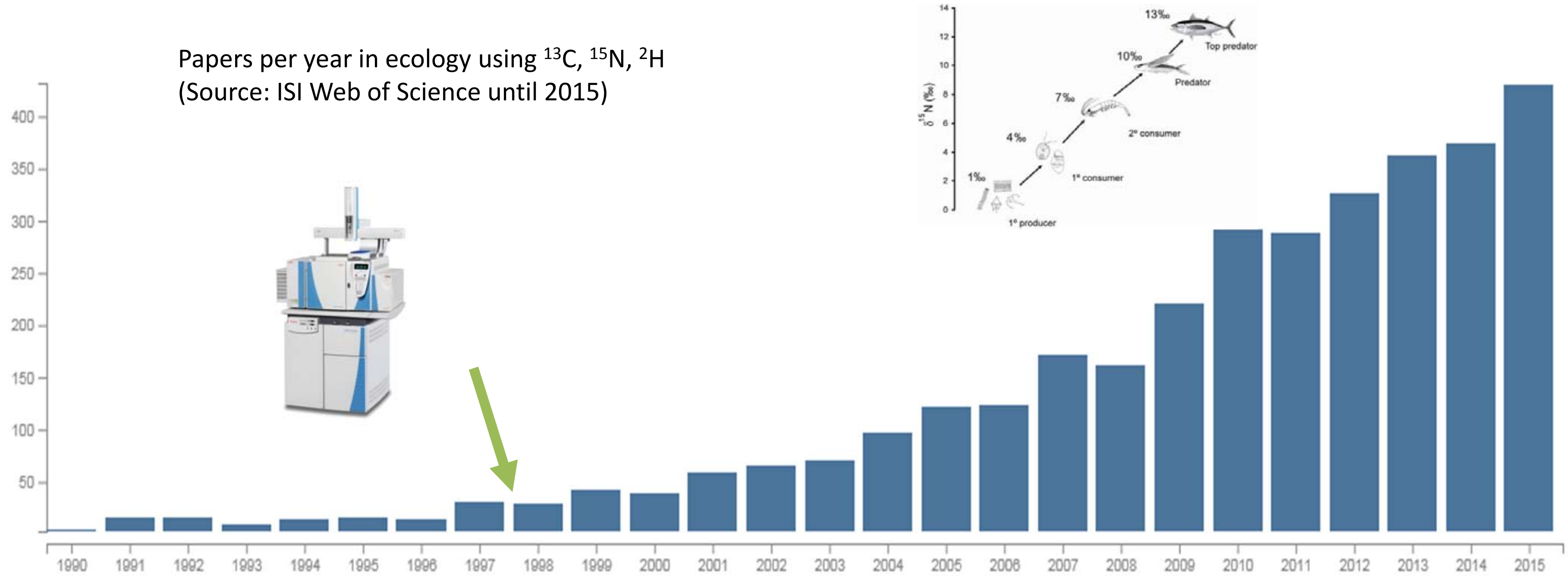
First Mass Commercial Mass Spectrometer created by MAT in Bremen, Germany (1953)

1970's

1990's

2000's

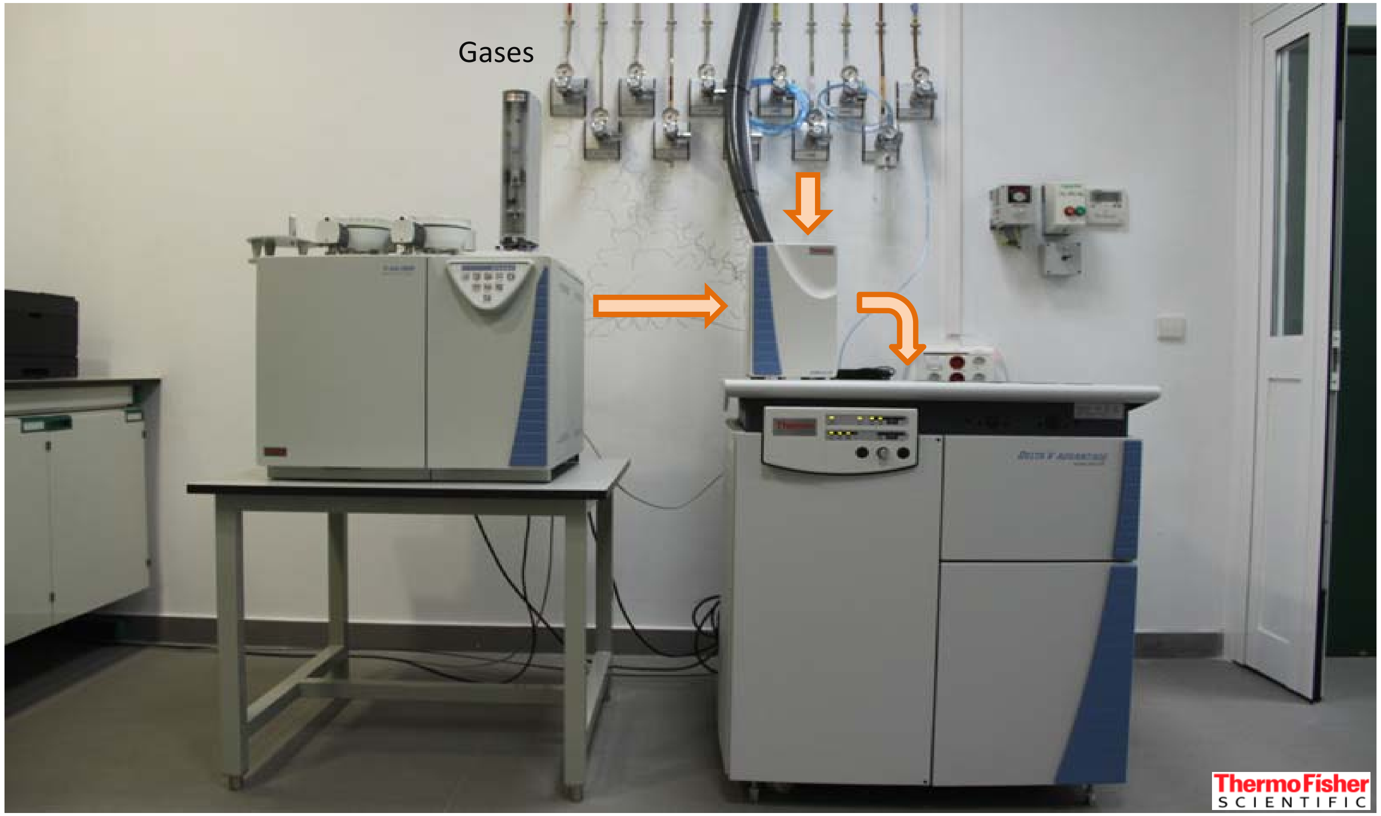
Isotopes in Ecology: Since 1990



Advent of stable isotopes in **ecology** in the mid-1990s was catalyzed by introduction of Elemental Analyzers linked to isotope-ratio mass spectrometers (CF-IRMS, Continous Flour Mass Spectometry)

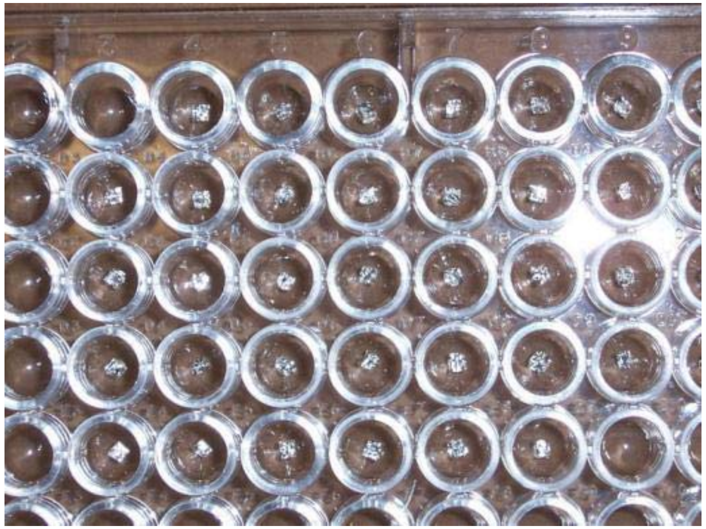
SIA by CF-IRMS

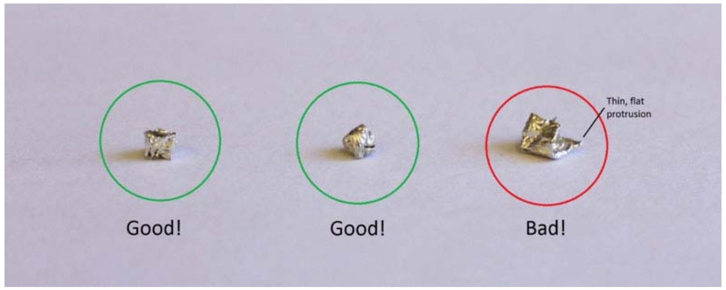
Flash HT *Plus* elemental analyzer coupled to a **Delta-V Advantage** isotope ratio mass spectrophotometer via a **CONFLO IV** interface (Thermo Fisher Scientific, Bremen, Germany)



Sample preparation:

- Dried
- Ground
- Lipid extraction (sometimes)
- Weighted (3 mg) accurately (.000)
- Roll in a tin cup





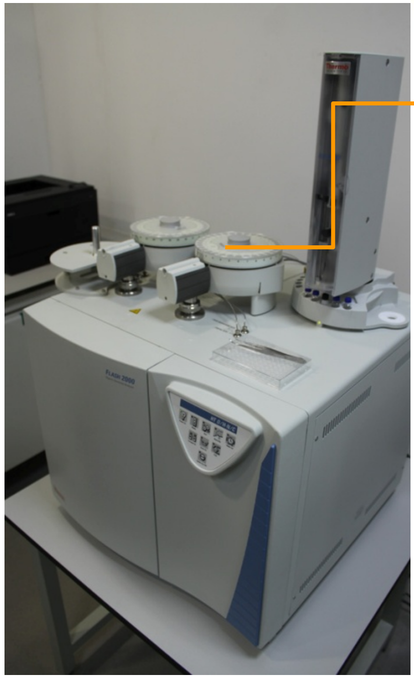
https://www.youtube.com/watch?v=dLo5Ftie2h8&feature=youtu.be -

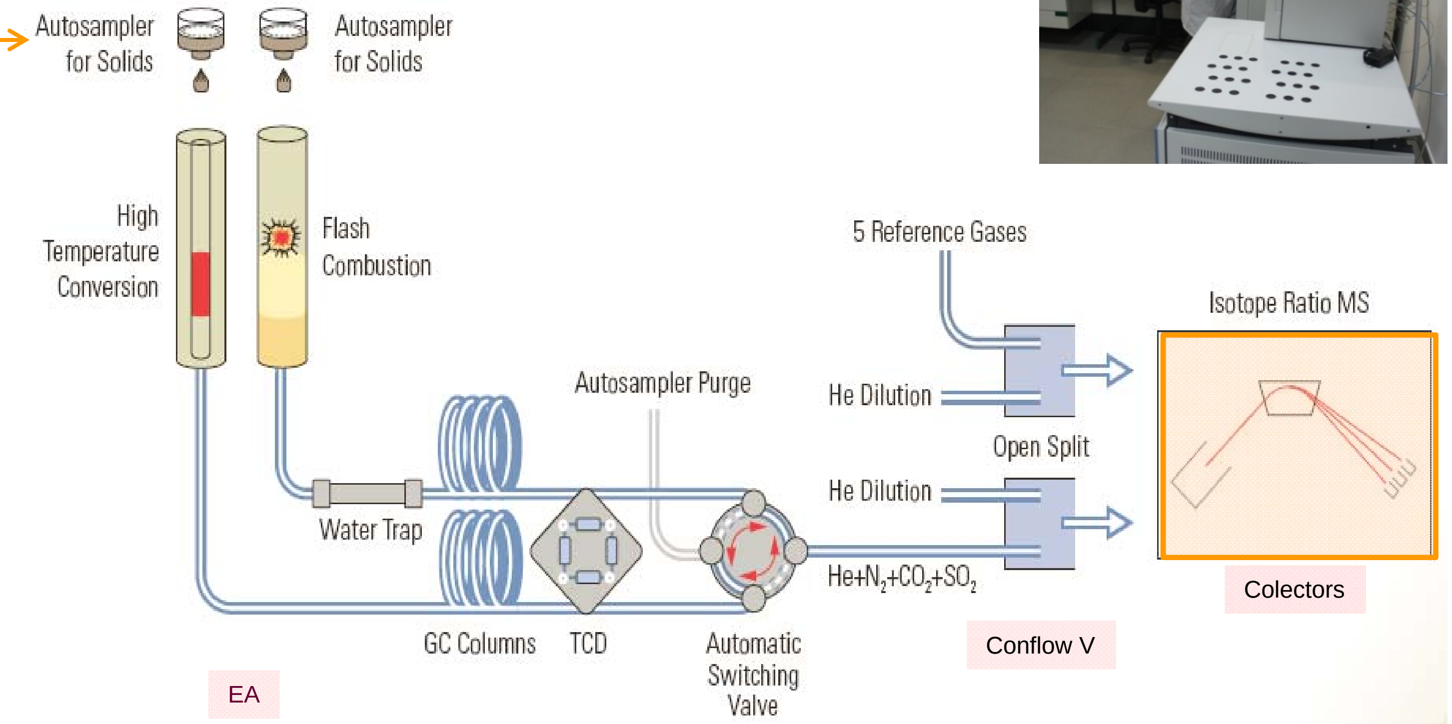
Estación Biológica Doñana CSIC

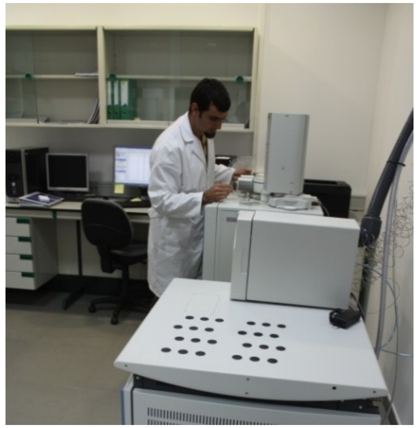
LIE EBD Laboratorio de Isótopos Estables Estación Biológica de Doñana

Sample preparation:

SIA by CF-IRMS



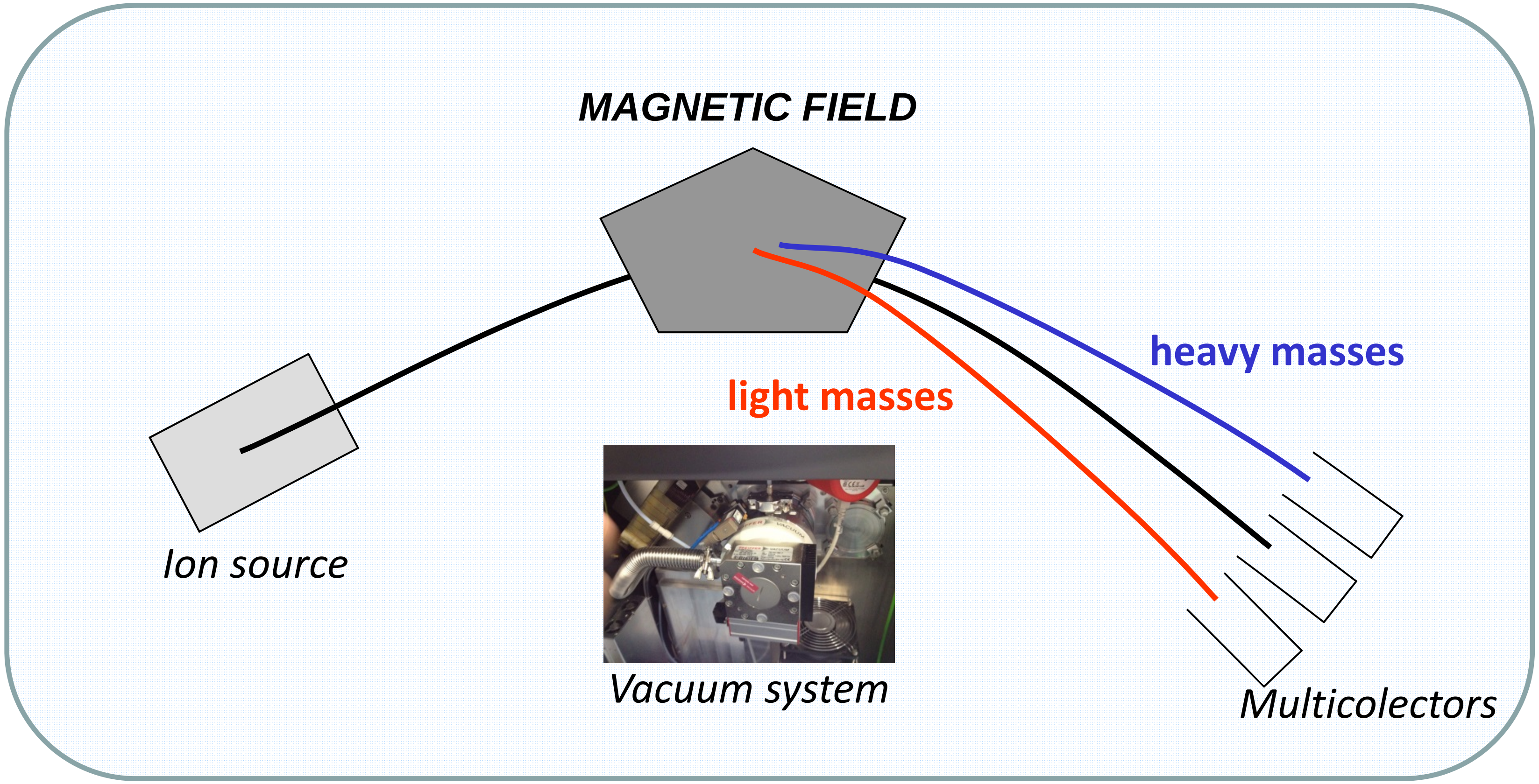




- Chemecally converts sample material (biological samples, rocks, water…) into gas

Anatomy of an IRMS

All the IRMSs have a common anatomy



- Separates isotopes in relation to their mass
- Reference gases as controls

Isotope notation (a ratio of ratios)

$$\delta^{A}X = 1000 \times \frac{{}^{A}R_{sample} - {}^{A}R_{std}}{{}^{A}R_{std}} \quad (‰)$$

Percentage of heavy isotope mass ($\delta^{15}N$, $\delta^{13}C$..)

Element (C, H, S…)

Ratio of heavy to light isotope of the element or the standart (^{2}H:^{1}H, ^{13}C:^{12}C….)

The value for all standars is 0!!

- Differences in abundances expresed as ratio of heavy to light form
- Standardized against international standards (δ, ‰)

Patrones de Isótopos Estables (Definidos como $\delta = 0$)

Patrón	Isótopos	Ratio	Materiales de referencia
Standard Mean Ocean Water (SMOW) y Vienna - SMOW	$^{2}H/^{1}H$ $^{18}O/^{16}O$ $^{17}O/^{16}O$	0,00015576 ± 0,00000010 0,00200520 ± 0,00000043 0,0003799 ± 0,0000015	GISP, SLAP, IAEA-OH1 a IAEA-OH4
Fósil "Pee Dee Belemnite" (PDB)	$^{13}C/^{12}C$ $^{16}O/^{16}C$ $^{17}O/^{18}O$	0,0112372 ± 0,0000090 0,0020671 ± 0,0000021 0,0003859 ± 0,0000016	NBS-18, NBS-19 y 22, IAEA-CH-6 y CO-1, USGS-24, etc
Nitrógeno atmosférico	$^{15}N/^{14}N$	0,0036765 ± 0,0000018	Aire, IAEA-N-1 a IAEA-N-3, NSVEC
Meteorito "Canyon Diablo Troilite" (CDT)	$^{34}S/^{32}S$	0,0450045 ± 0,0000093	NIST-8557, IAEA-S3

- Some distributed by International Atomic Energy Agency in Vienna, Austria
- Some free, nitrogen and oxigen gas in atmosphera
- Some internal reference material

Laboratory reference material

Three keratin lab standards for daily laboratory use in isotopic studies

Ricardo Álvarez*, Francisco Ramírez and Manuela G. Forero
Estación Biológica de Doñana (EBD-CSIC), Laboratorio de Isótopos Estables** (LIE-EBD)
*email: ralvarez@ebd.csic.es
**Web Page: http://www.ebd.csic.es/lie/index.html

Introduction

- Reference materials provided by the International Atomic Energy Agency (IAEA) are expensive and distributed in small quantities, so that they are not intended for daily use, but only for calibration of in-house lab standards
- No suitable certified organic reference material for δ^2H and $\delta^{18}O$ determinations in keratinous tissues are yet available from the IAEA
- In order to measure δ^2H in keratin material is necessary to have a keratin reference accounting for H that is exchanged with ambient laboratory air moisture

Objectives

- To prepare three different keratin reference standards for routine daily use in the stable isotope laboratory

Keratin Standard Origins

1- Fin whale (*Balaenoptera physalus*) baleen (**BB**): beached whale found in Malaga in 2012
2- Cow (*Bos taurus*) horn (**CV**): Obtained from a endemic cow from the Natural Area of Doñana
3- Razorbill (*Alca torda*) feathers (**PA**): Beached razorbill found in Huelva

Preparation

1) Cleaned keratin samples were cut into <1 cm pieces
2) Samples were solvent cleaned to remove fats and oils
3) Ball Milling to < 120 um particle size
4) Shaker sieving with < 120 um mesh, followed by blending
5) Random sampling for isotopic variance testing

Analyses

- Isotopic analyses in the "Laboratorio de Isótopos Estables" of the Estación Biológica de Doñana (LIE-EBD) were carried by means of a Thermo-Scientific Flash HT Plus elemental analyzer coupled to a Delta-V advantage isotope ratio mass spectrometer via a CONFLO IV interface (Thermo Fisher Scientific)
- Analyses of $\delta^{15}N$, $\delta^{13}C$ and $\delta^{34}S$ were performed in Dynamic Flash Combustion mode, and δ^2H and $\delta^{18}O$ by High Temperature Conversion, with the following configurations:

N/C analyses

S analyses

H/O analyses

Result

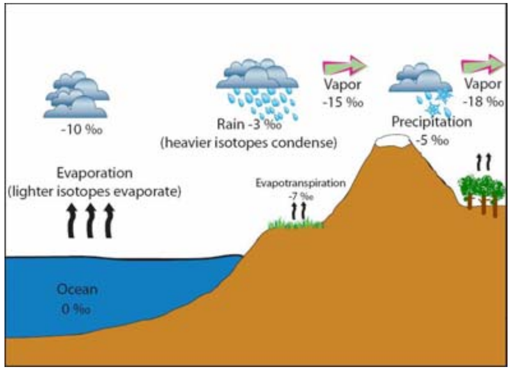
BB $\delta^{15}N$= +9.97 +/- 0.07 air (n=6) $\delta^{13}C$= -18.6 +/- 0.08 V-PDB (n=6) $\delta^{34}S$= +19.17 +/- 0.2 VCDT (n=2)* δ^2H = -46 +/-1.8 ‰VSMOW (n=11) **	**CV** $\delta^{15}N$= +10.27 +/- 0.08 air (n=5) $\delta^{13}C$= -22.24 +/- 0.07 V-PDB (n=5) $\delta^{34}S$= +10.89 +/- 0.2 VCDT (n=2)* δ^2H = -82.1 +/-1.8 ‰VSMOW (n=3) ** $\delta^{18}O$ = +16.69 +/-0.3 ‰ VSMOW (n=3) **
PA $\delta^{15}N$= +16.47 +/- 0.09 air (n=8) $\delta^{13}C$= -15.69 +/- 0.08 V-PDB (n=8) δ^2H = +24 +/-1.2 ‰VSMOW (n=9) **	**Spectrum Keratin Fine Powder Lot# 2AJ3011***** $\delta^{15}N$ = 6.02 +/-0.04 ‰ air (n=5) $\delta^{13}C$ = -15.02 +/-0.03 ‰ V-PDB (n=5)

*Pending for validation at other stable isotope laboratories.
**Isotopic values were normalized with Caribou Hoof Standard (CBS) and Kudu Horn Standard (KHS) supplied by Environment Canada, which are pending for international validation.
***Powdered keratin from Spectrum Chemicals.

BB and CV were simultaneously analyzed at LIE-EBD, the Reston Stable Isotope Laboratory (RSIL, U.S. Geological Survey, USA), and the Stable Isotope Hydrology and Ecology Research Laboratory (SIHERL, Environment Canada, Canada), whereas PA has been exclusively analyzed at LIE-EBD and SIHERL.

To the best of our knowledge, no positive standards for δ^2H are available to apply a suitable correction to PA. This material was normalized with CBS and KHS with value of δ^2H of -197 and -54.1 respectively. Therefore, this value is provisional, pending of further calibrations. At the present, PA is the only available keratin standard with positive value for δ^2H analyses.

Acknowledgements. We thank to Carlos Gutiérrez Expósito, Renaud de Stephanis and the Scientific Collections of the EBD for providing pictures and samples. We also thank RSIL and SIHERL for the complementary analyses

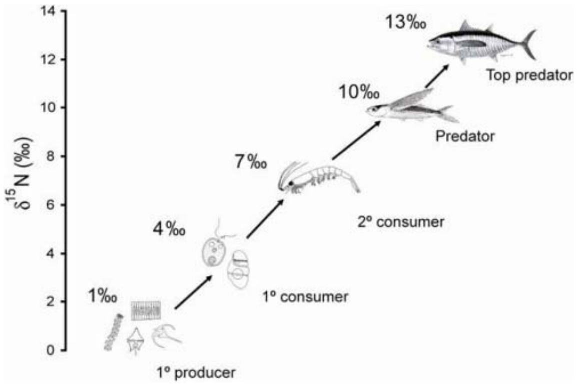


Isotope fractionation (α)

Very small differences in mass cause subtle differences in their behaviour during chemical reactions and diffusion which alter the ratio of heavy to light isotopes

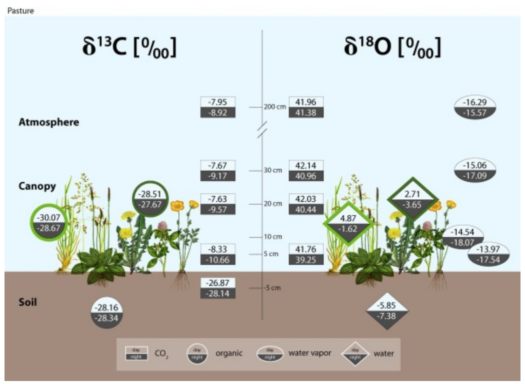
↓

Differences among organisms and across different habitats in their stable isotope ratios



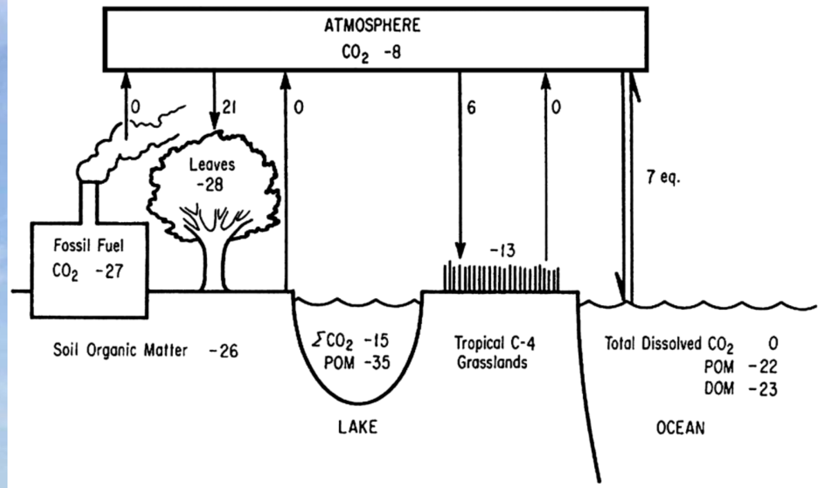
Sources of fractionation:

- Diffusion, evaporation, metabolic effects (respiration, digestion, photosynthesis…)

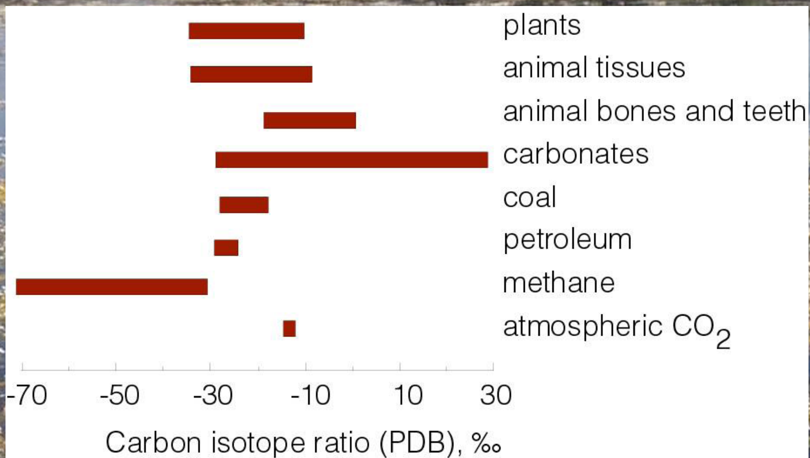


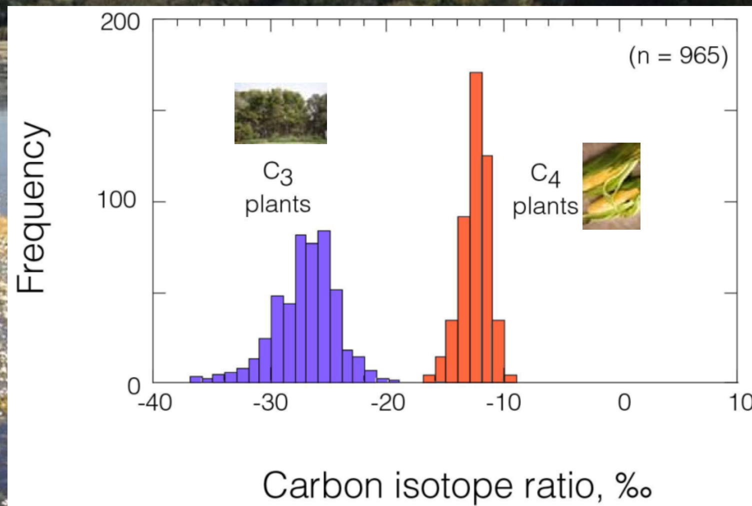
$\delta^{13}C$ GRADIENTS

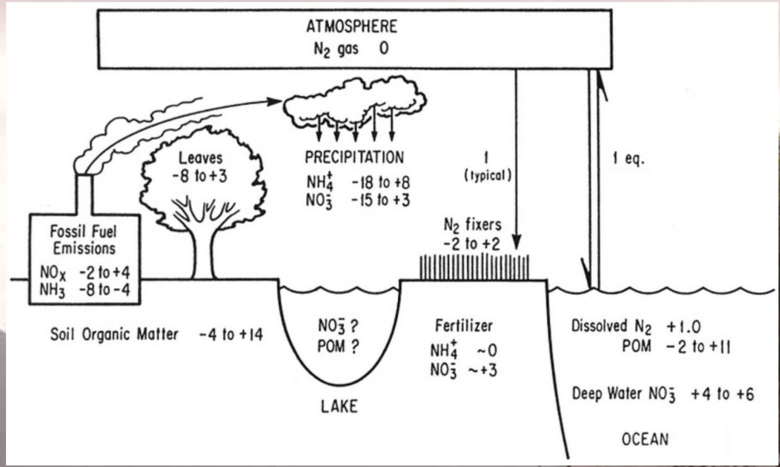
- Photosynthetic pathway (C3, C4, CAM) and water-use efficiency.
- Marine vs. freshwater/terrestrial C sources.
- Spatial Indicator of aquatic primary production (benthic/inshore vs pelagic/offshore).
- Tracer of C source and mechanisms related to atmospheric and global change.



Fry 2006, *Stable Isot Ecol.*







Fry 2006, *Stable Isot Ecol.*

$\delta^{15}N$ UTILITIES

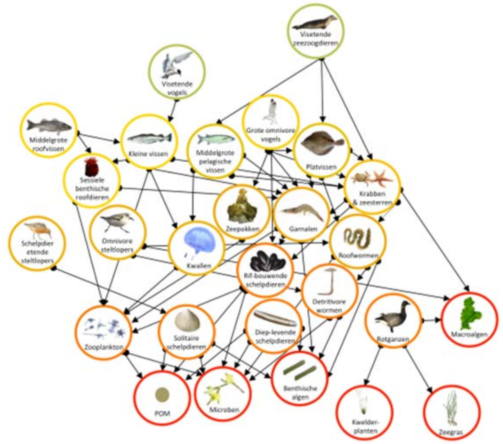
- Identify sources o nitrate to aquatic systems
- Tracer of nitrification and denitrification
- N sourcing to plants
- Terrestrial vs. marine inputs to foodwebs
- Foodweb trophic level indicator

ALSO $\delta^{34}S$

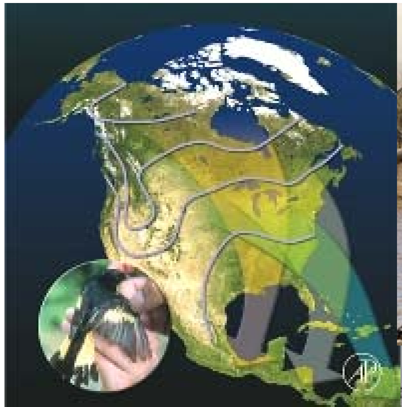
- Animal studies: marine vs. terrestrial, estuaries, marshes; S-amino acids …..

Why should we use them (CNS, HO in Ecology)?

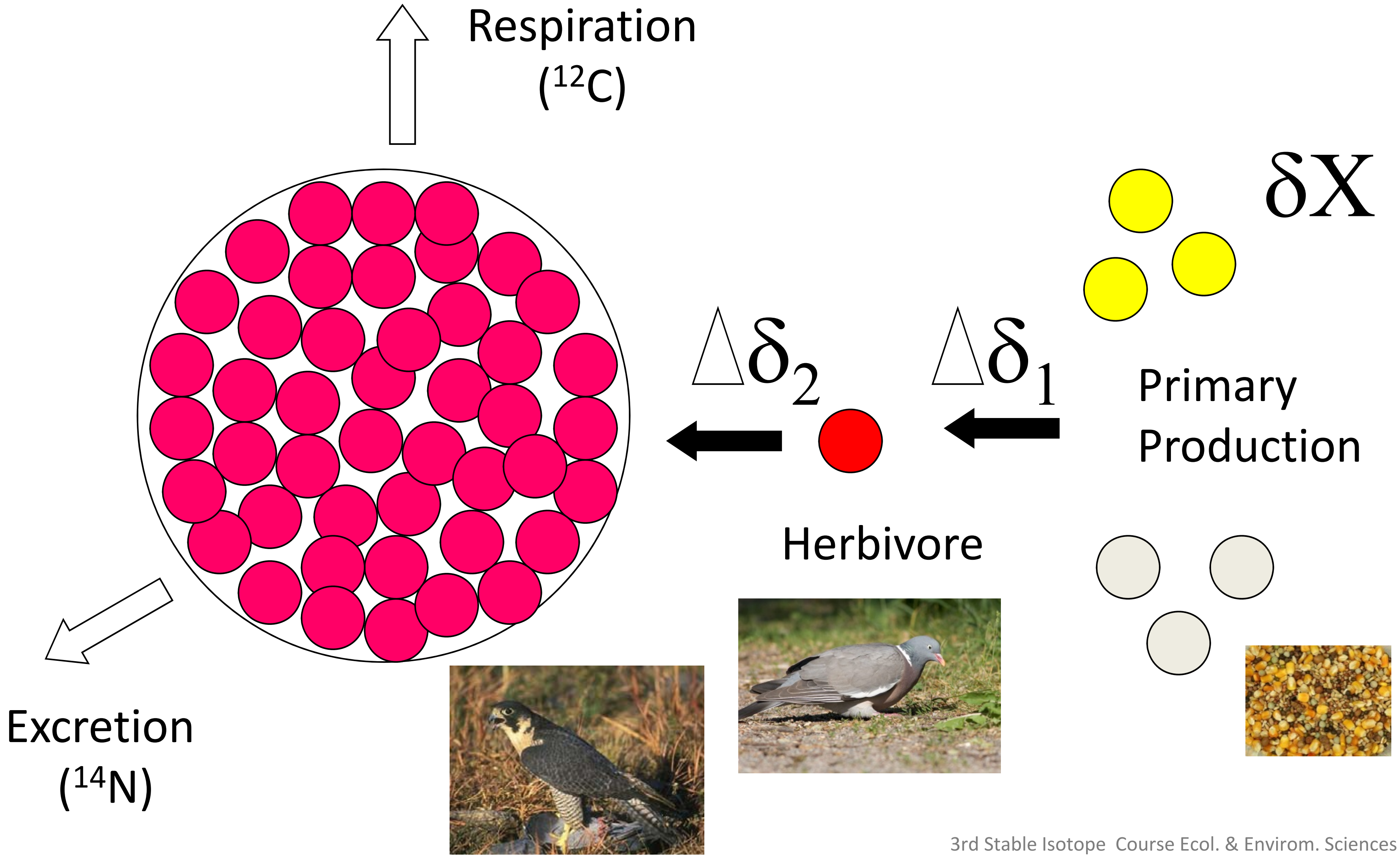
- Trophic and nutricional ecology, food webs



- Animal movement, migration research



Foodweb markers the basic principles of trophic level and sources determinations:



Discrimiantion Factors (Δδ)

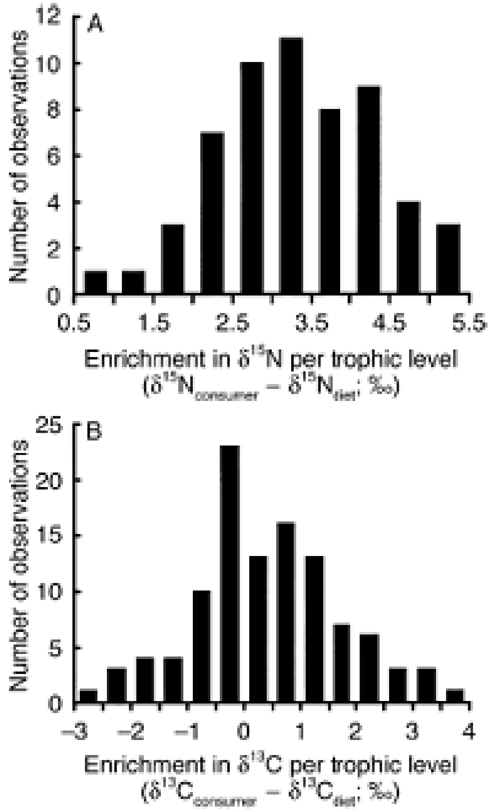


FIG. 6. Frequency distributions of the enrichment in (A) $\delta^{15}N$ and (B) $\delta^{13}C$ per trophic level. The means are 3.4‰ for $\delta^{15}N$ (SD = 0.98, n = 56) and 0.39‰ for $\delta^{13}C$ (SD = 1.3, n = 107), and neither distribution is significantly different from normal. See *Methods* for the list of studies used to produce these figures.

June 1997 REPORTS 1271

Ecology, 78(4), 1997, pp. 1271–1276
© 1997 by the Ecological Society of America

STABLE ISOTOPES IN ANIMAL ECOLOGY: ASSUMPTIONS, CAVEATS, AND A CALL FOR MORE LABORATORY EXPERIMENTS

LEONARD Z. GANNES,[1] DIANE M. O'BRIEN,[1] AND CARLOS MARTÍNEZ DEL RIO[2]

BIOLOGICAL REVIEWS Cambridge Philosophical Society

Biol. Rev. (2009), **84**, pp. 91–111. 91
doi:10.1111/j.1469-185X.2008.00064.x

Isotopic ecology ten years after a call for more laboratory experiments

Carlos Martínez del Rio[1*], Nathan Wolf[1], Scott A. Carleton[1] and Leonard Z. Gannes[2]

Journal of Applied Ecology

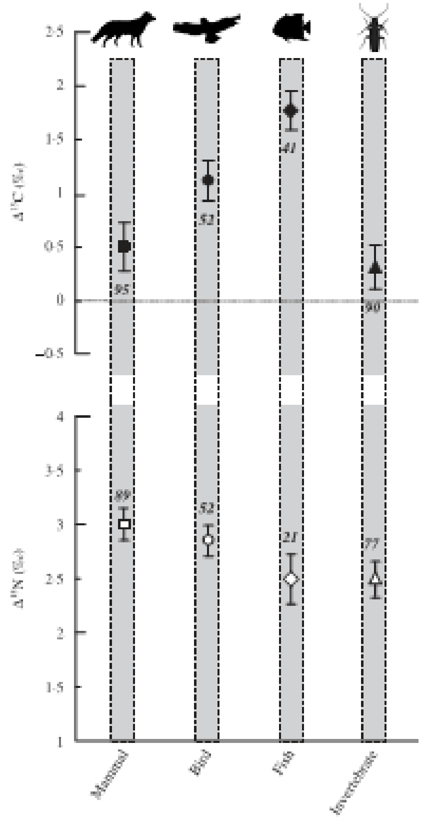
Journal of Applied Ecology 2009, 46, 443–453 doi: 10.1111/j.1365-2664.2009.01620.x

REVIEW

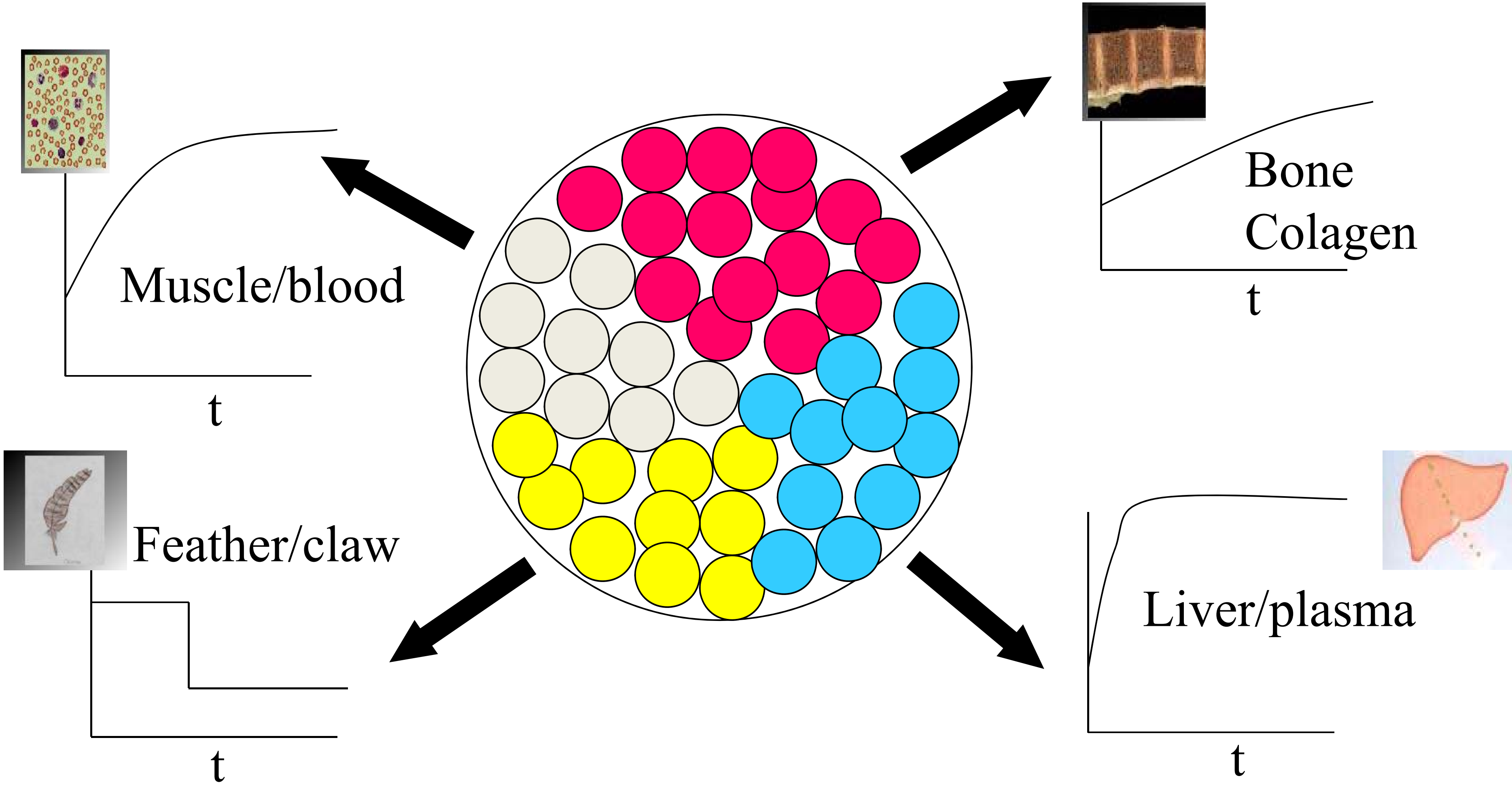
Variation in discrimination factors ($\Delta^{15}N$ and $\Delta^{13}C$): the effect of diet isotopic values and applications for diet reconstruction

Stéphane Caut[1,2*], Elena Angulo[2] and Franck Courchamp[1]

Vander Zanden et al. 2015. *PLOS one*



Stable isotope Turnover (λ, % day^{-1}) and Isotopic Half-Life (ln(2)/ λ , days): choice of tissue….



Experimental isotopic diet shift studies: discrimination factors and turnover rates

Journal of Experimental Marine Biology and Ecology 475 (2016) 54–61

Contents lists available at ScienceDirect

Journal of Experimental Marine Biology and Ecology

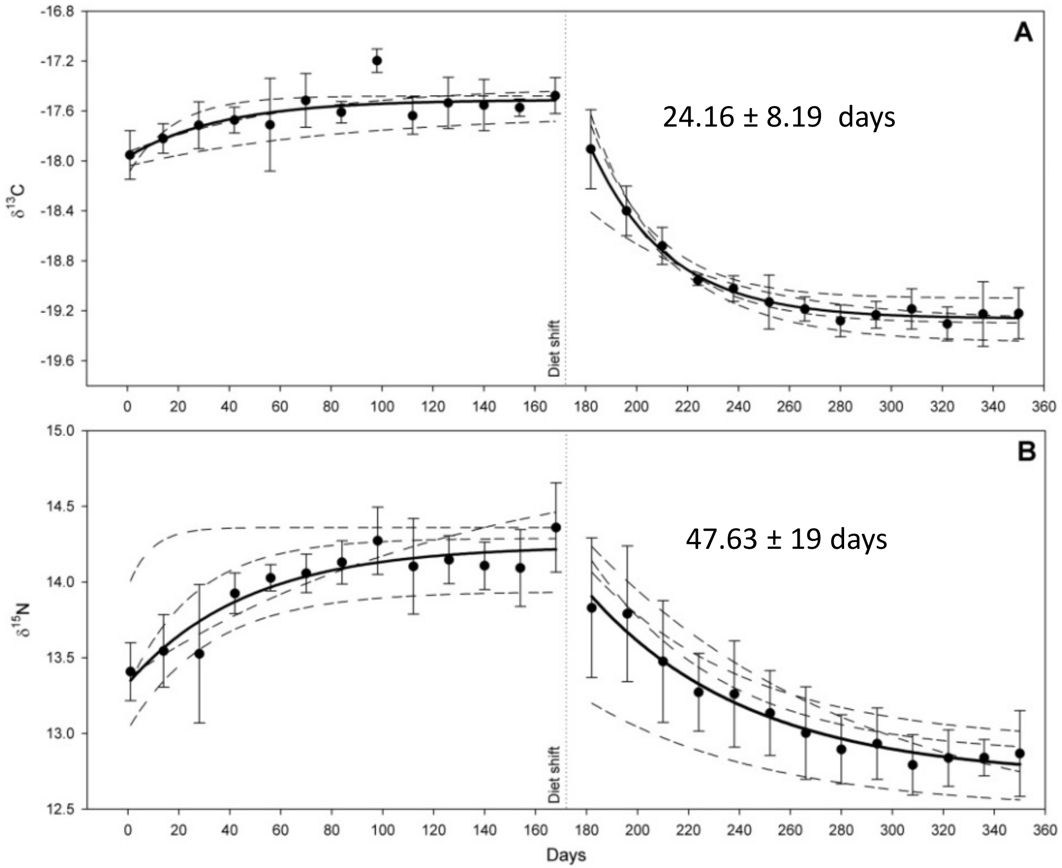
journal homepage: www.elsevier.com/locate/jembe

From the pool to the sea: Applicable isotope turnover rates and diet to skin discrimination factors for bottlenose dolphins (*Tursiops truncatus*)

Joan Giménez [a,*], Francisco Ramírez [a], Javier Almunia [b], Manuela G. Forero [a], Renaud de Stephanis [a,c]

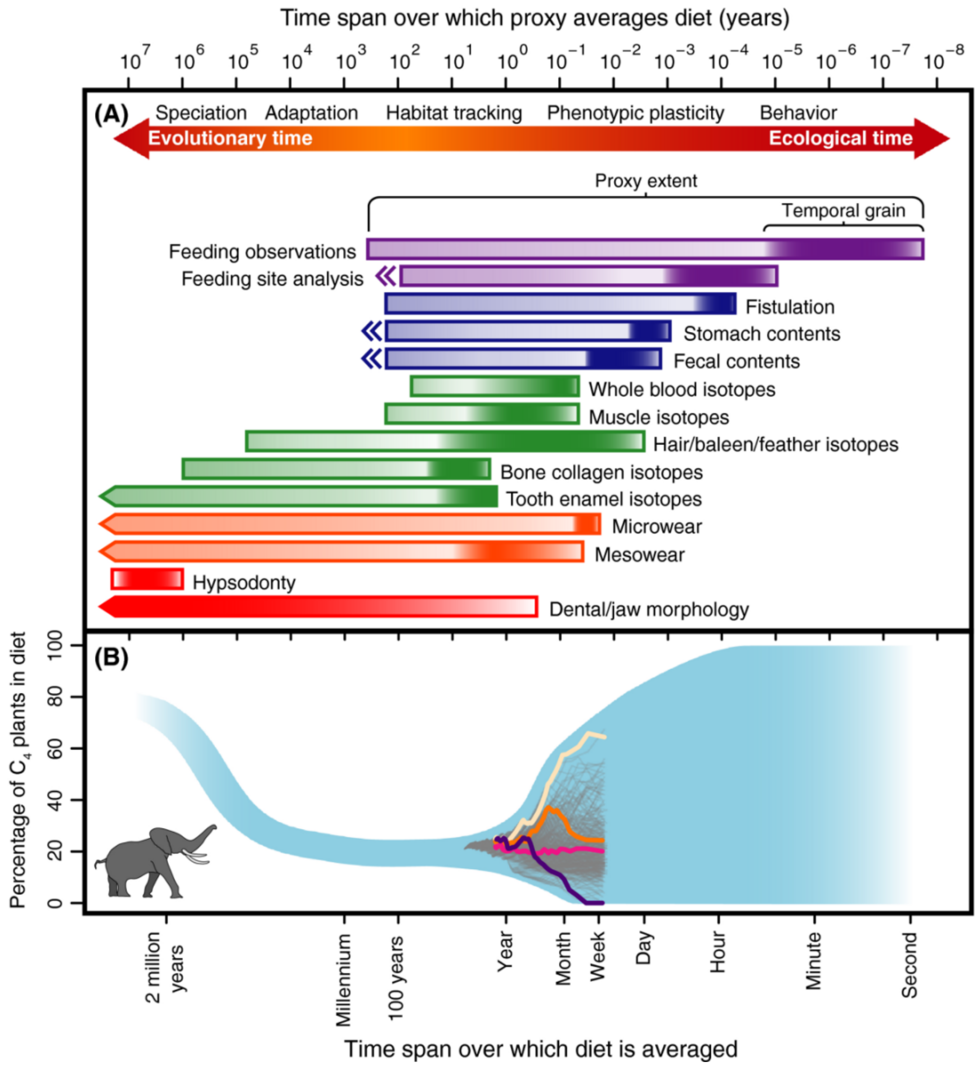
Experimental isotopic diet shift studies: discrimination factors and turnover rates

	$\delta^{13}C$		$\delta^{15}N$	
	fish muscle	**whole fish**	**fish muscle**	**whole fish**
Δ Clara	1.31	1.23	1.89	2.06
Δ Luna	0.94	0.86	1.25	1.42
Δ Pacina	0.85	0.77	1.7 1.87	
Δ Paco	0.94	0.86	1.43	1.6
Mean Δ ± sd	***1.01 ± 0.37***	***0.93 ± 0.56***	***1.57 ± 0.52***	***1.74 ± 0.55***



Giménez *et al.* 2016 *J Exp Mar Biol & Ecol*

Important to match scale of our questions with the scale of our data

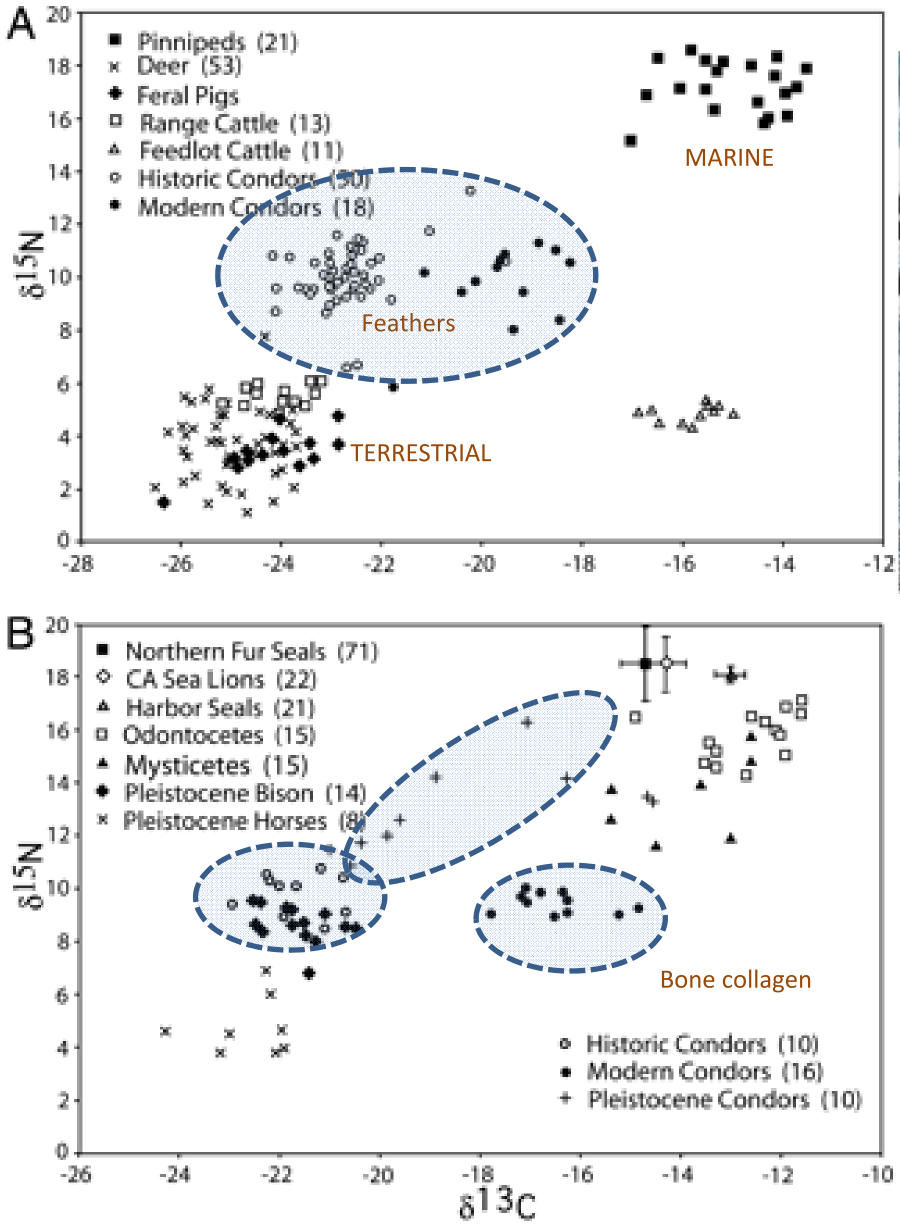


Loxodonta africana

"Stable isotopes: assimilated diet from short to long temporal scales"

Davis & Pineda Muñoz 2016. *Ecol and Evol.*

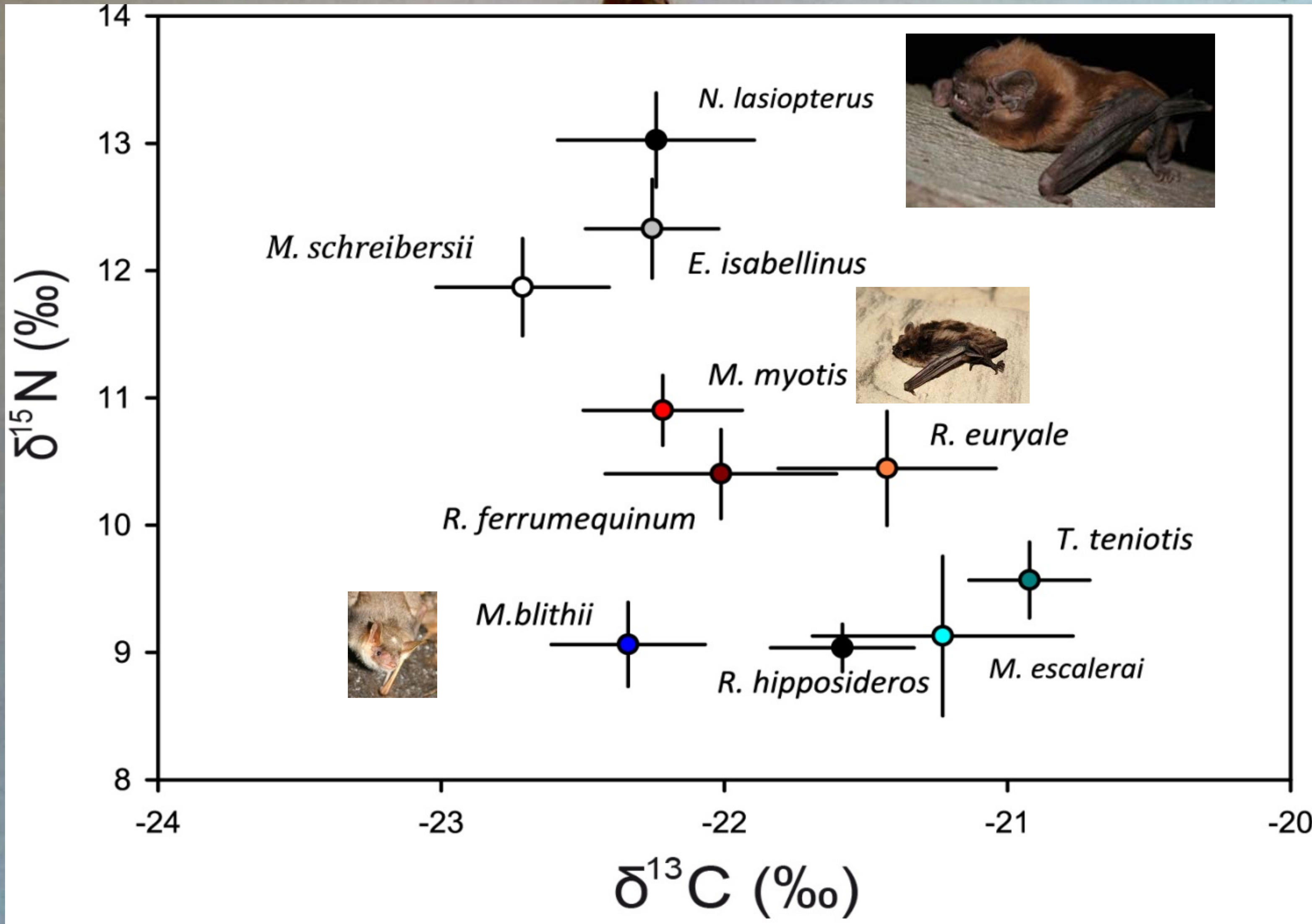
Long temporal scale: stable isotopes in fossil material, bone collagen, feathers from scientific collections



Gymnogyps californianus
Chamberlain et al. 2005, *PNAS*

Pleistocene: terestrial megafauna & marine mammals
Historic Condors: Terrestrial
Moderm Condors: Human subsides

Evolutive approach using long integrating tissues: looking for a species-specific signal in bat skeletons



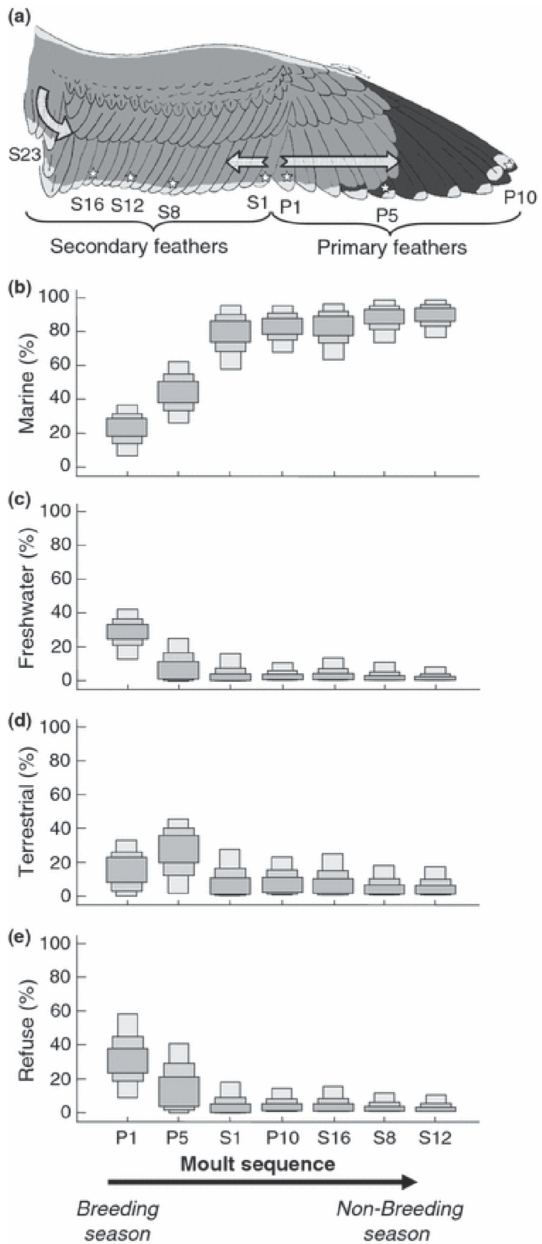
Juste, Forero et al., *in prep*

Innert tissues: spatiotemporal components of trophic ecology

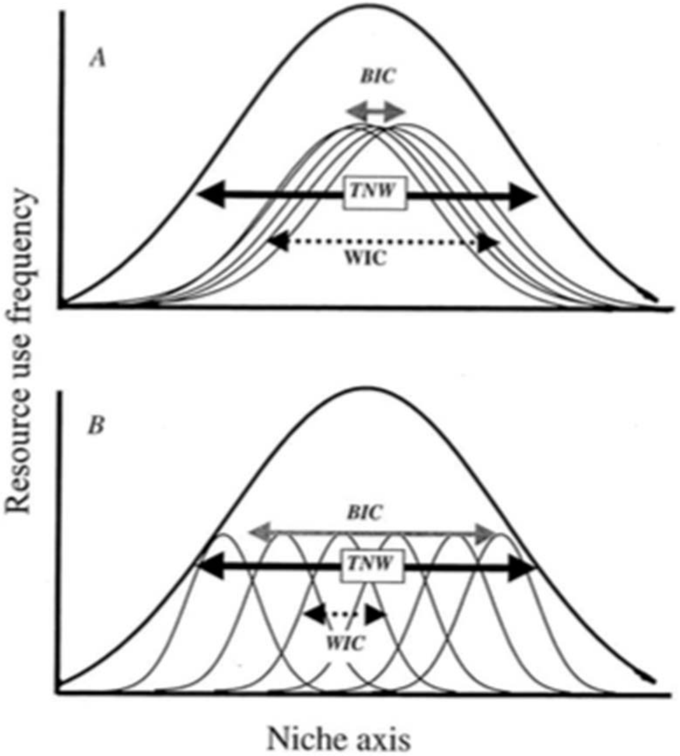
Stable Isotope Analises in R mixing moddeling (C, N, S)

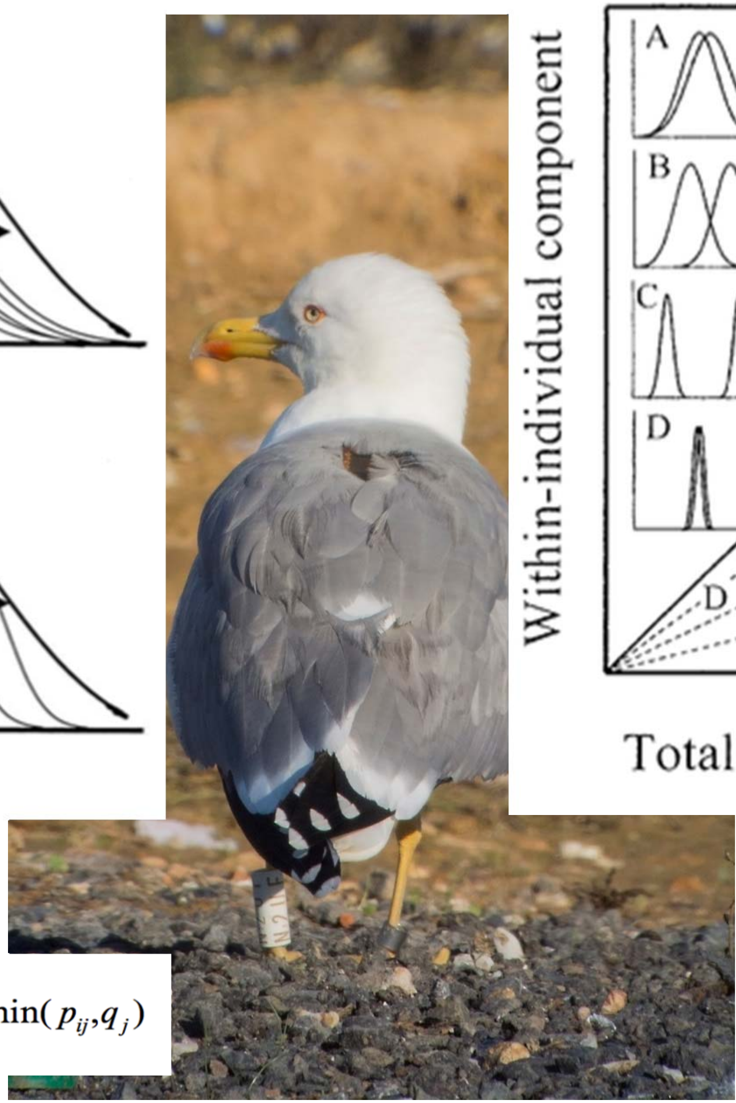
Larus michaellis

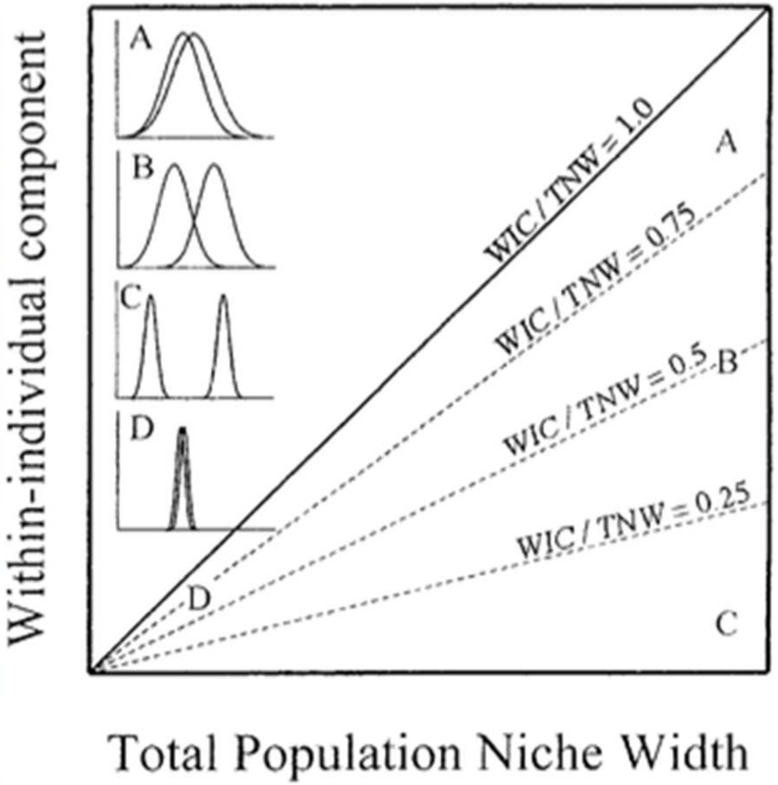
Ramos *et al*. 2011, *Diversity & Distributions*



Dietary and isotopic specialization



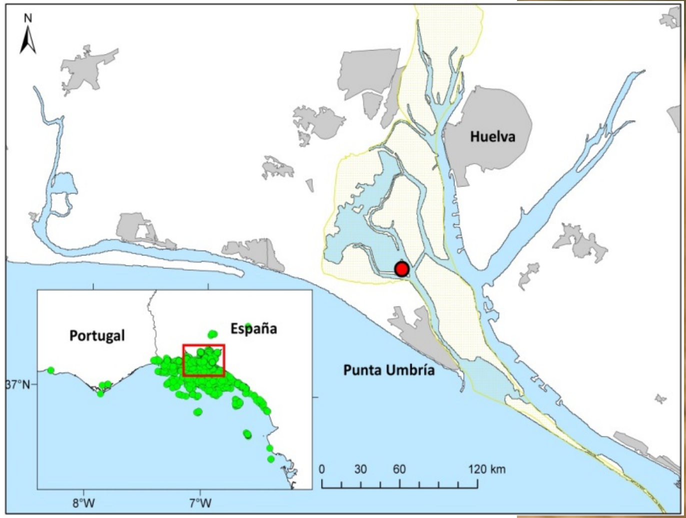




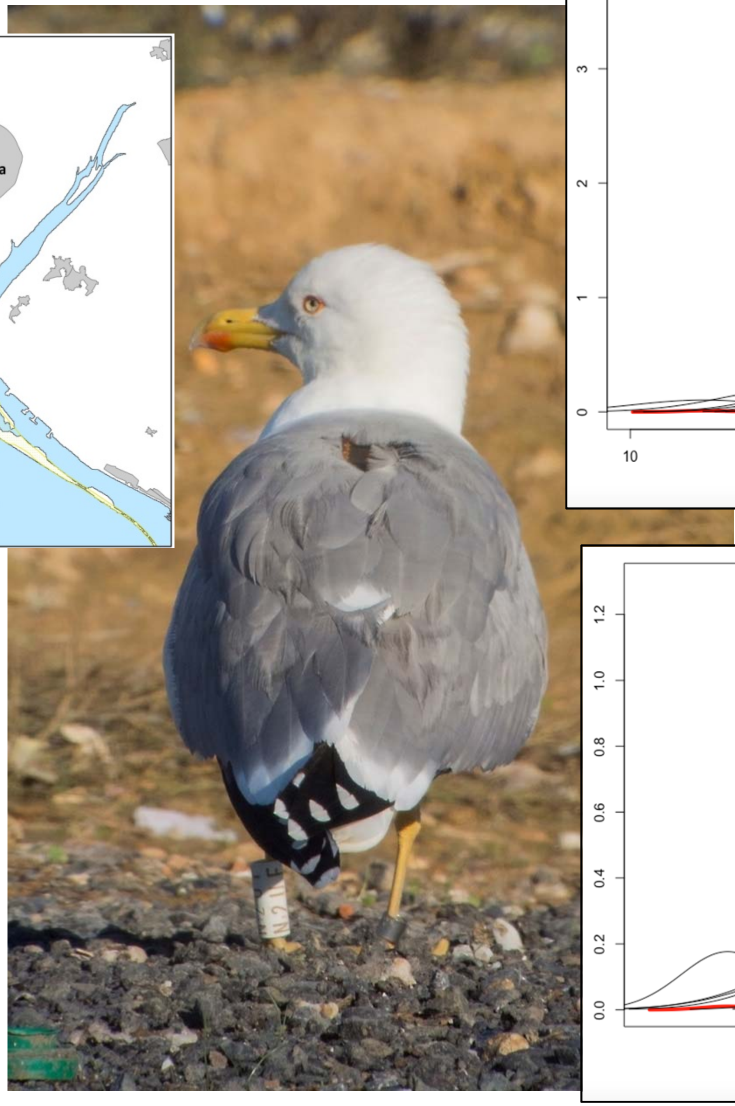
$$PS_i = 1 - 0{\cdot}5 \sum_j | p_{ij} - q_j | = \sum_j \min(p_{ij}, q_j)$$

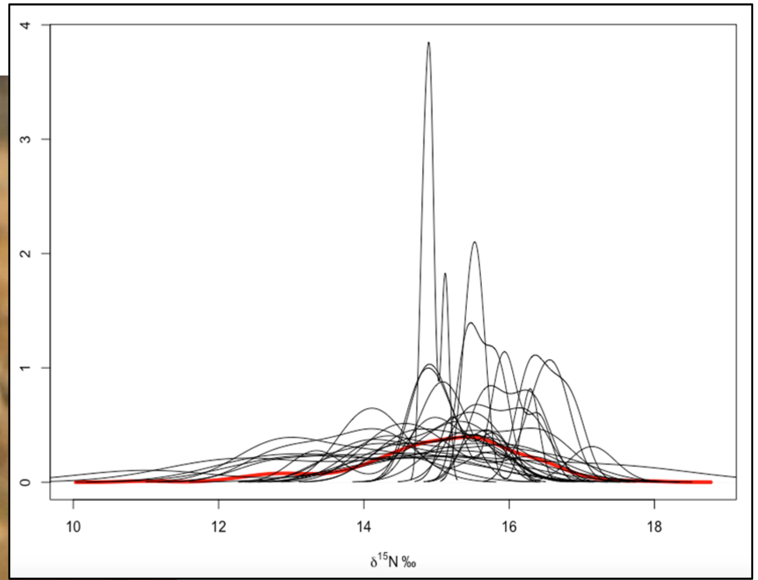
Bolnick *et al.* 2003, *American Naturalist*

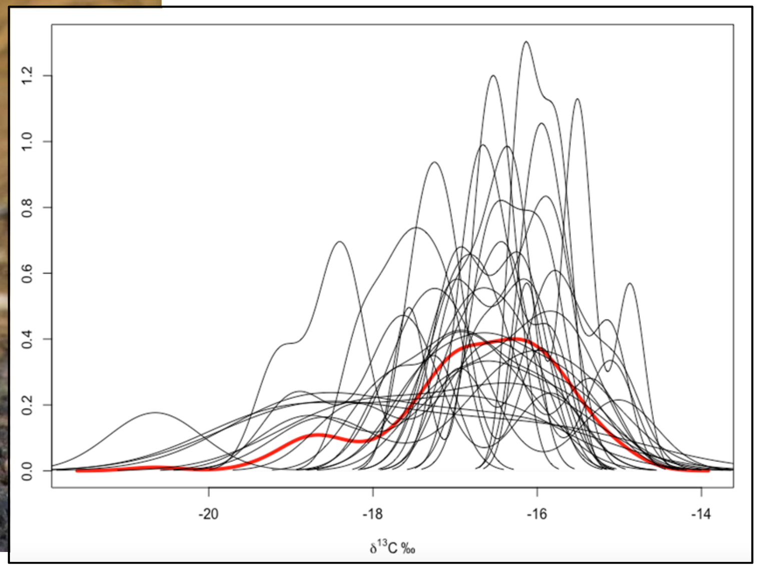
Dietary and isotopic individual specialization



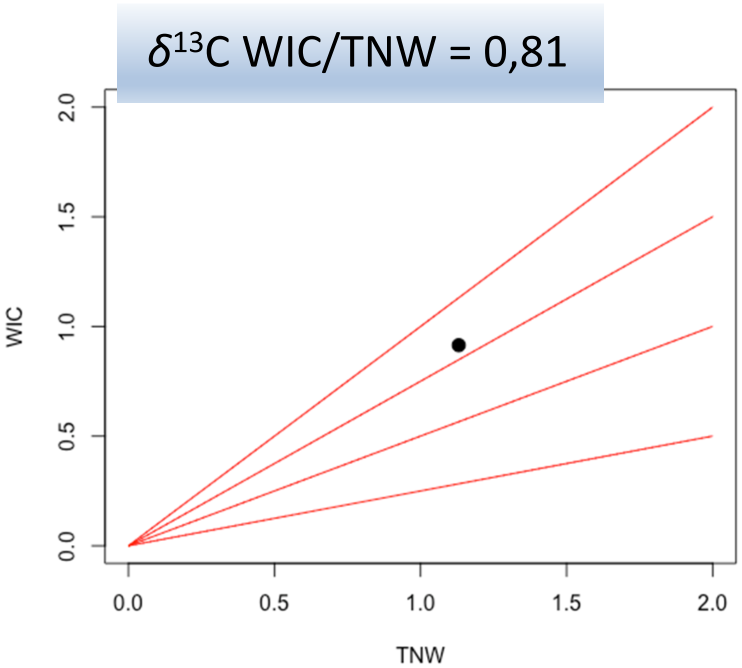
Marismas del Odiel
Larus michaellis, 4 tejidos, C, N

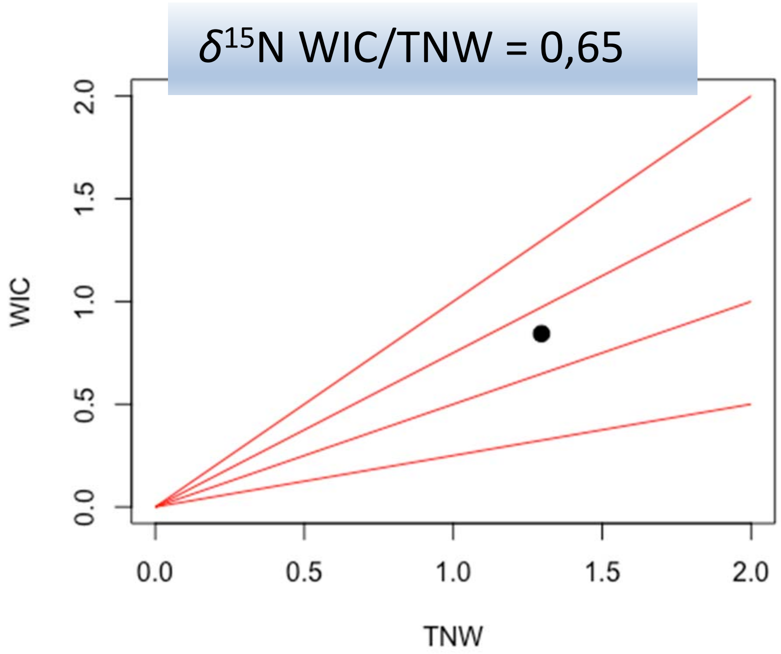






Dietary and isotopic specialization at population level

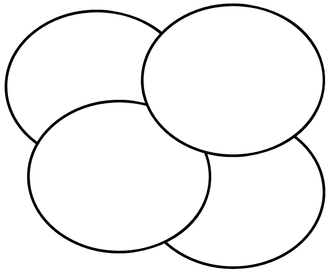


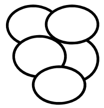


Tejido	PS_i
Sangre	0,91
Pluma corporal	0,91
Primera Primaria	0,95
Octava Secundaria	0,92

Lopezosa *et al. In prep (trabajo fin de master)*

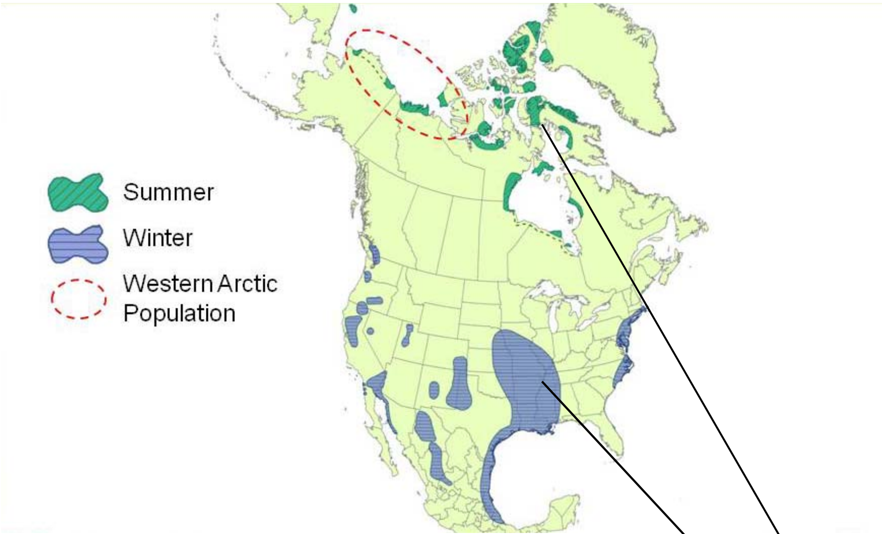
Understanding nutrient allocations to reproduction: capital vs. income breeders





Drent & Daan 1980, *Ardea*

Endogenous vs. exogenous nutrients



Greater snow goose (*Chen caerulescens atlantica*)
~~Tradicional Capital breeder~~

Contribution of:

- Reserves: 16-34%
- Food ingested in breeding grounds: 65-84%

$\delta^{13}C$ and $\delta^{15}N$ in:

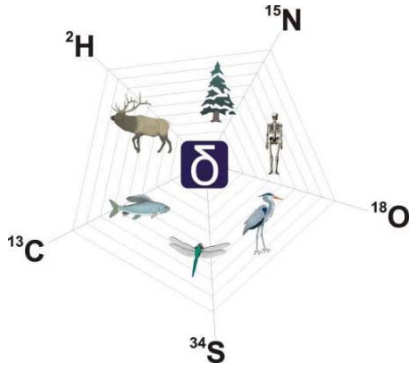
- ✓ Yolk & albumen
- ✓ Goose tissues (liver, breast muscle, abdominal fat)
- ✓ Graminoids and forbs from Artic

MIXED CAPITAL/INCOME BREEDING STRATEGY

Gautier et al. 2003, *Ecology*

So far…

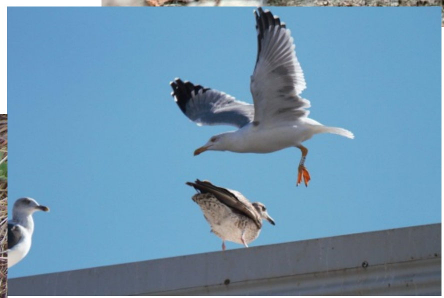
Isotope fractionation and Turnovers: ***covering different temporal and spatial scales in applications***



Intraespecific studies:

- Paleodiet reconstruction
- Long term changes in diet, evolutionary approach
- Spatio-temporal variation in trophic ecolgy
- Individual and population specialisation level
- Nutrient allocation





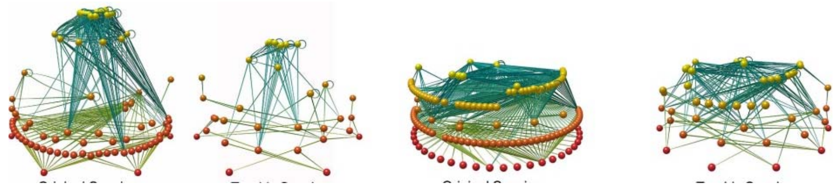

From specific to community approach: quatitative measures of trophic structures

Ecology, 88(1), 2007, pp. 42–48

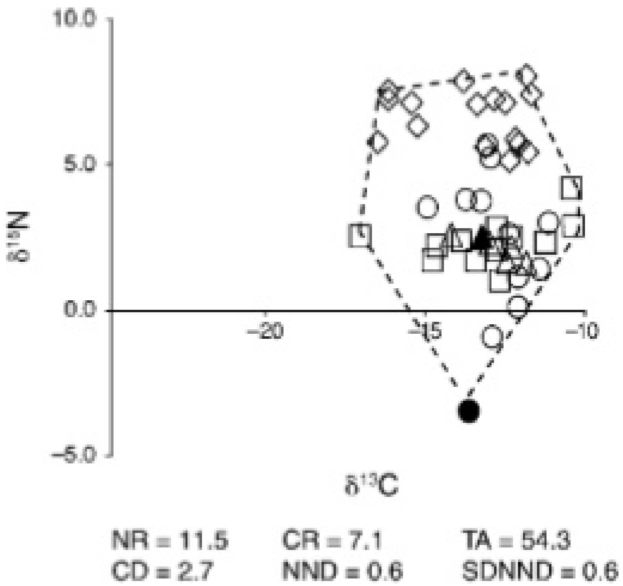
CAN STABLE ISOTOPE RATIOS PROVIDE FOR COMMUNITY-WIDE MEASURES OF TROPHIC STRUCTURE?

Craig A. Layman,[1,5] D. Albrey Arrington,[2] Carmen G. Montaña,[3] and David M. Post[4]

[1]Marine Science Program, Department of Biological Sciences, 3000 NE 151st Street, North Miami, Florida 33181 USA
[2]Loxahatchee River District, 2500 Jupiter Park Drive, Jupiter, Florida 33458-8964 USA
[3]Universidad de Los Llanos Ezequiel Zamora, UNELLEZ, Guanare, Apartado Postal 3310 Venezuela
[4]Department of Ecology and Evolutionary Biology, Yale University, New Haven, Connecticut 06520-8106 USA



Layman metrics: community-wide metrics



1. δ^{15}N range (NR)
2. δ^{1}3C range (CR)
3. Convex hull area (TA), niche width
4. Species space, distance to centroid (CD)
5. Density and clustering of species, mean nearest neigbourg distance (MNND)
6. Estándar deviation of MNND, evennness of spatial density and packing (SDNND)

Journal of Animal Ecology

Journal of Animal Ecology 2011, 80, 595–602 doi: 10.1111/j.1365-2656.2011.01806.x

Comparing isotopic niche widths among and within communities: SIBER – Stable Isotope Bayesian Ellipses in R

Andrew L. Jackson[1*], Richard Inger[2], Andrew C. Parnell[3] and Stuart Bearhop[2]

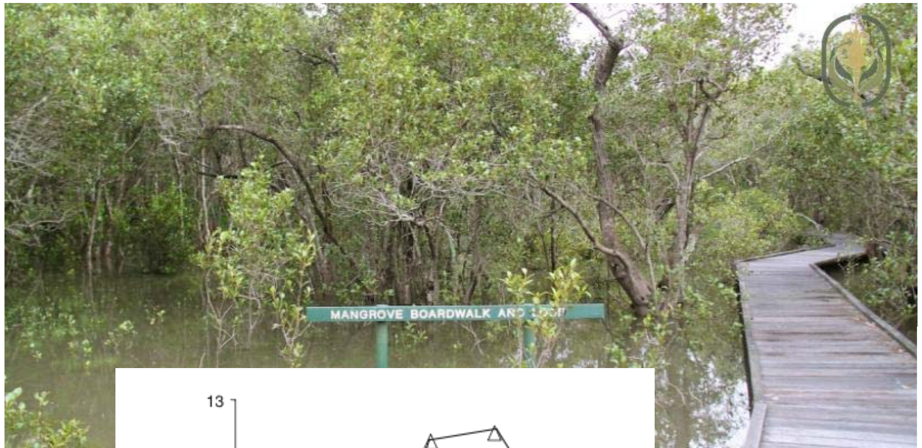
https://github.com/AndrewLJackson/SIBER

Conservation approaches: habitat framentation and changes in the trophic webs

Ecology Letters, (2007) **10**: 937–944 doi: 10.1111/j.1461-0248.2007.01087.x

LETTER

Niche width collapse in a resilient top predator following ecosystem fragmentation



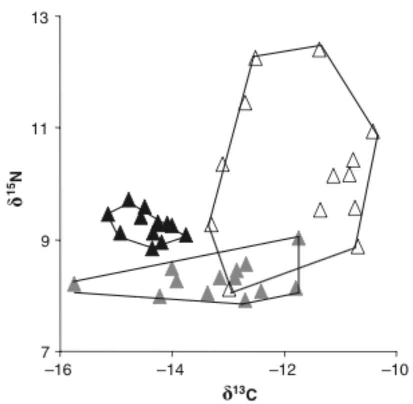


Figure 1 Each symbol represents an individual snapper and the lines represent the convex hull area used as a measure of niche width. White triangles are individuals from an unfragmented site, grey triangles from a partially fragmented site, and black triangles from a highly fragmented site (Cross Harbour, Sucking Fish, and Marsh Harbour, respectively, in Table 1).

Grey Snaper *(Lutjanus griseus)*

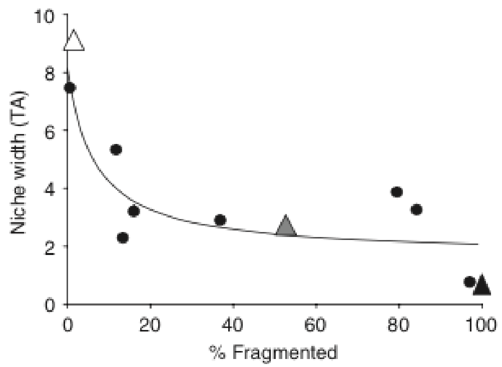
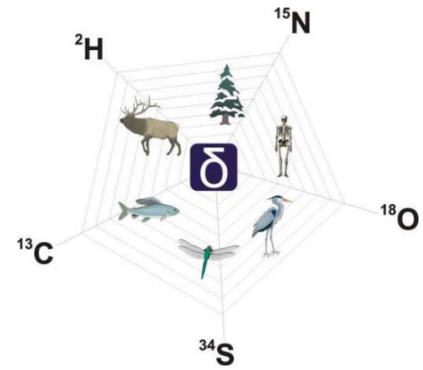


Figure 2 Niche width plotted as a function of percent fragmentation, with niche width estimated as convex hull area (TA) encompassing 13 individuals in each population. Each symbol represents the estimated niche width of a gray snapper population in one of 11 tidal creek systems, with triangles corresponding to the three sites depicted in Fig. 1.

So far…



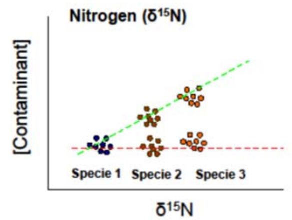
Intraespecific studies:

- Paleodiet reconstruction
- Long term changes in diet, evolutionary approach
- Spatio-temporal variation in trophic ecolgy
- Individual and population specialisation level
- Nutrient allocation

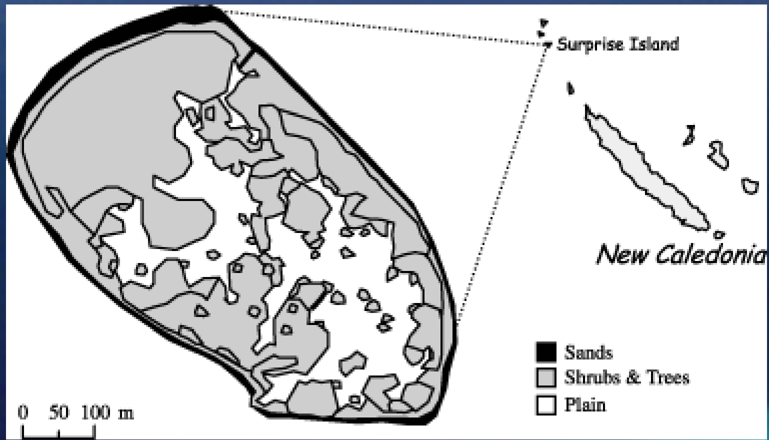
Community studies: Layman metrics, SEA, TA (SIBER)

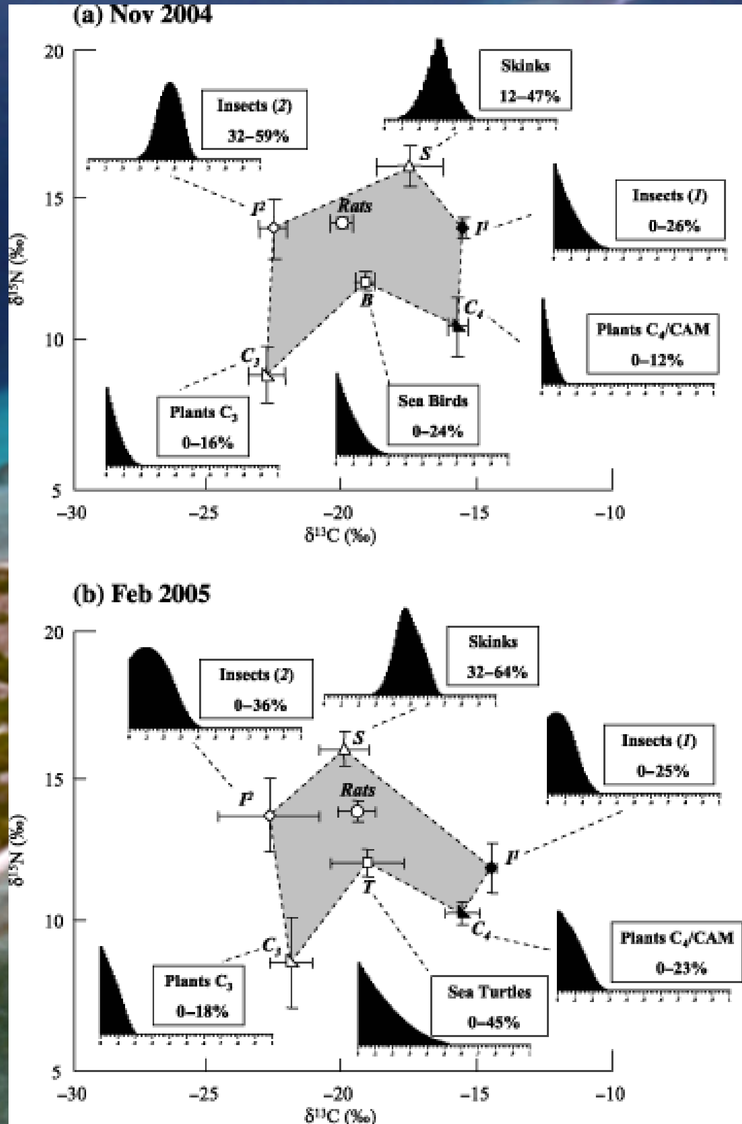
Conservation approaches:

- Use of antrophogenic feeding habitats
- Biomagnification and bioaccumulation
- Impact of introduced predators



Conservation approaches: impact of introduced predators on native species and habitats



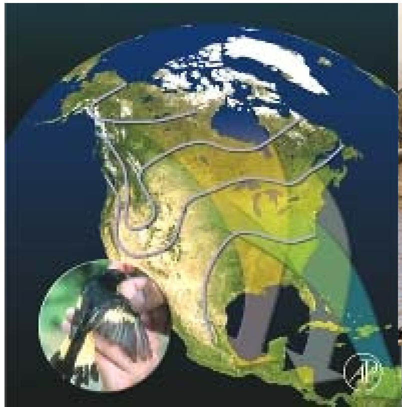


Caut *et al.* 2008. *J Applied Ecol*

Why should we use them (CNS, HO)?

- Trophic and nutricional ecology, food webs

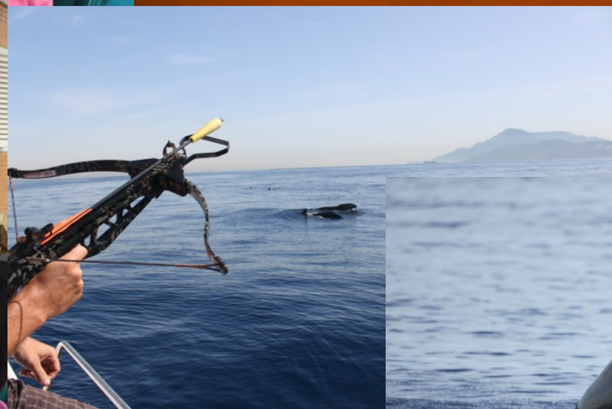
- Animal movement, migration research



Traditional way of study animal movement: extrinsic markers



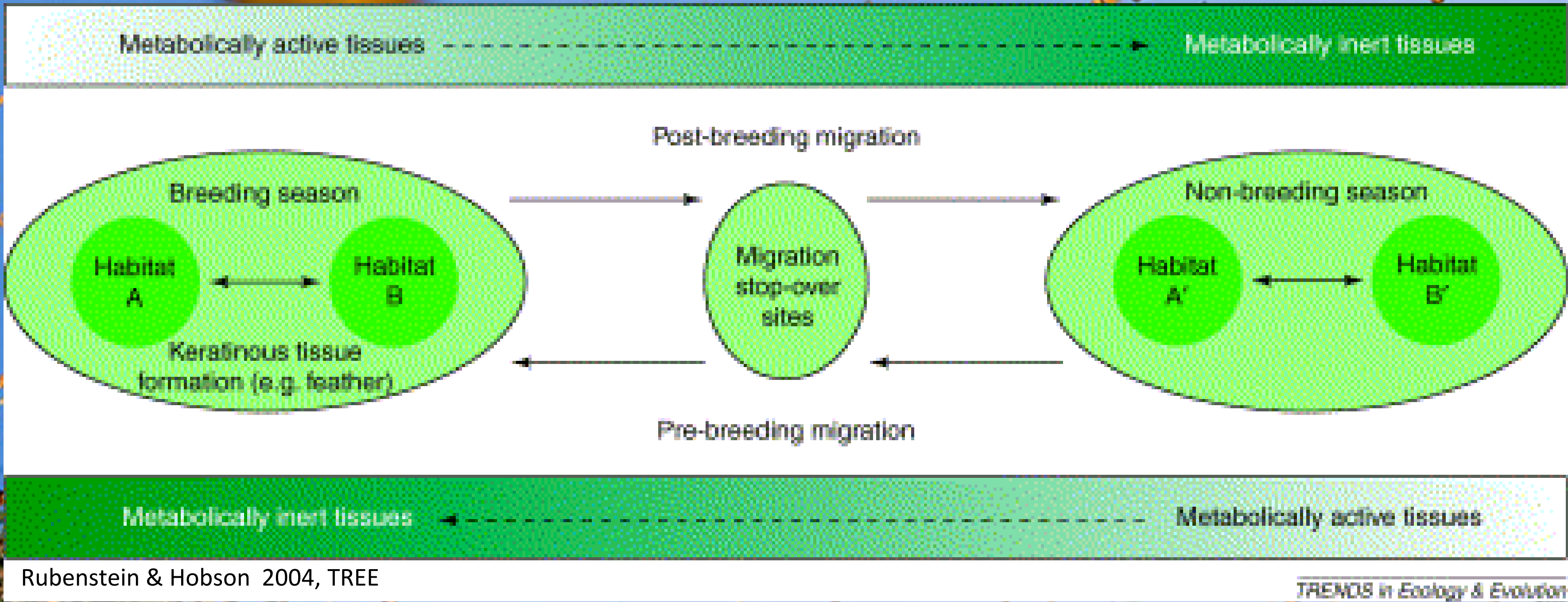


Problems /SIA advantages:

- Organism often needs to be recoverd /No recovery
- Expensive/No very expensive
- Body size requierements /Any species and size
- Biased to original marked population/Capture is a recapture

Application of stable isotopes to study animal movement: the isotopic clock



Rubenstein & Hobson 2004, TREE

Early attemps to study migration by stable isotopes

"You are where you swim in"

California gray whales (*Eschrichtius robustus*)

$\delta^{18}O$ in barnacles (water temperature and salinity)

Killingley 1980, *Science*

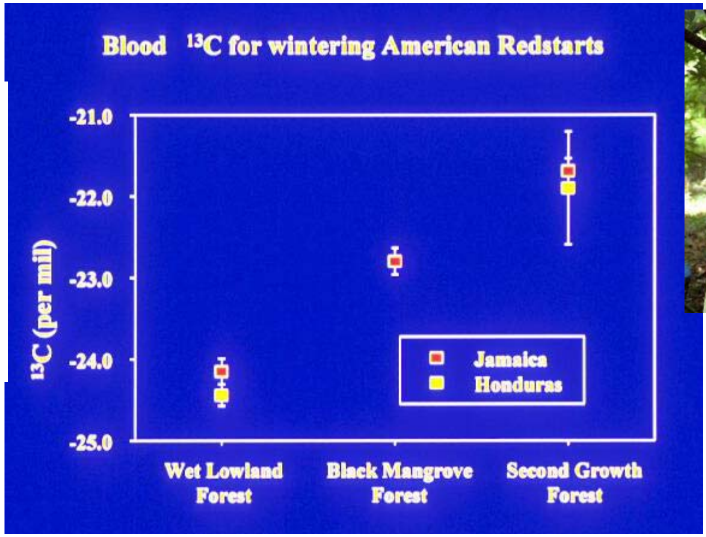
Longgerhead turtle (*Caretta caretta*)

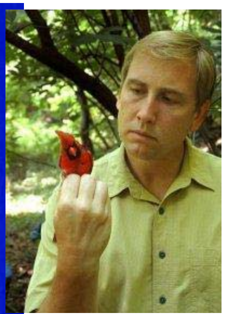
$\delta^{13}C$ and $\delta^{18}O$ (estuarine vs coastal waters)

Killingley and Lutcavage 1983, *Coastal Shelf Sci*

Setophaga ruticilla

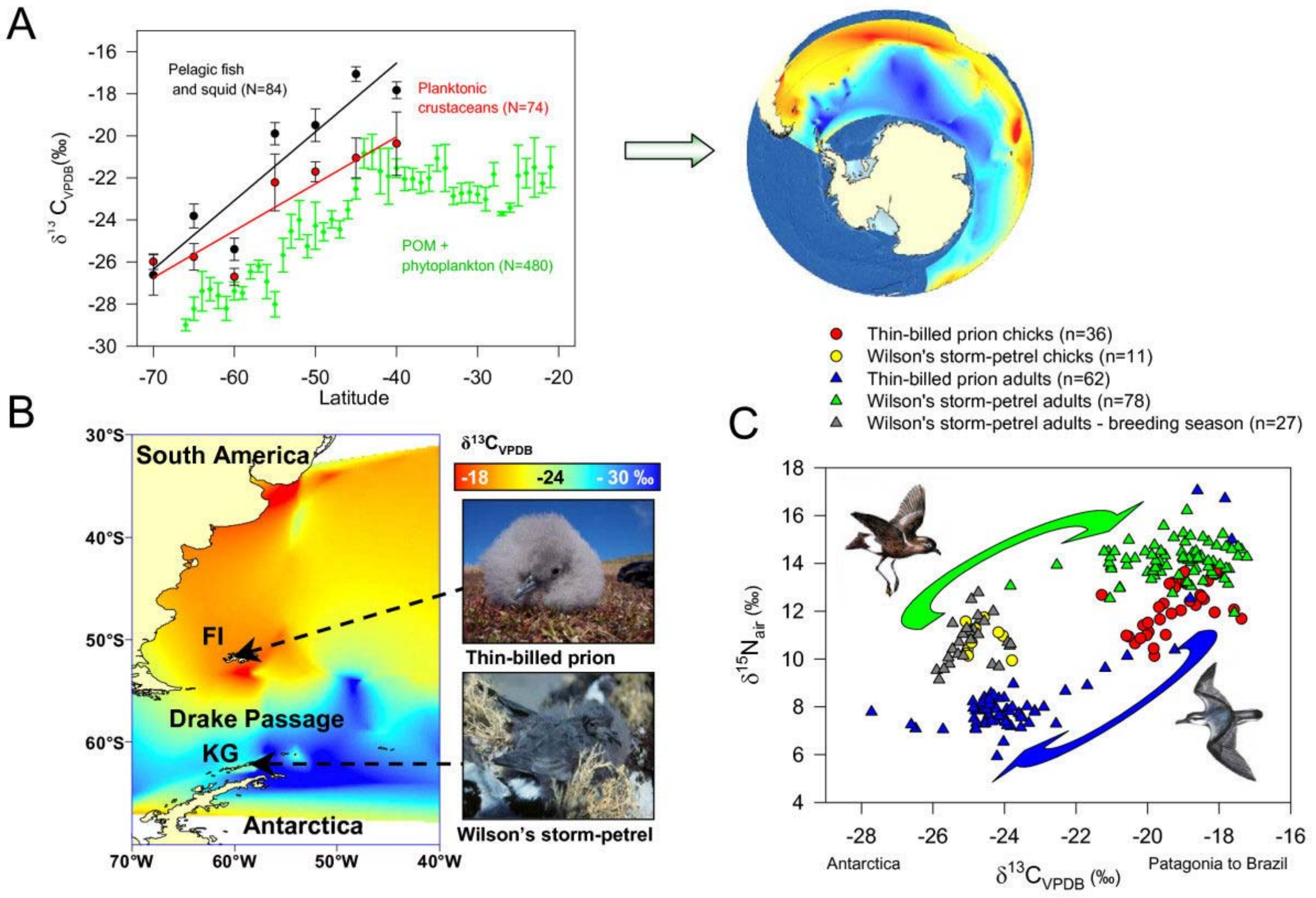
$\delta^{13}C$, wintering habitat determine arrival time on breeding grounds





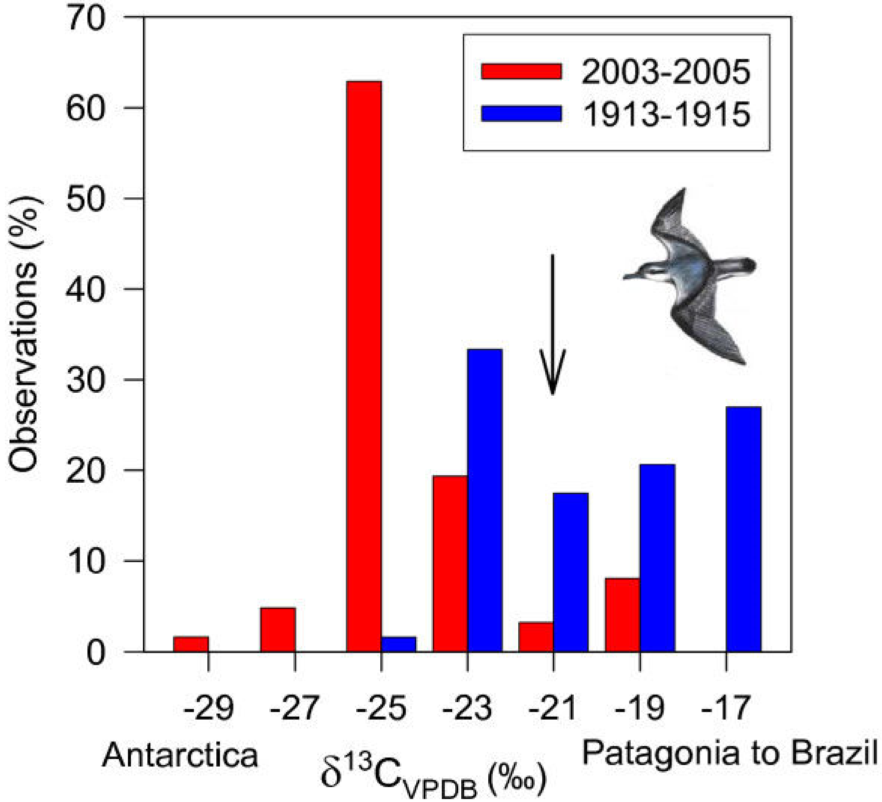
Marra et al. 1998, *Science*

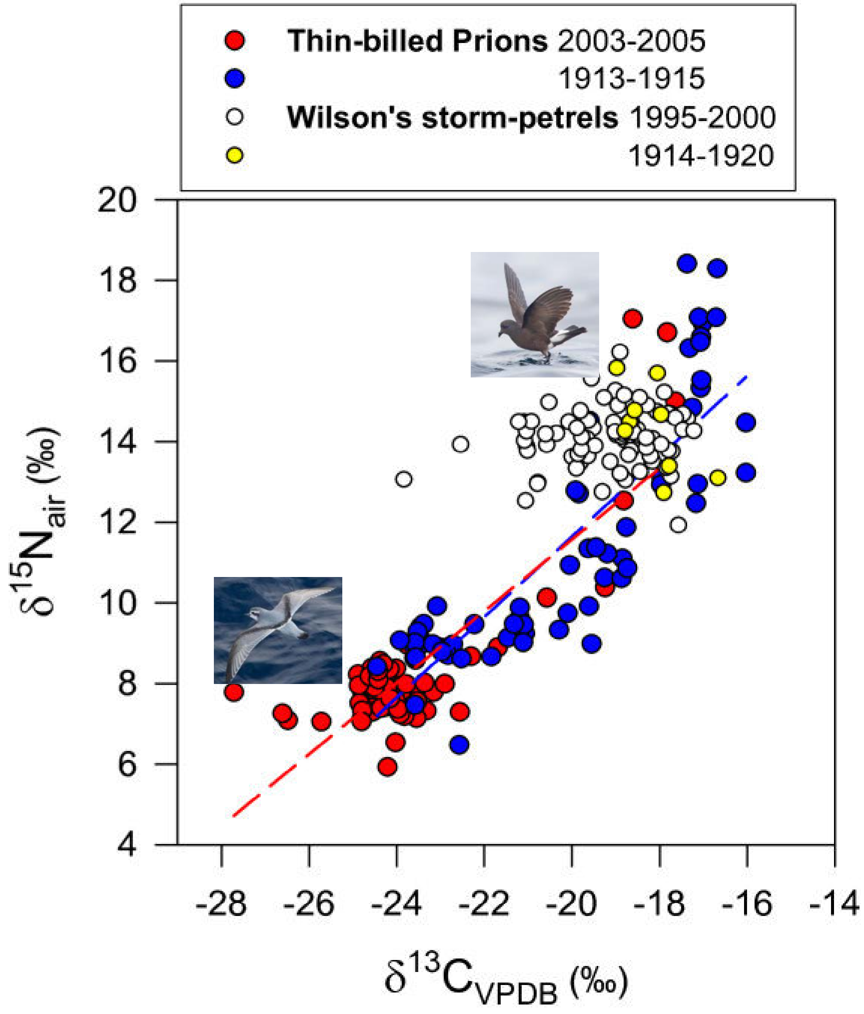
More recent approaches in the marine environment: changes in migratory strategies



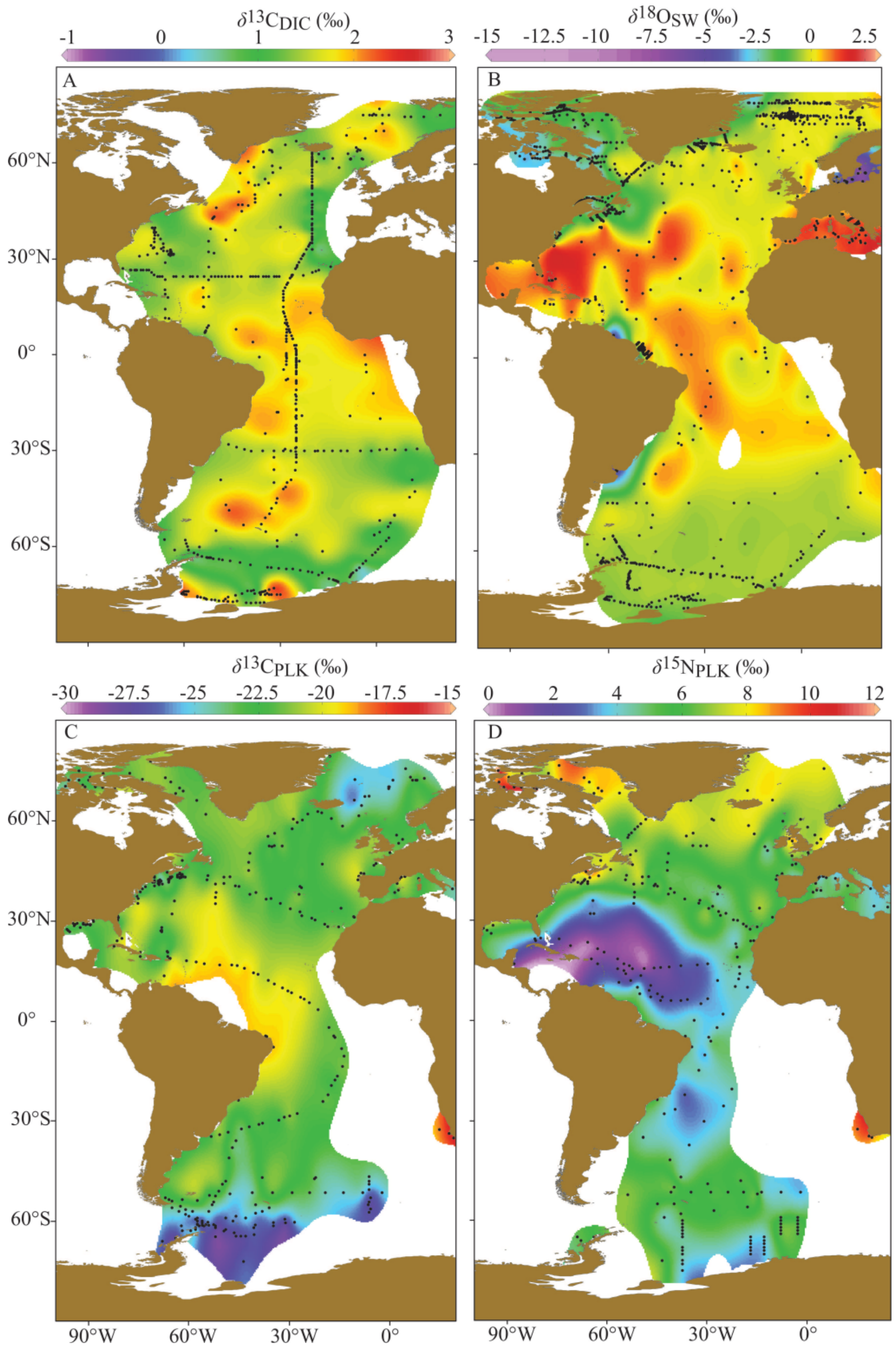
Quillfeldt et al. 2010 *Frontiers in Zool*

More recent approaches in the marine environment: rapid changes in migratory strategies

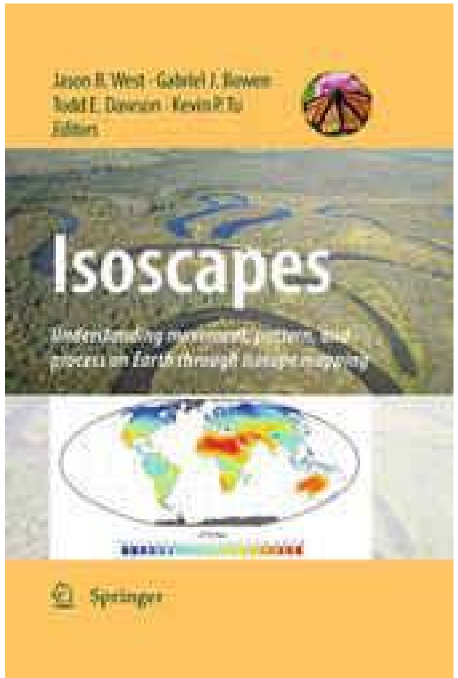




Quillfeldt et al. 2010 *Frontiers in Zool*



More recent marine Isoscapes



McMahon et al. 2013, *Limnol. Oceanogr*

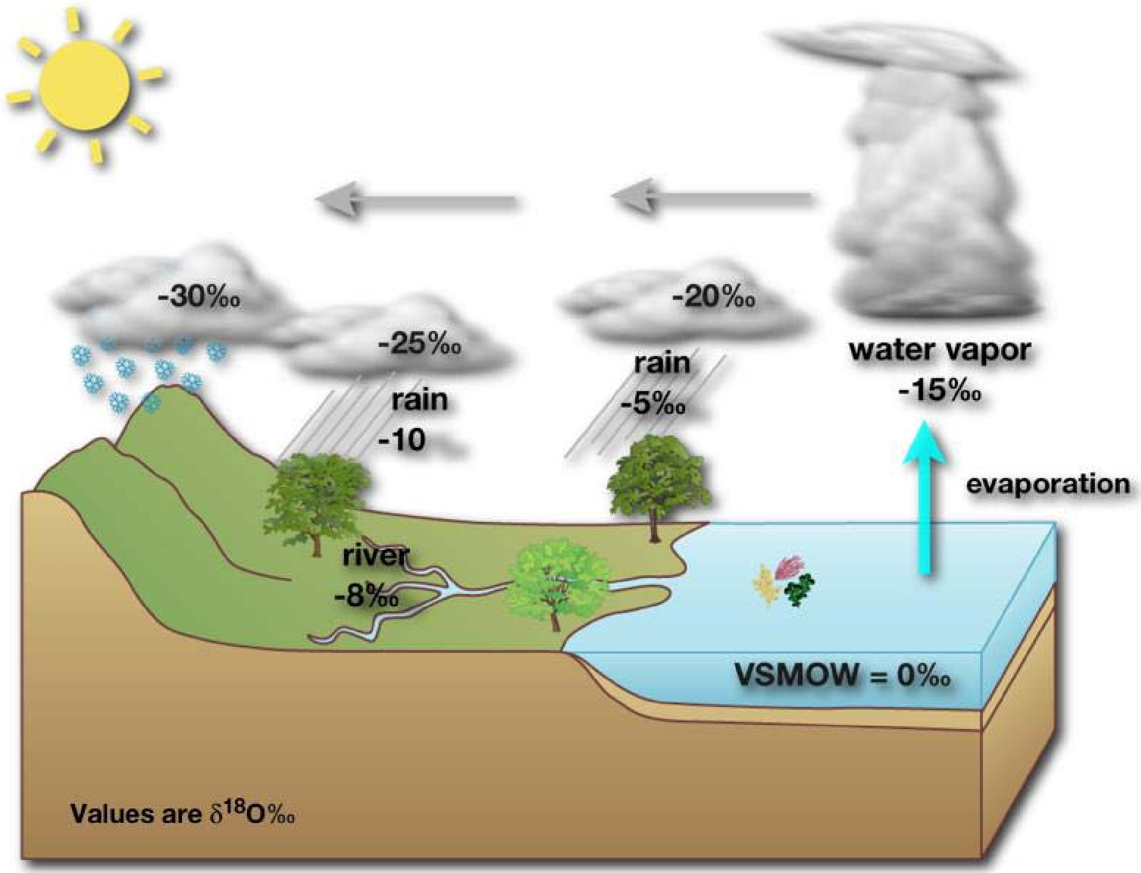
Application of stable isotopes to study animal movement: Water, δD and $\delta^{18}O$ Isoscapes

High elevation
High latitud

Light ← Heavy

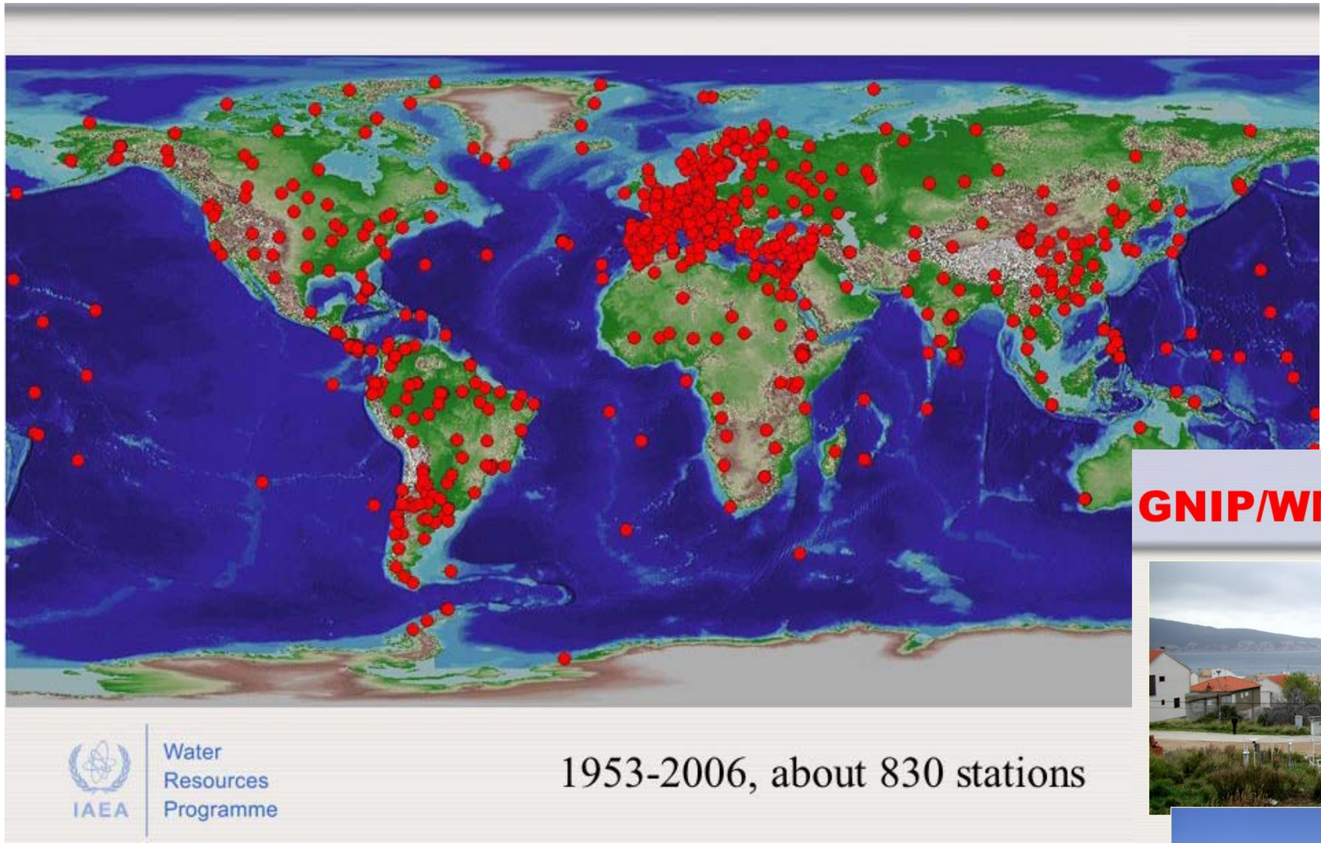
Low elevation
Low latitud

Light ↕ Heavy



Partitioning of isotopes in Vapor and Precipitation

Application of stable isotopes to study animal movement: Water, δD and $\delta^{18}O$ Isoscapes



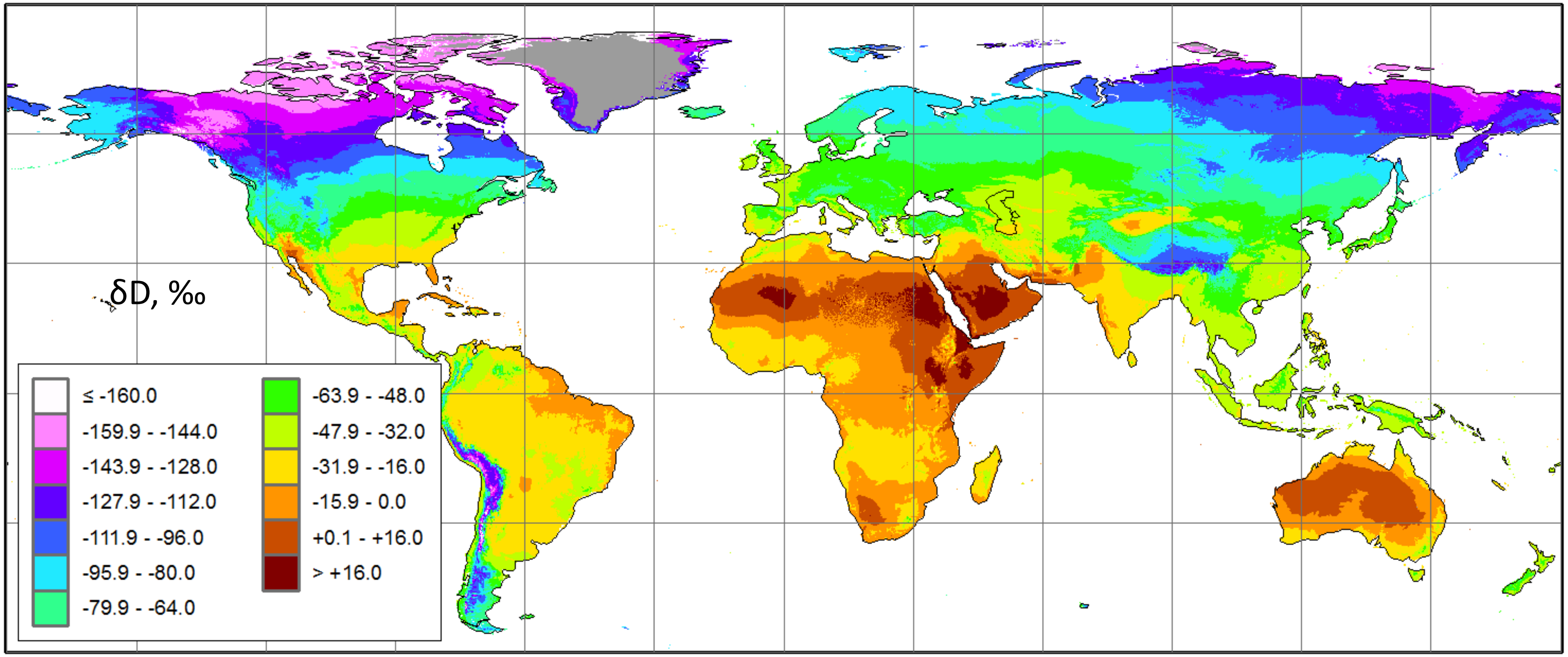
Isotopic content in precipitation, monthly basis (Global Network of Isotopes in Precipitation, GNIP)

The IAEA's Water Resources Programme and the World Meteorological Organization (WMO)

http://www-naweb.iaea.org/napc/ih/IHS_resources_gnip.html

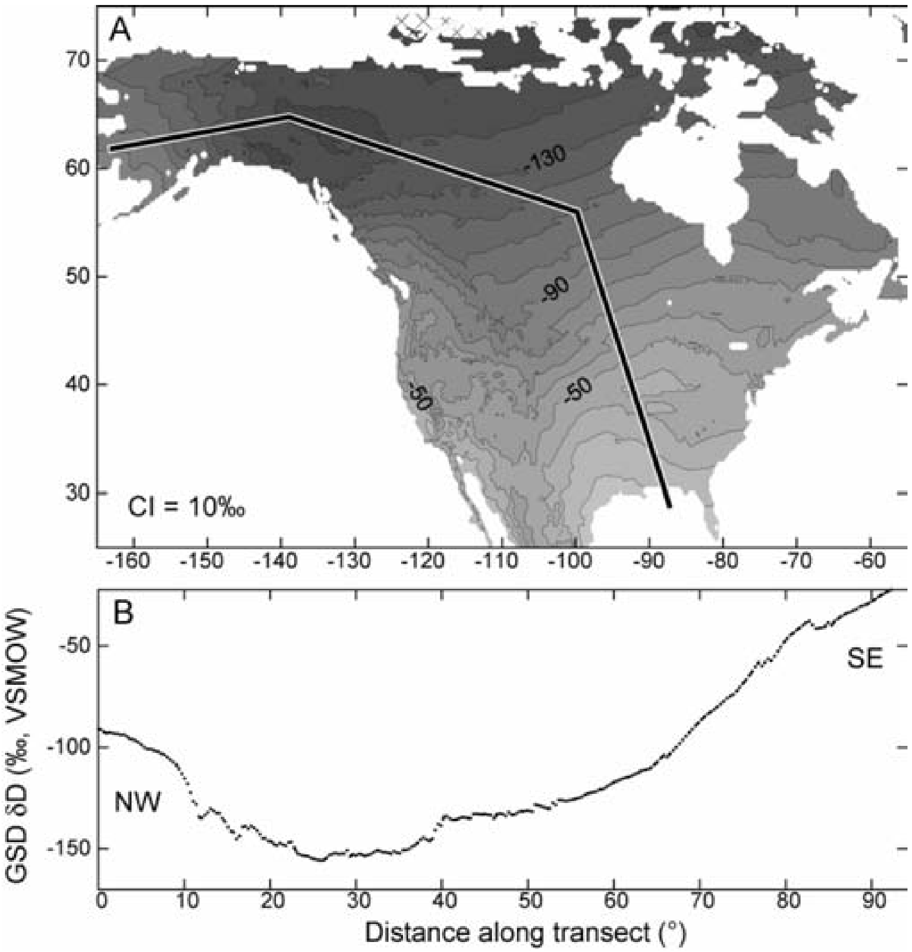
Application of stable **isotopes** to study animal movement: Water, δD and $\delta^{18}O$ Isoscapes

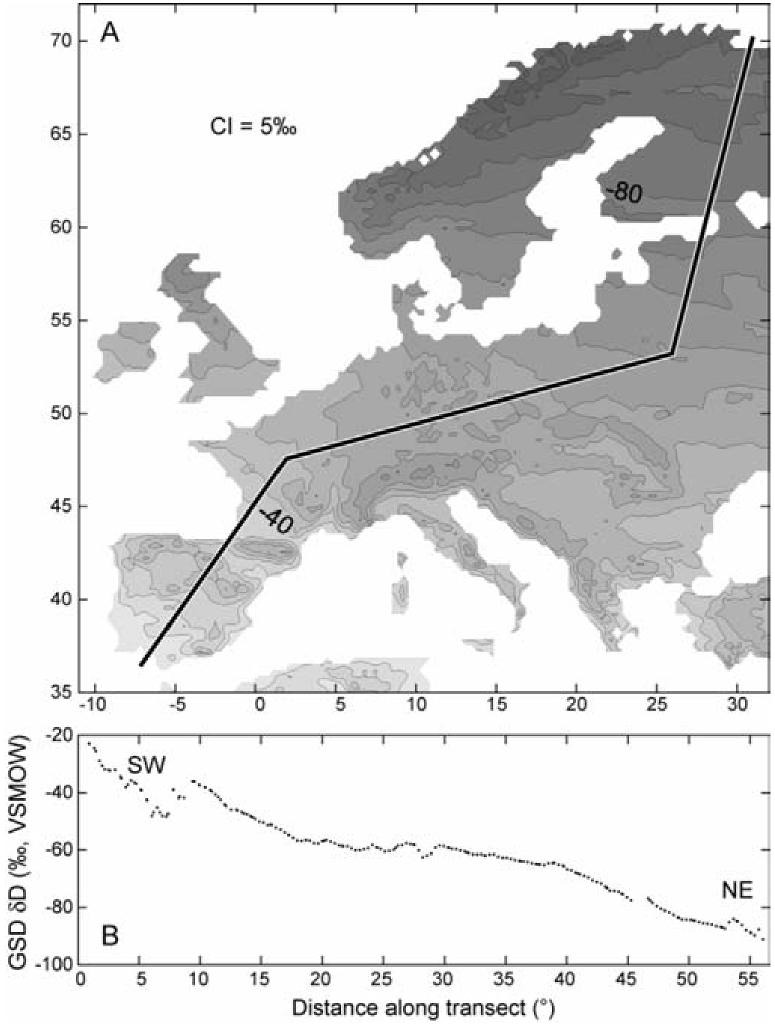
- Water Resource Program of IAEA Stable Isotope Laboratory (analyse data from 40% of GINP stations, rest other labs)
- Detrended Interpolation models (latitud, longitud, smothing algorithm): Spacially continous grids (*Bowen et al. 2003, Water Resour Res*)



www. waterisotopes.org

Global application of stable hydrogen and oxigen isotopes to wildlife forensic

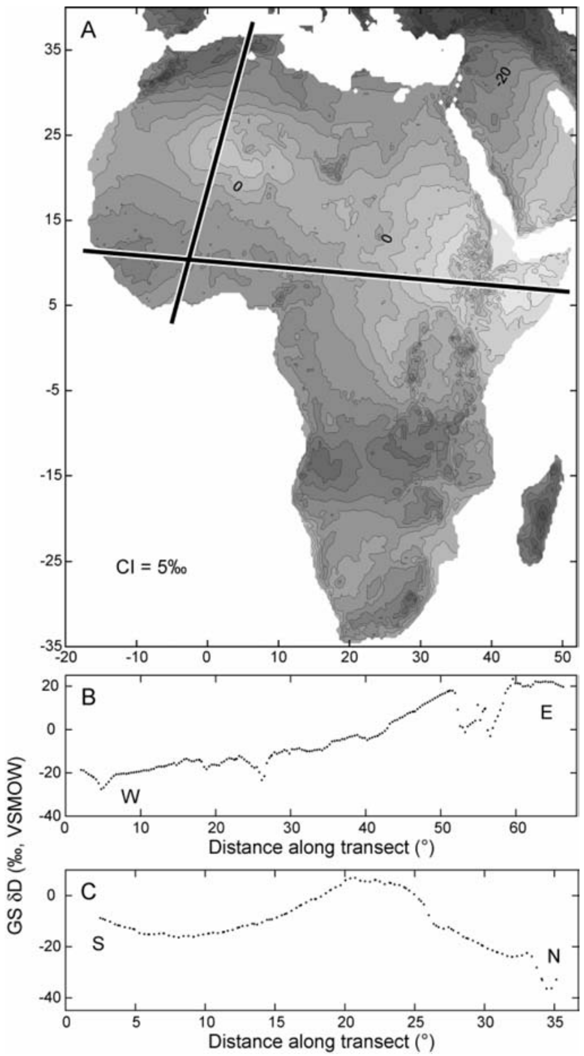


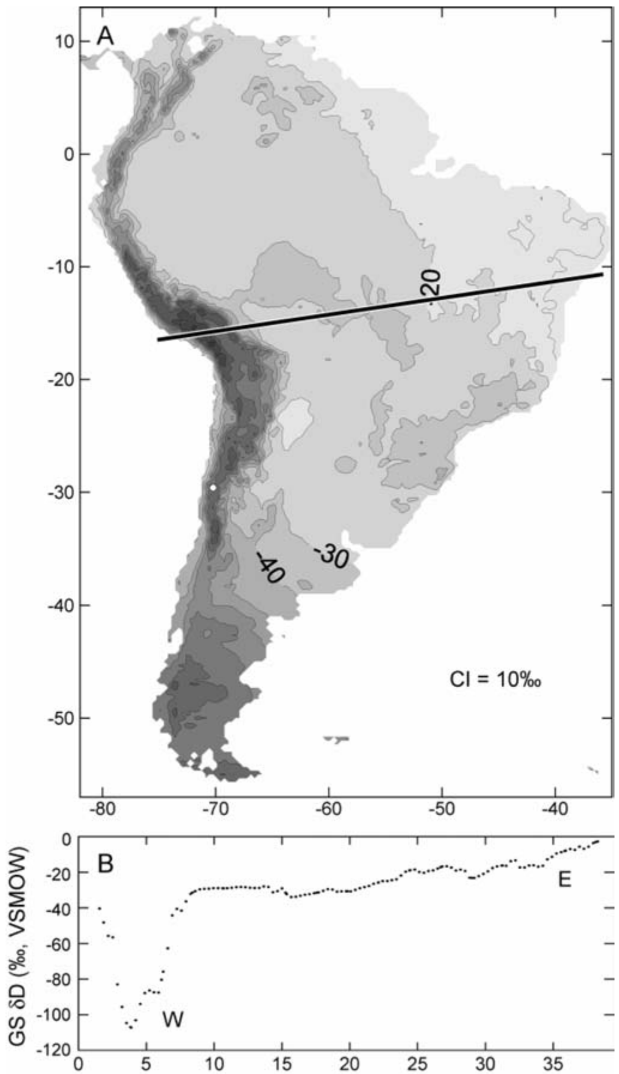


Latitudinal better than longitudinal

Bowen *et al.* 2005, *Oecologia*

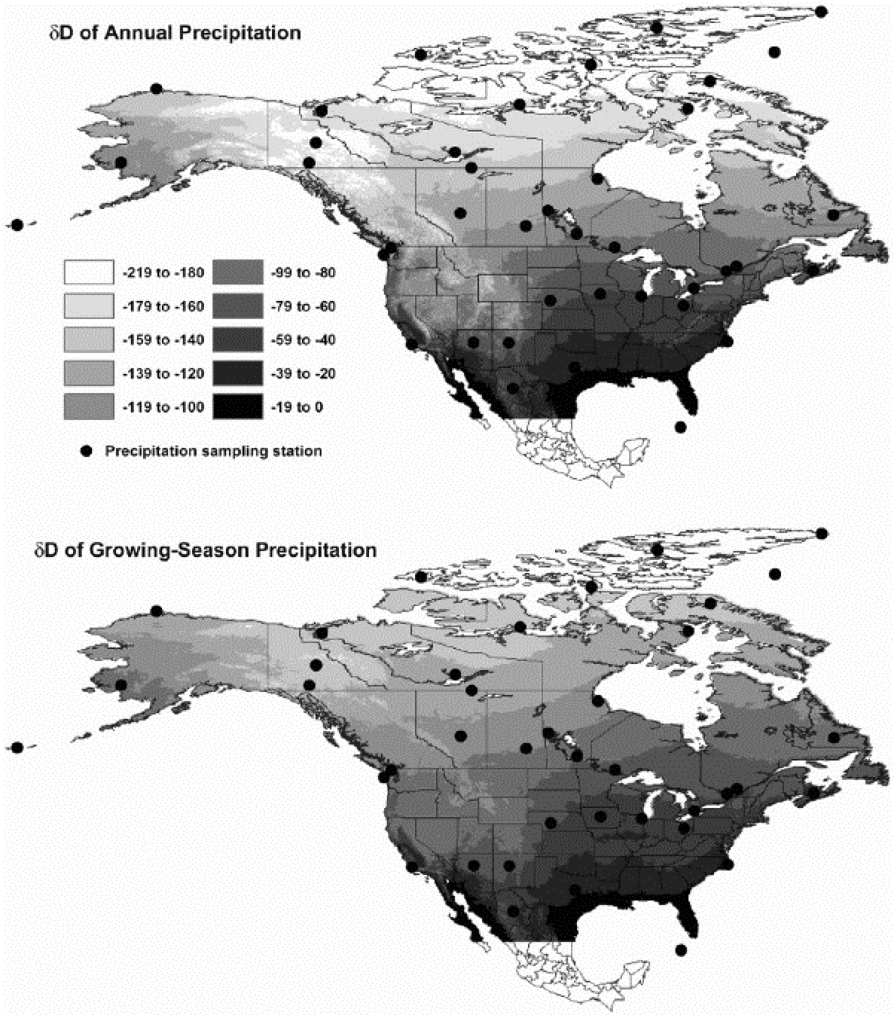
Global application of stable hydrogen and oxigen isotopes to wildlife forensic

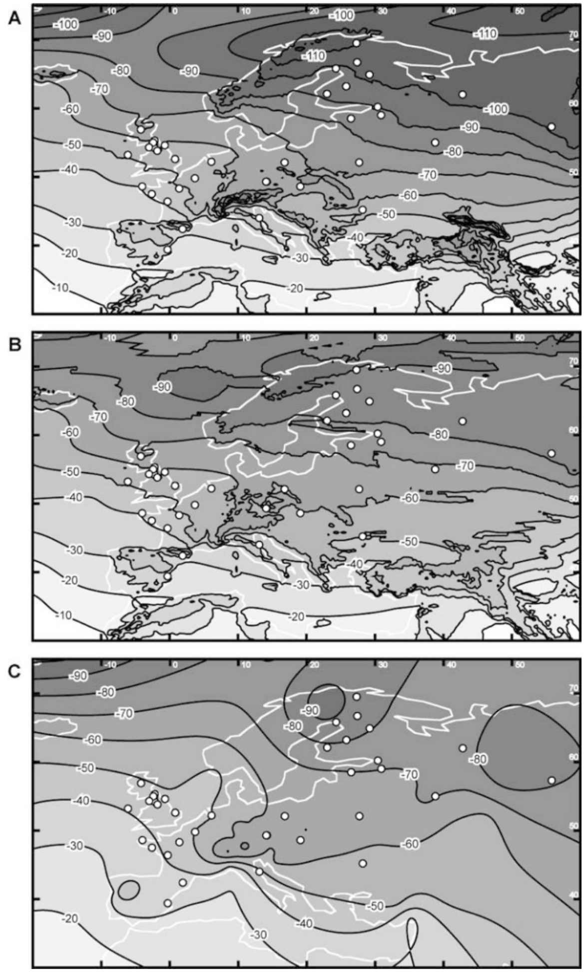




Bowen *et al.* 2005, *Oecologia*

Global application of stable hydrogen and oxigen isotopes to wildlife forensic: refining gradients, elevational models





δD $_p$ mean annual elevational explicit model

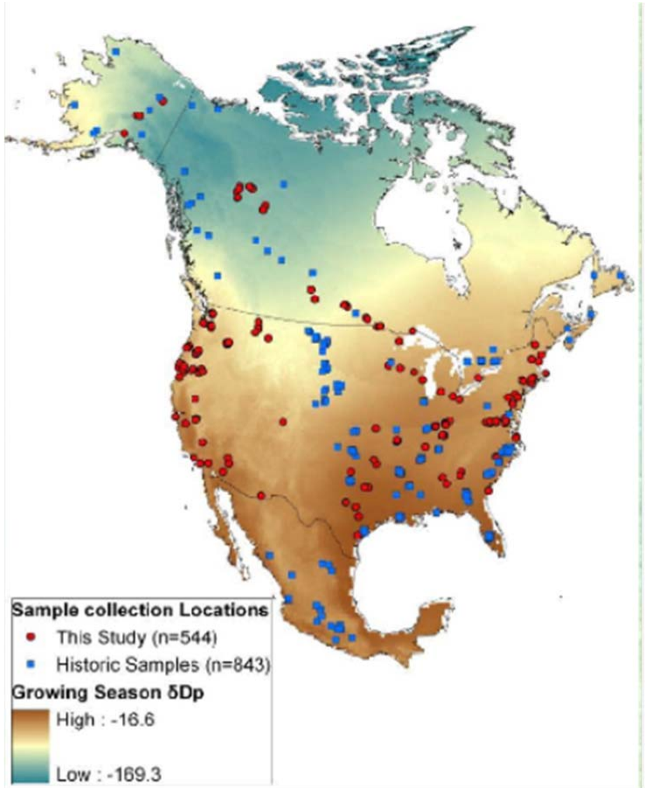
δD $_p$ growing season elevational explicit model

δD $_p$ growing season simple Kriging, no elevation effect

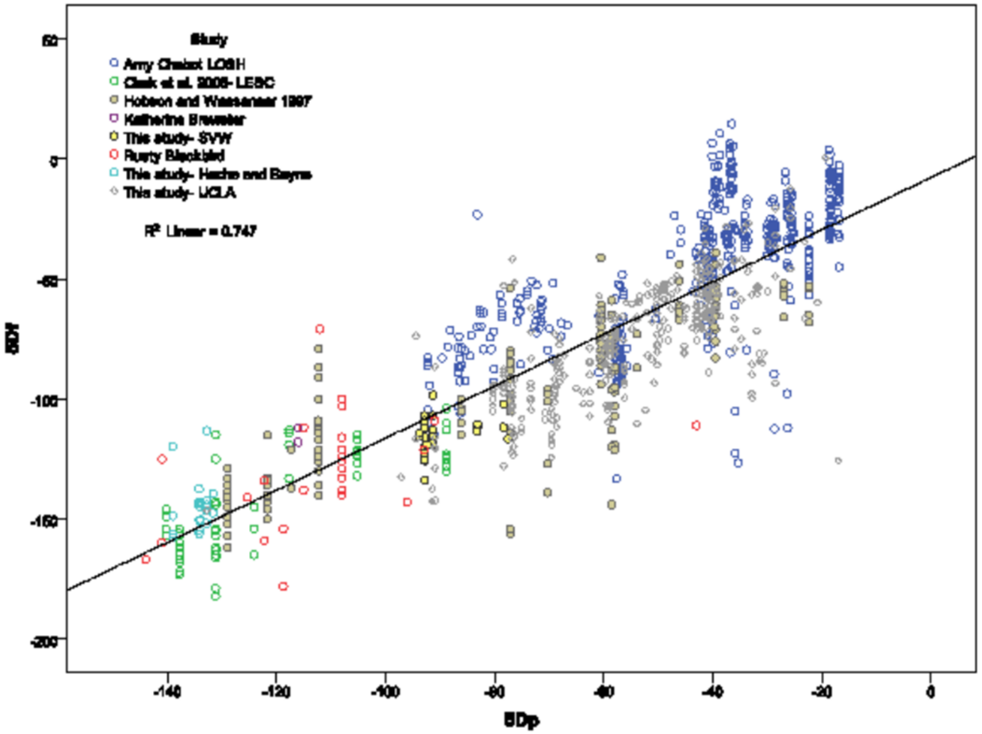
Meehan et al. 2004, *Isotopes Environm Health Stud, North America*
Hobson et al. 2004, *Oecologia, Europe*

Stable Isotopes and Migratory Connectivity: δD_p and δD_f

First studies in Norht America:



Chamberlain *et al.* 1997
Hobson & Wassenaar 1997



Discrimination factor: -25—30‰

Stable Isotopes and Migratory Connectivity: δD_p and δD_f

Species	Equation	r^2	Model	Source
Birds:				
6 species of North American songbird	δD=-31 + 0.9δDp	0.83	H	Hobson and Wassenaar (1997)
6 species of North American songbird	δD=-25 + 0.9δDp	0.88	B	Clark et al. (2006)
6 species of North American songbird	δD=-19.4 + 1.07δDp	0.86	B	Bowen et al. (2005)
Black-throated Blue Warbler	δD=-51 + 0.5δDp	0.86	CH	Chamberlain et al. (1997)
Red-winged blackbird	δD=-27 + 1.1δDp	0.83	H	Wassenaar and Hobson (2000)
Bicknell's Thrush	δD=-26 + 0.7δDp	0.48	H	Hobson et al (2001)
Wilson's Warbler	δD=-51.7 + 0.4δDp	0.36	B	J. Kelly (unpublished)
Wilson's Warbler	δD=+14.47 + 1.41δDp	0.91	M	Paxton et al. (2007)
Wilson's Warbler	δD=-21 + 0.7δDp	0.48	M	Meehan et al. (2004)
Mountain Plover	δD=+17.4 + 1.26δDp	0.36	B	Wunder (2007)
23 species of European birds	δD=-7.8 + 1.27δDp	0.65	B	Hobson et al. (2004d)
23 species of European birds	δD=-22.3+0.77δDp	0.85	B	Bowen et al. (2005)
Cooper's Hawk	δD=-34 + 1.0δDp	0.83	H	Meehan et al. (2001)
Inland generalist raptors	δD=-40+0.62δDp	0.59	H	Lott et al. (2003)
Inland bird-eating raptor	δD=-44.2+0.54δDp	0.37	H	Lott et al. (2003)
Coastal generalist raptors	δD=-38.8+0.55δDp	0.19	H	Lott et al. (2003)
Coastal bird-eating raptors	δD=-104.7-0.59δDp	0.12	H	Lott et al. (2003)
Non-coastal bird-eating raptors	δD=-41.1+0.58δDp	0.46	H	Lott et al. (2003)
9 species of raptors	δD=-52.2+0.28δDp	0.09	H	Lott et al. (2003)
9 species of diurnal raptors	δD=-37 + 0.6δDp	0.51	M	Meehan et al. (2004)
Raptors in South Carolina	δD=-25 + 0.7δDp	0.18	M	Meehan et al. (2004)
Flammulated Owl	δD=-8 + 0.9δDp	0.66	M	Meehan et al. (2004)
12 species of raptors	δD=-5.6 + 0.91δDp	0.62	M	Lott and Smith (2006)
Scaup	δD=-27.8 + 0.9δDp	0.64	B	Clark et al. (2006)
Mallards and Northern Pintail	δD=-57 + 0.83δDp	0.56	M	Hebert and Wassenaar (2005)
Other animals:				
Deer collagen	δD=4 + 1.02δDp	0.94	C	Cormie et al. (1994)
Hoary bat	δD=-25 + 0.8δDp	0.60	M	Cryan et al. (2004)
Monarch butterfly	δD=-79 + 0.62δDp	0.69	H	Hobson et al. (1999)
Beetle (chitin)	δD=33.2 + 1.60δDp	0.74	B	Gröcke et al. (2006)

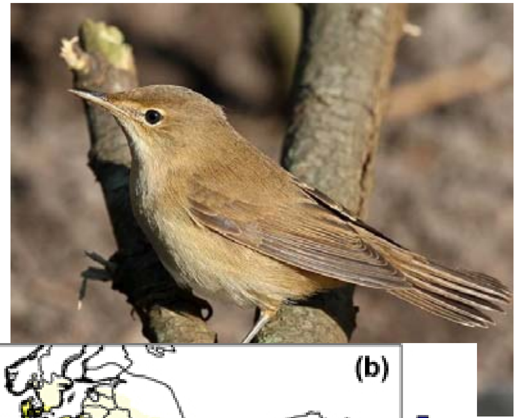
Best model: migratory guild, foraging substrate. Further, effect of year, age…, still working

Journal of Avian Biology 44: 541–550, 2013
doi: 10.1111/j.1600-048X.2013.00185.x

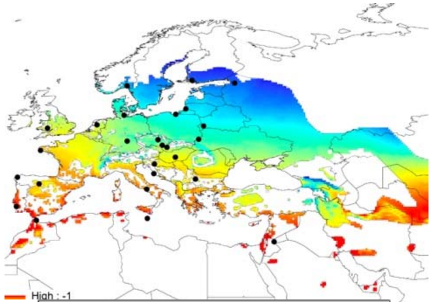
Subject Editor: Staffan Bensch. Accepted 27 July 2013

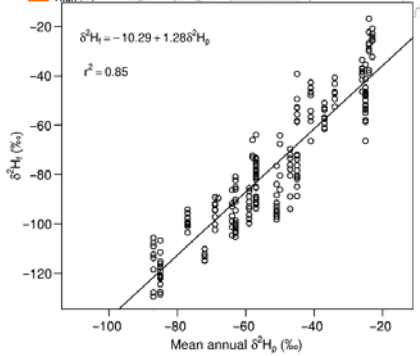
Using stable hydrogen isotopes (δ^2H) and ring recoveries to trace natal origins in a Eurasian passerine with a migratory divide

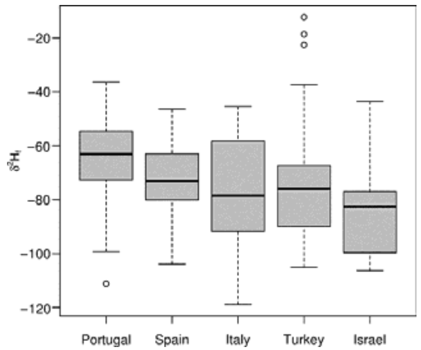
Petr Procházka, Steven L. Van Wilgenburg, Júlio M. Neto, Reuven Yosef and Keith A. Hobson

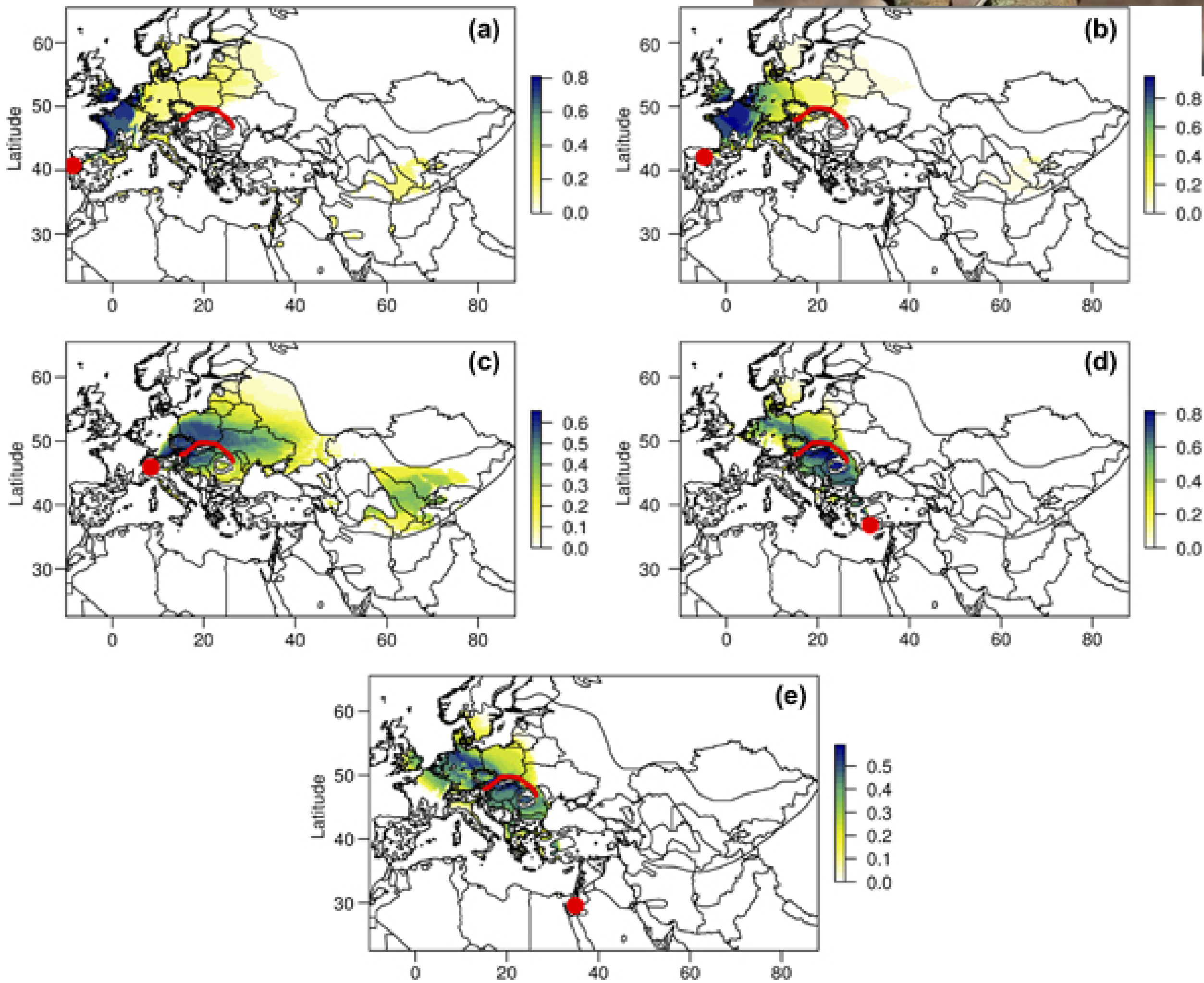


Acrocephalus schoenobaenus









Procházka *et al. J Avian Biol* 2013

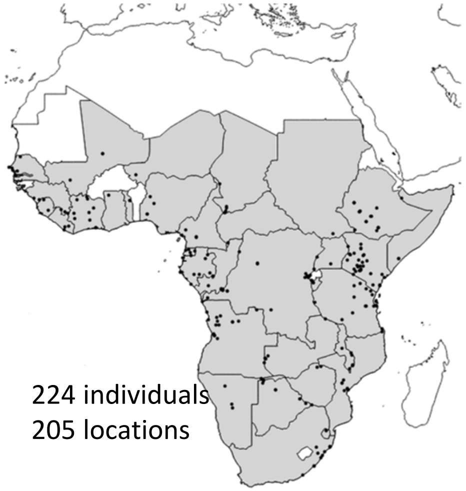
PLOS ONE

RESEARCH ARTICLE

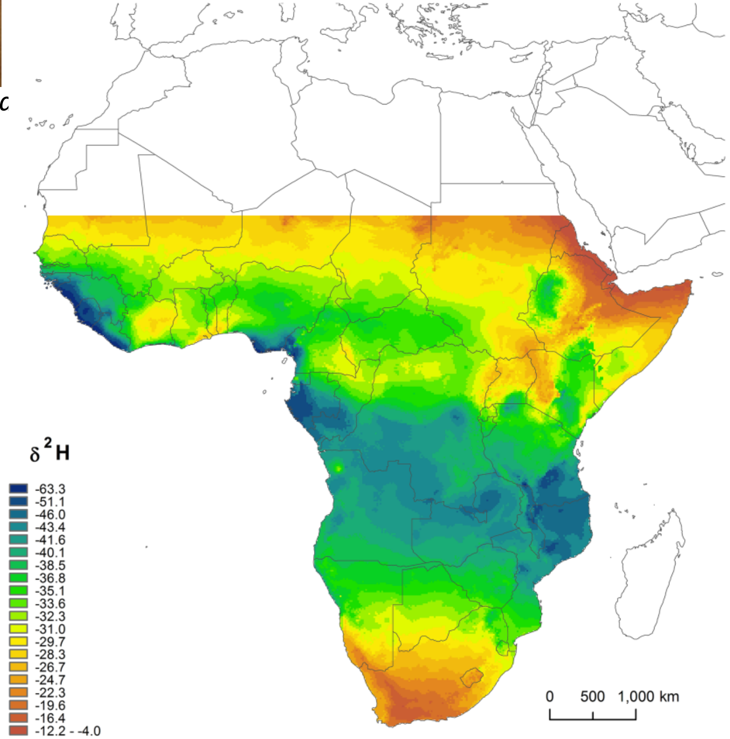
Toward a Deuterium Feather Isoscape for Sub-Saharan Africa: Progress, Challenges and the Path Ahead

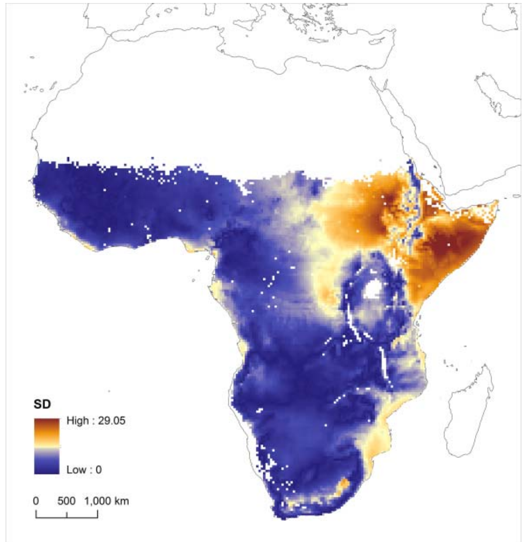
Carlos Gutiérrez-Expósito[1]*, Francisco Ramírez[1], Isabel Afán[2], Manuela G. Forero[1], Keith A. Hobson[3]

Camaroptera brachyura/brevicaudata



224 individuals
205 locations



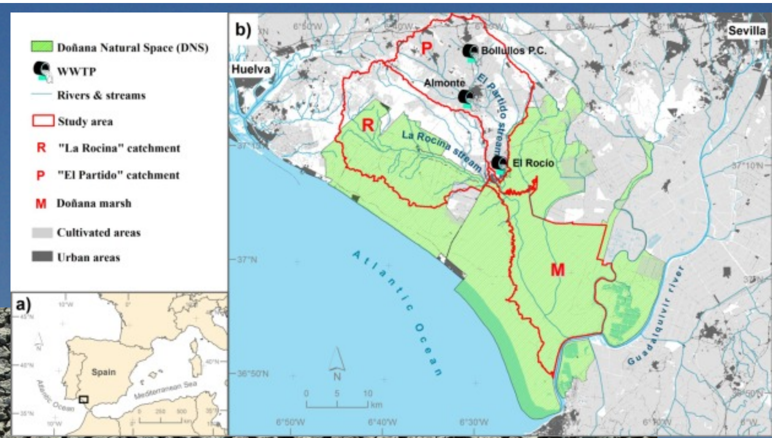


Hobson et al. 2012. *Ecosphere*

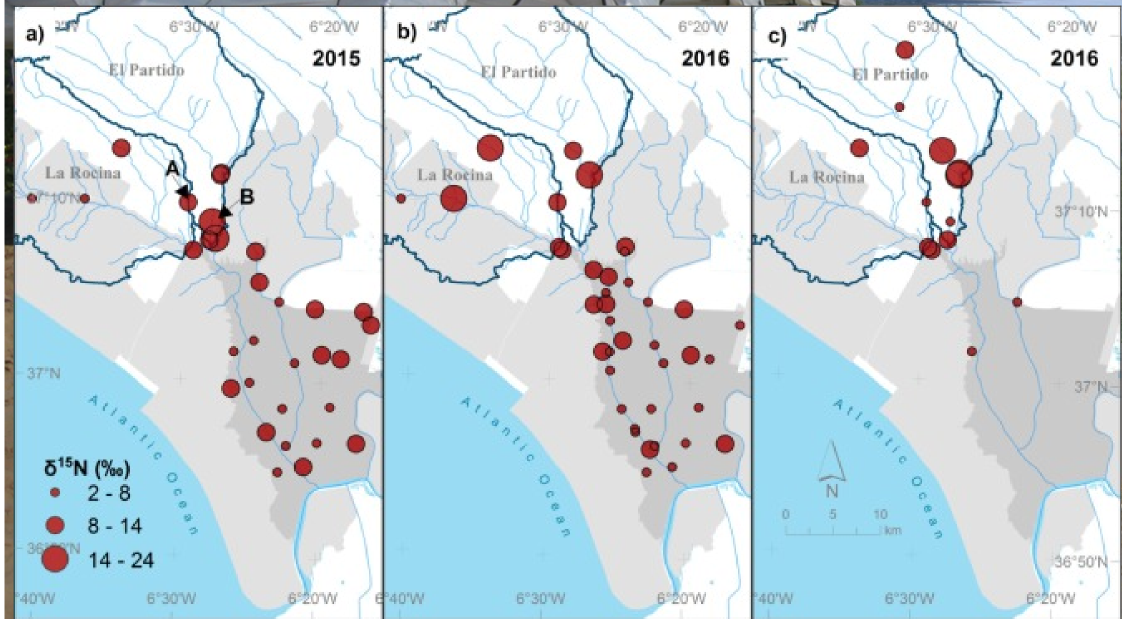
Gutiérrez-Expósito *et al.* 2015. *PLOS one*

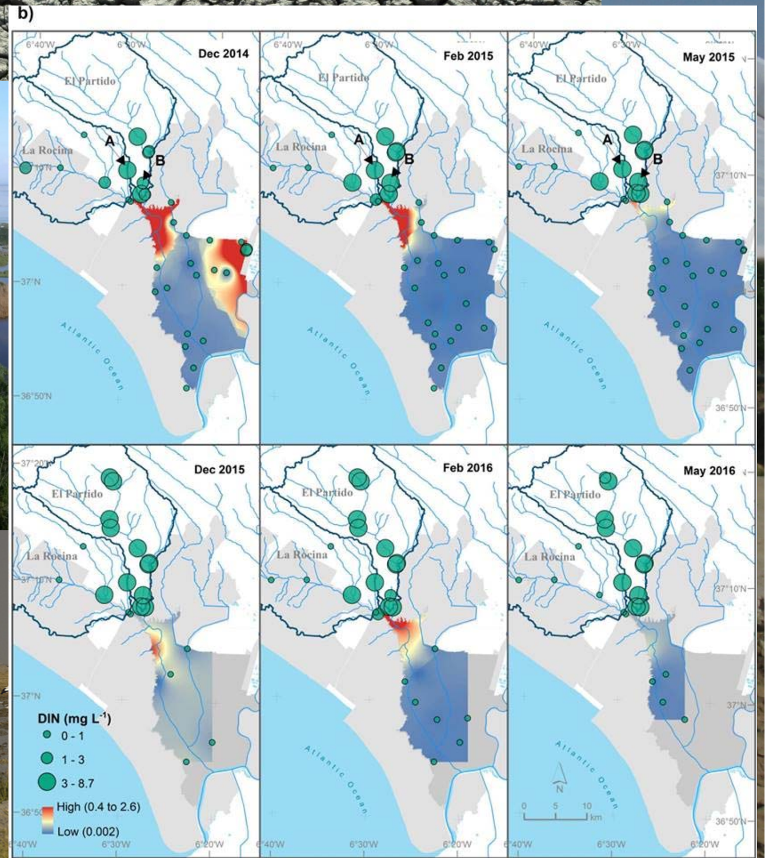
Other Isoscape studies (Doñana):

- Water dynamic (δ^2H y $\delta^{18}O$)
- Imputs of antrophogenic $\delta^{15}N$



Paredes et al. 2018. *Ecol. Applic.*





Stable Isotopes beyong limits of ecology

thermoscientific

Isótopos Estables, Metodologías y Aplicaciones – Reunión de Usuarios IRMS 2017

Hotel Sevilla Macarena

18-19 de Octubre
Sevilla

PROGRAMA

Miércoles 18 Octubre

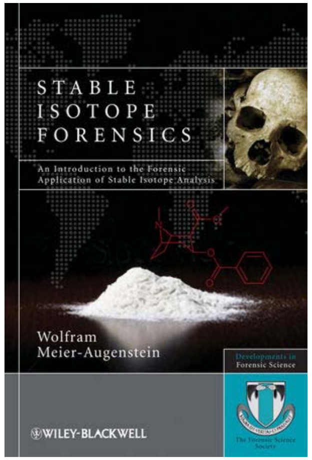
21:00	**Cóctel de bienvenida.** Bar El Pasaje Sevilla (Calle Pje. de Vila, 10, 41004 Sevilla)

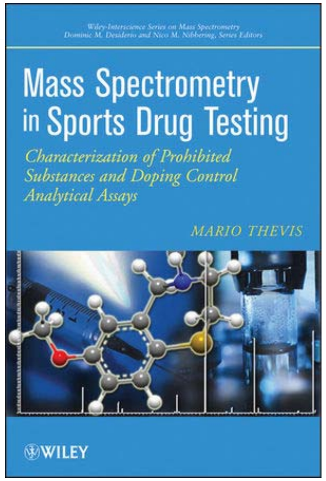
Jueves 19 Octubre

10:00	**Bienvenida – Café**
10:15	**Introducción.** Ricardo Alvarez – Thermo Fisher Scientific
10:30	**The Delta Ray Connect (Isotope Ratio Infrared Spectrometer).** Magda Mandic – Thermo Fisher Scientific
11:00	**Biomarcadores e Isótopos Estables en Astrobiología.** Daniel Carrizo - Centro de Astrobiología (INTA-CSIC)
11:30	**Trazabilidad de productos ecológicos utilizando isótopos estables.** José Manuel Moreno Rojas - Instituto de Investigación y Formación Agraria y Pesquera (IFAPA)
12:00	**Unidad de Espectrometría de Masas del SCAI de la Universidad de Málaga: Desarrollo de un método para la determinación de δ^2H y $\delta^{18}O$ en aguas de alto contenido salino.** Pedro Cañada Rudner; Sara Fernández-Palacios Campos - Servicios Centrales de Apoyo a la Investigación. Universidad de Málaga
12:30	**Paleoclimatología basada en relaciones isotópicas de carbonatos continentales: Aplicaciones en alta resolución.** Javier Martín-Chivelet - Departamento de Estratigrafía - Facultad de Ciencias Geológicas & Instituto de Geociencias (CSIC-UCM). Universidad Complutense de Madrid
13:00	**Aplicación de la composición isotópica del nitrato ($\delta^{15}N$ y $\delta^{18}O$) disuelto para evaluar la eficiencia de actuaciones de desnitrificación inducida a escala de campo.** Albert Soler Gil - Departament de Cristal·lografia, Mineralogia i Dipòsits Minerals Universitat de Barcelona (UB)
13:30	**Almuerzo**
14:30	**What's new in IRMS.** Lionnel Mounier – Thermo Fisher Scientific
15:00	**Análisis isotópico directo de compuestos específicos mediante pirólisis (Py-CSIA). Aplicaciones.** José A González-Pérez; Nicasio T Jiménez-Morillo - Instituto de Recursos Naturales y Agrobiología de Sevilla (IRNAS-CSIC).
15:30	**Ventajas del nuevo EA IsoLink** Ricardo Alvarez – Thermo Fisher Scientific
16:00	**Café y demostración en vivo del Thermo Scientific™Delta Ray™ Connect.**
17:00	**Conclusiones.**

Ricardo Álvarez (Sales Representative)
e-mail: ricardo.alvarez@thermofisher.com.
Móvil: 663 160 000

ThermoFisher SCIENTIFIC





Laboratorio de Isótopos Estables de la Estación Biológica de Doñana

LIE-EBD, CSIC

[Web Page]

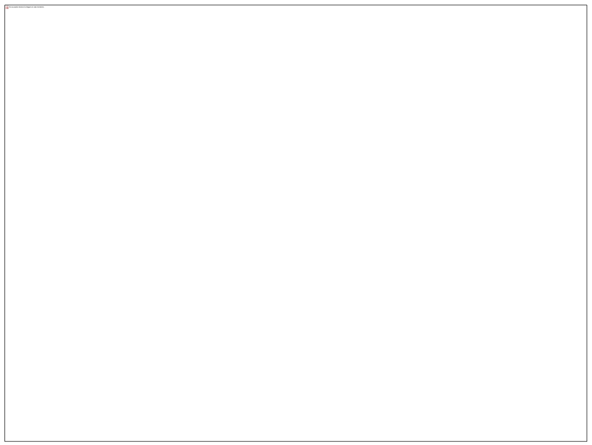
Foto personal del LIE, decir que hemos hecho dos cursos (foto), y que estamos Abiertos a asesorar y participar en cualqueir proyecto

Stable Isotope Laboratory & Scientific Collections

+

✓ High potential to study long-term changes on trophic ecology and migratory connnectivity

Incorporation of SIA to long-term field monitoring programs



CENTRALIZED REPOSITORY FOR ISOTOPIC DATA

ISOBANK

Database Structure

Pauli et al. 2017, PNAS

Isotopes are magic,
Can’t see them,
Can’t touch them,
Can’t hear or smell them,
Yet they are all around and inside us,
Making chemical commotion.
They are nature’s dyestuffs,
A world of pastel isotope landscapes
Awaits those who venture untrodden paths,

Discovery this way

by Brian Fry