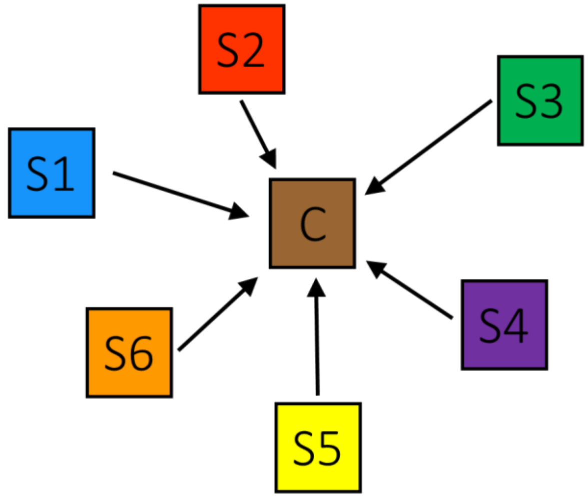
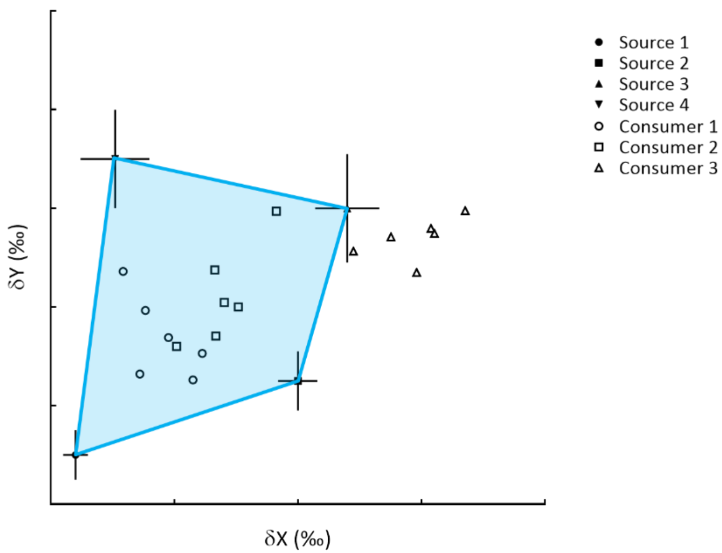
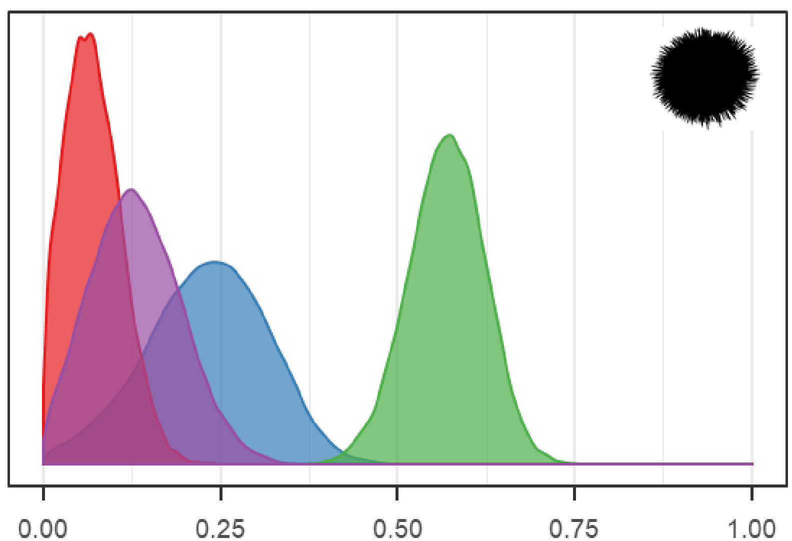
Stable isotope mixing models







Loïc MICHEL – loicnmichel@gmail.com
Course "Etude des isotopes stables et applications au milieu marin"

What do animals feed on?

What do animals feed on?



There is (nearly) an infinity of ecological questions somehow linked to animal diet

- Which are the resources essential for a consumer's nutrition?
- Do species A and B feed on the same resources?
- Does species A consumes a different amount of a given resource than species B?
- Is the diet of this animal stable in time, or does it shift to match seasonal resource availability?
- …

What do animals feed on?

There is (nearly) an infinity of ecological questions somehow linked to animal diet

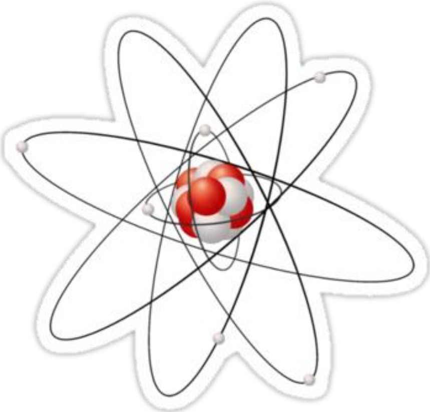
- Which are the resources essential for a consumer's nutrition?
- Do species A and B feed on the same resources?
- Does species A consumes a different amount of a given resource than species B?
- Is the diet of this animal stable in time, or does it shift to match seasonal resource availability?
- …

To answer such questions, we need tools that allow us to delineate animal diet, i.e. to quantify the contribution of each potential food item to the diet of a consumer

→ Stable isotope mixing models

Stable isotopes: you are what you eat

Mixing law: stable isotope composition of an animal is a proportional mix of its food sources' isotopic composition



Stable isotopes: you are what you eat

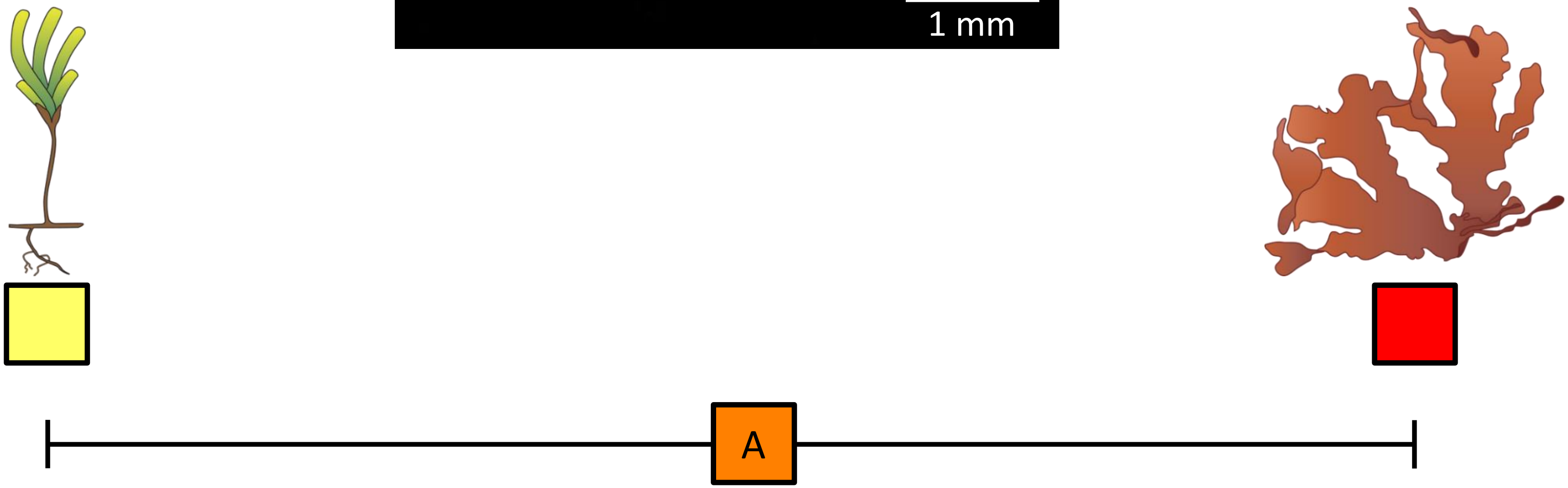
Mixing law: stable isotope composition of an animal is a proportional mix of its food sources' isotopic composition

Stable isotopes: you are what you eat

Mixing law: stable isotope composition of an animal is a proportional mix of its food sources’ isotopic composition

Stable isotopes: you are what you eat

Mixing law: stable isotope composition of an animal is a proportional mix of its food sources' isotopic composition



Stable isotopes: you are what you eat

Mixing law: stable isotope composition of an animal is a proportional mix of its food sources' isotopic composition

Stable isotopes: you are what you eat

Mixing law: stable isotope composition of an animal is a proportional mix of its food sources' isotopic composition

If you measure the isotopic compositions of an animal and its food item, you can calculate contributions of each food item to this animal's diet

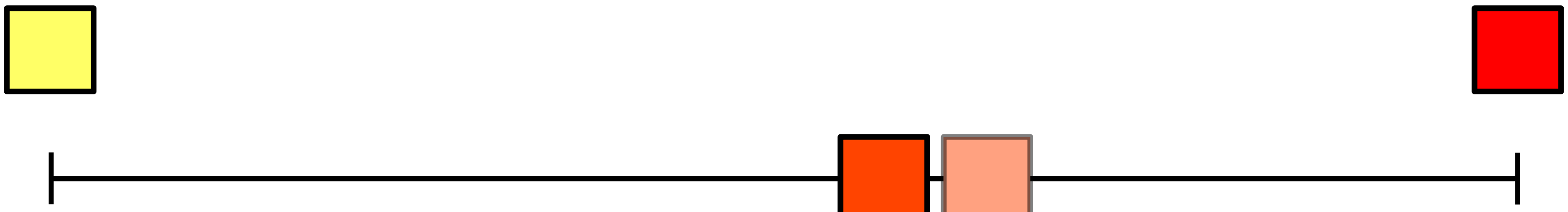
A simple mixing model

$$\begin{cases} \delta X_{Cons} = p1.\delta X_{S1} + p2.\delta X_{S2} \\ p1 + p2 = 1 \end{cases}$$

$$\Updownarrow$$

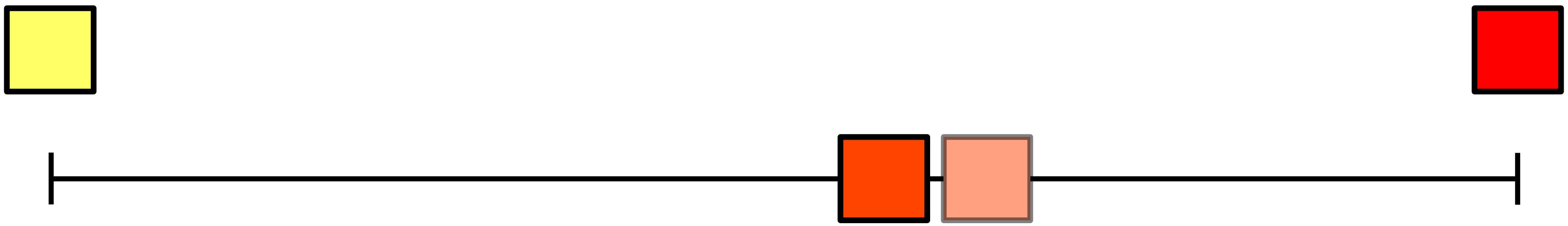
$$\begin{cases} p1 = \dfrac{\delta X_{Cons} - \delta X_{S2}}{\delta X_{S1} - \delta X_{S2}} \\ p2 = 1 - p1 \end{cases}$$

You are what you eat… plus a few ‰!



We need to take into account trophic fractionation (most cases: enrichment in heavy isotope, hence "trophic enrichment factor" or TEF

You are what you eat… plus a few ‰!



$$\begin{cases} \delta X_{Cons} - \Delta X = p1.\delta X_{S1} + p2.\delta X_{S2} \\ p1 + p2 = 1 \end{cases}$$

$$\Updownarrow$$

$$\begin{cases} p1 = \dfrac{\delta X_{Cons} - \Delta X - \delta X_{S2}}{\delta X_{S1} - \delta X_{S2}} \\ p2 = 1 - p1 \end{cases}$$

What if you have 3 sources?

Just add a second isotopic ratio!

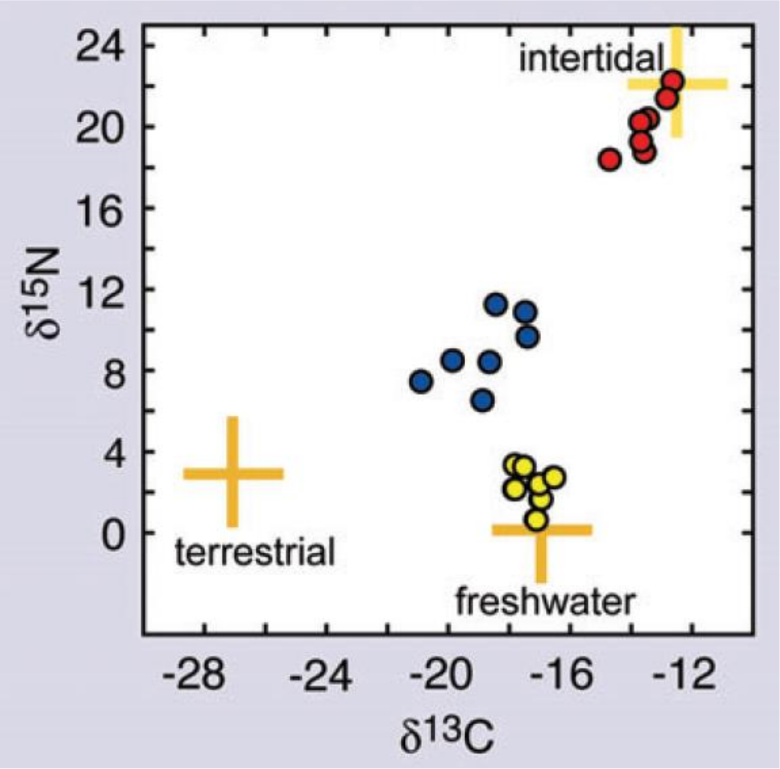
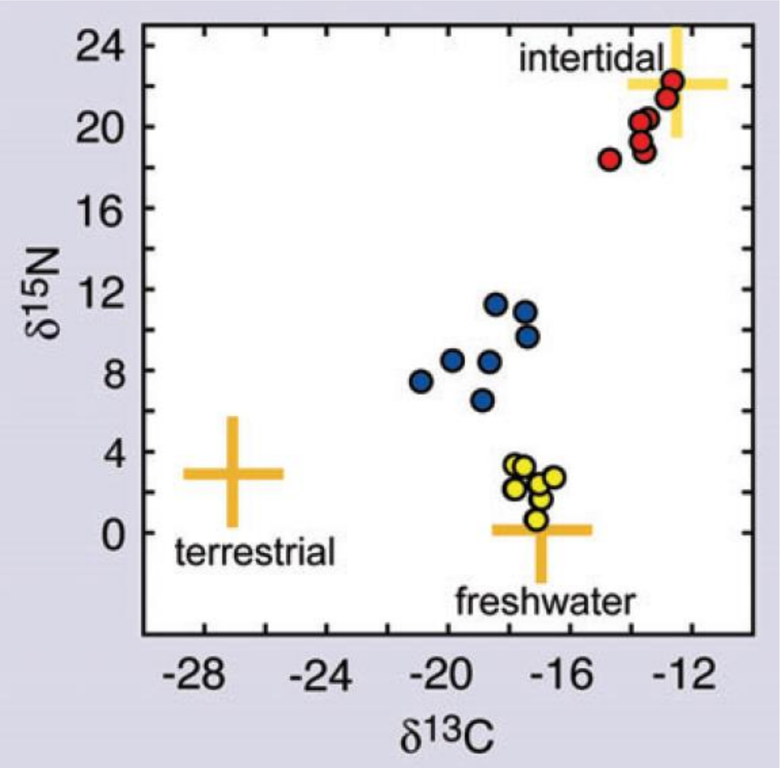


Figure: Newsome et al. 2007 Front Ecol Environ 5: 429-436

What if you have 3 sources?

Just add a second isotopic ratio!

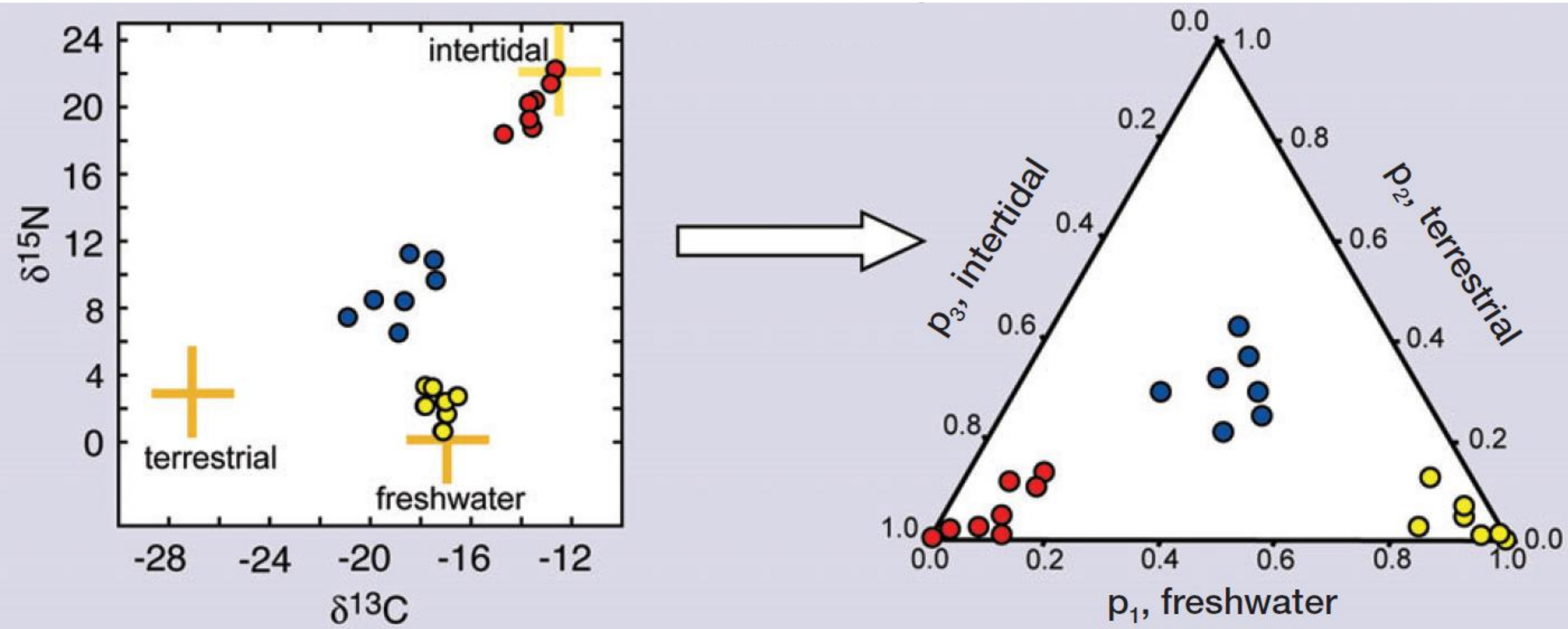


$$\begin{cases} \delta X_{Cons} - \Delta X = p1.\delta X_{S1} + p2.\delta X_{S2} + p3.\delta X_{S3} \\ \delta Y_{Cons} - \Delta Y = p1.\delta Y_{S1} + p2.\delta Y_{S2} + p3.\delta Y_{S3} \\ p1 + p2 + p3 = 1 \end{cases}$$

Figure: Newsome et al. 2007 Front Ecol Environ 5: 429-436

What if you have 3 sources?

Just add a second isotopic ratio!

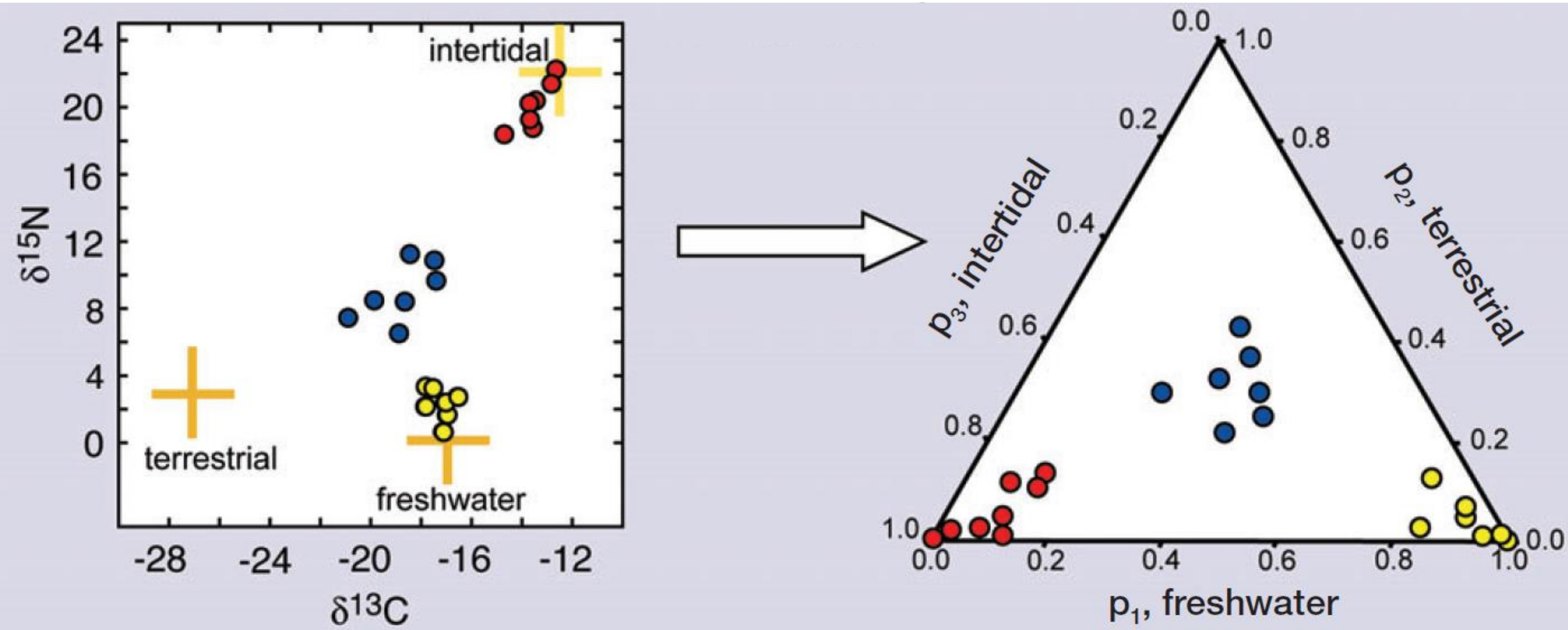


$$\begin{cases} \delta X_{Cons} - \Delta X = p1.\delta X_{S1} + p2.\delta X_{S2} + p3.\delta X_{S3} \\ \delta Y_{Cons} - \Delta Y = p1.\delta Y_{S1} + p2.\delta Y_{S2} + p3.\delta Y_{S3} \\ p1 + p2 + p3 = 1 \end{cases}$$

Figure: Newsome et al. 2007 Front Ecol Environ 5: 429-436

What if you have 3 sources?

Just add a second isotopic ratio!



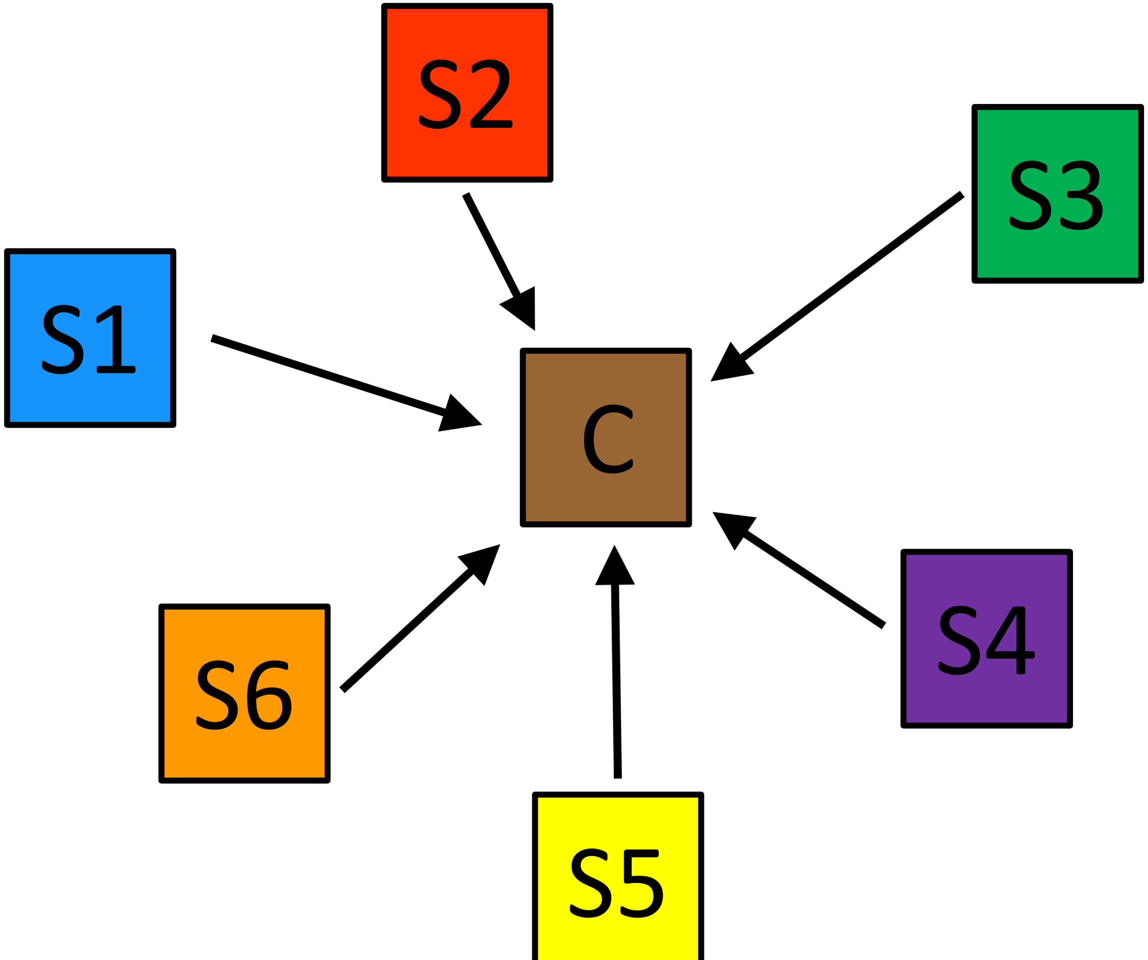
If you have n isotopic ratios, you can deal with n+1 sources

Figure: Newsome et al. 2007 Front Ecol Environ 5: 429-436

Real-world food webs

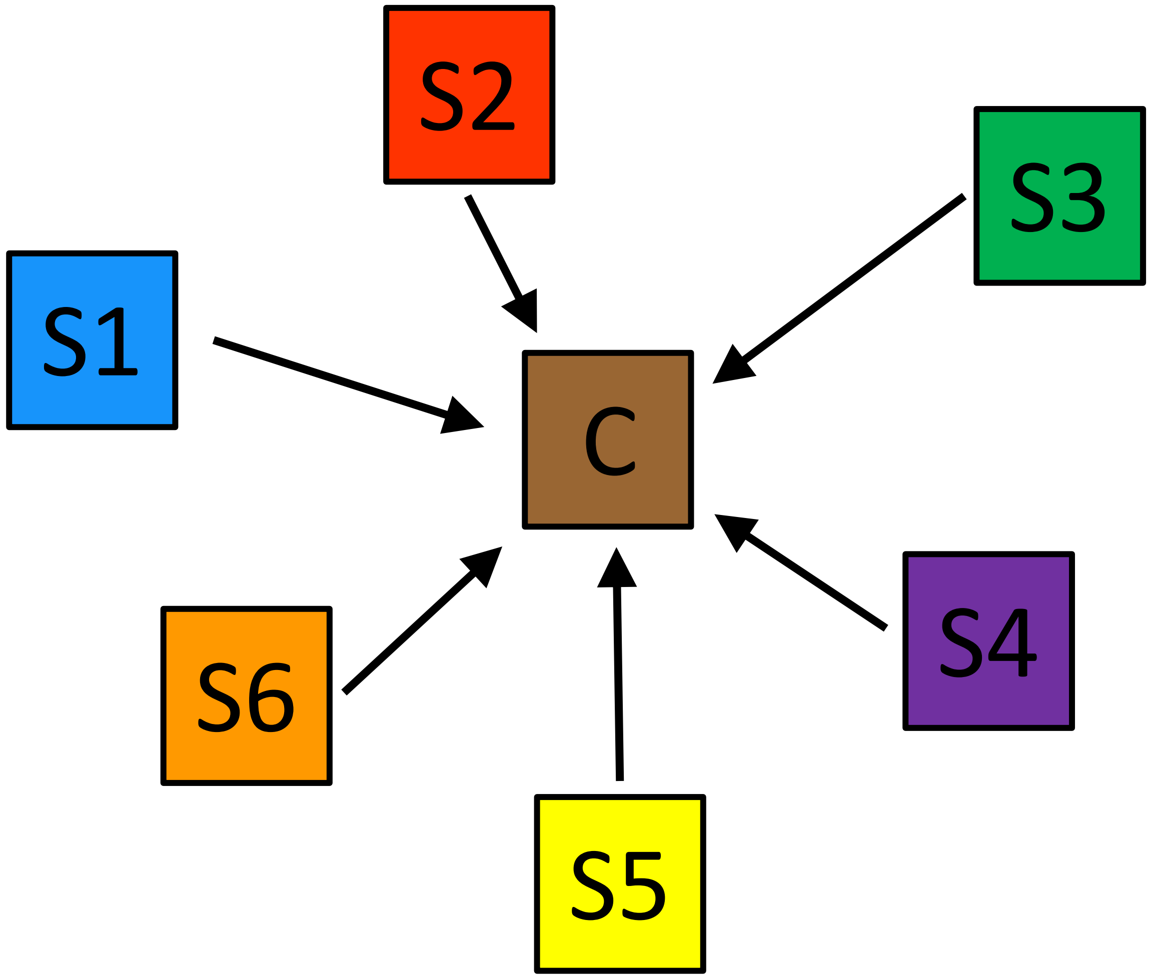
Real-world food webs are complex: animals feed on many food items…

Most isotopic studies: 2 isotopic ratios (C & N), sometimes 3



Real-world food webs

Many systems are underdetermined: more unknowns then equations
Need of more complex mathematical models



Dealing with underdetermined systems

Oecologia (2003) 136:261–269
DOI 10.1007/s00442-003-1218-3

ECOSYSTEMS ECOLOGY

Donald L. Phillips · Jillian W. Gregg

Source partitioning using stable isotopes: coping with too many sources

IsoSource model

Iterative procedure:

- All possible combinations of each source combination (0-100%) are examined in small increments (e.g. 1%).
- Combinations that sum to the consumer’s isotopic composition are considered feasible
- The program returns the ranges and frequencies of these solutions

There is no single solution! The model’s "solution" is the full distribution of feasible solutions

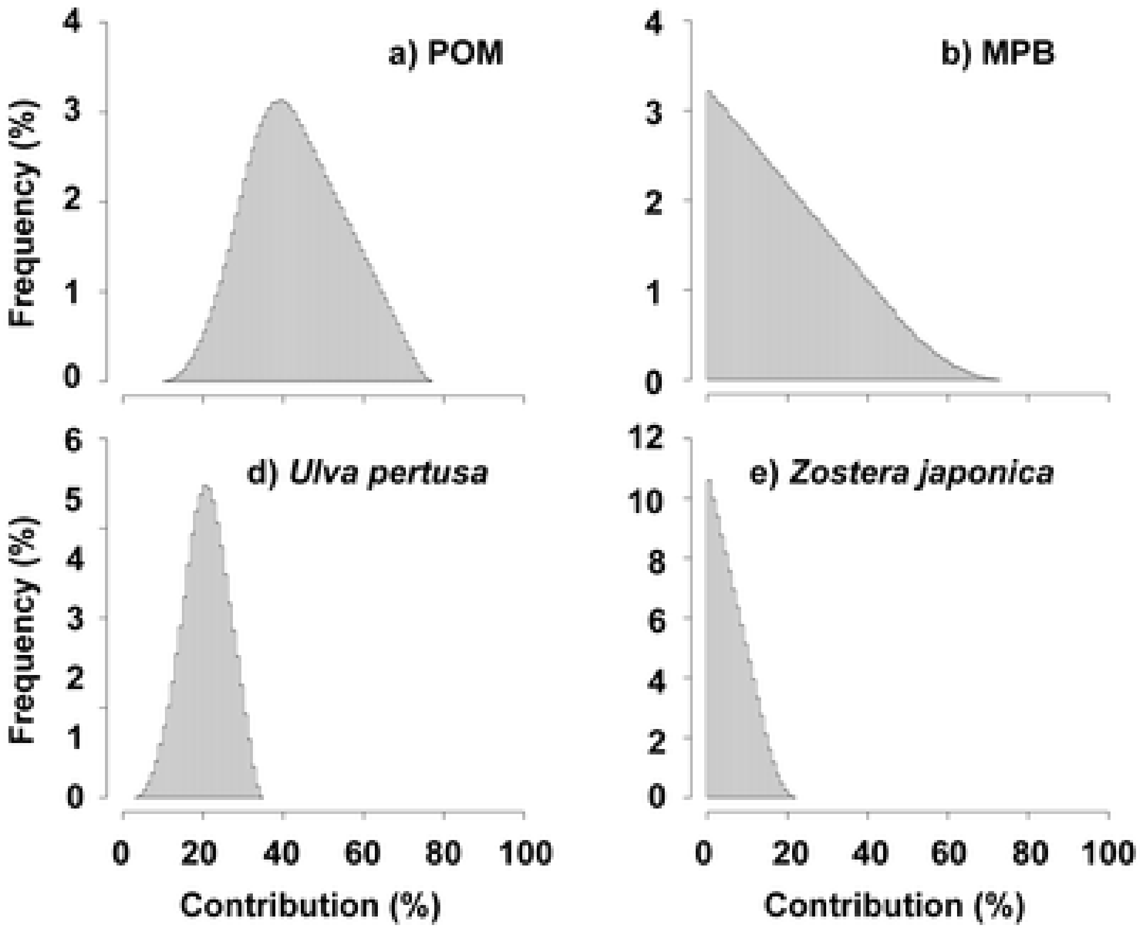
Dealing with underdetermined systems

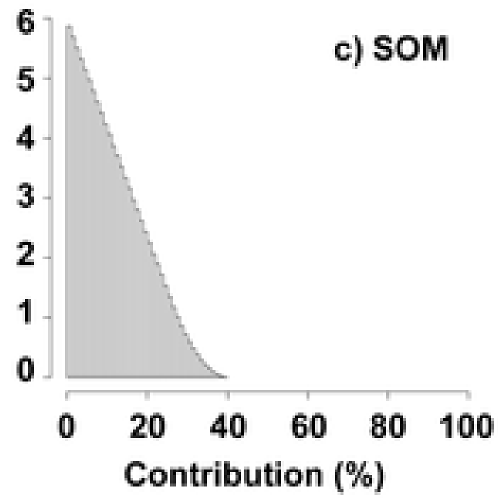
Oecologia (2003) 136:261–269
DOI 10.1007/s00442-003-1218-3

ECOSYSTEMS ECOLOGY

Donald L. Phillips · Jillian W. Gregg

Source partitioning using stable isotopes: coping with too many sources



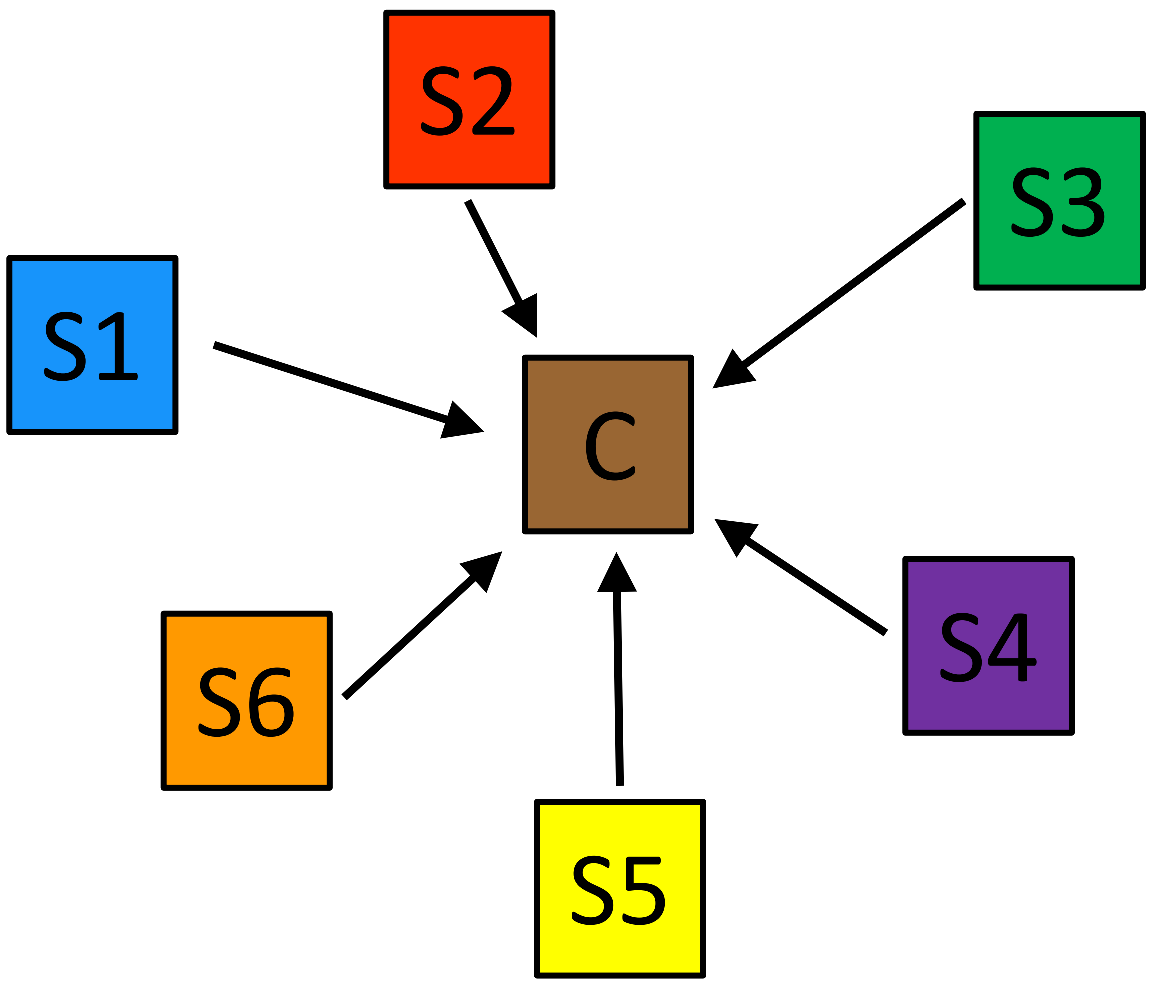


Example of IsoSource output: diet of the clam *Ruditapes philippinarum*

Komorita et al. 2014 PLoS ONE 9(1): e86732

Real-world food webs

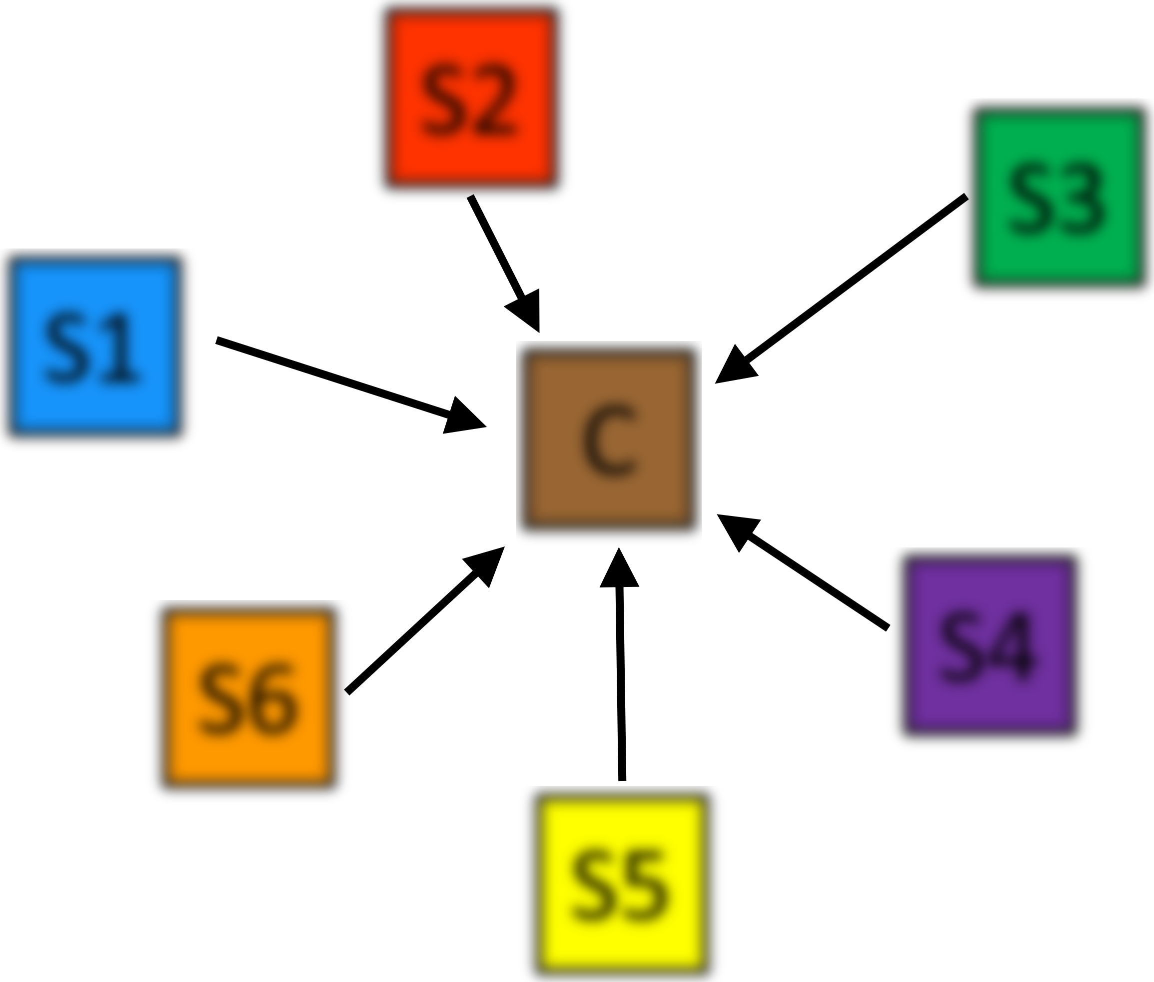
Isotopic compositions of consumers and food items are uncertain



Real-world food webs

Isotopic compositions of consumers and food items are uncertain

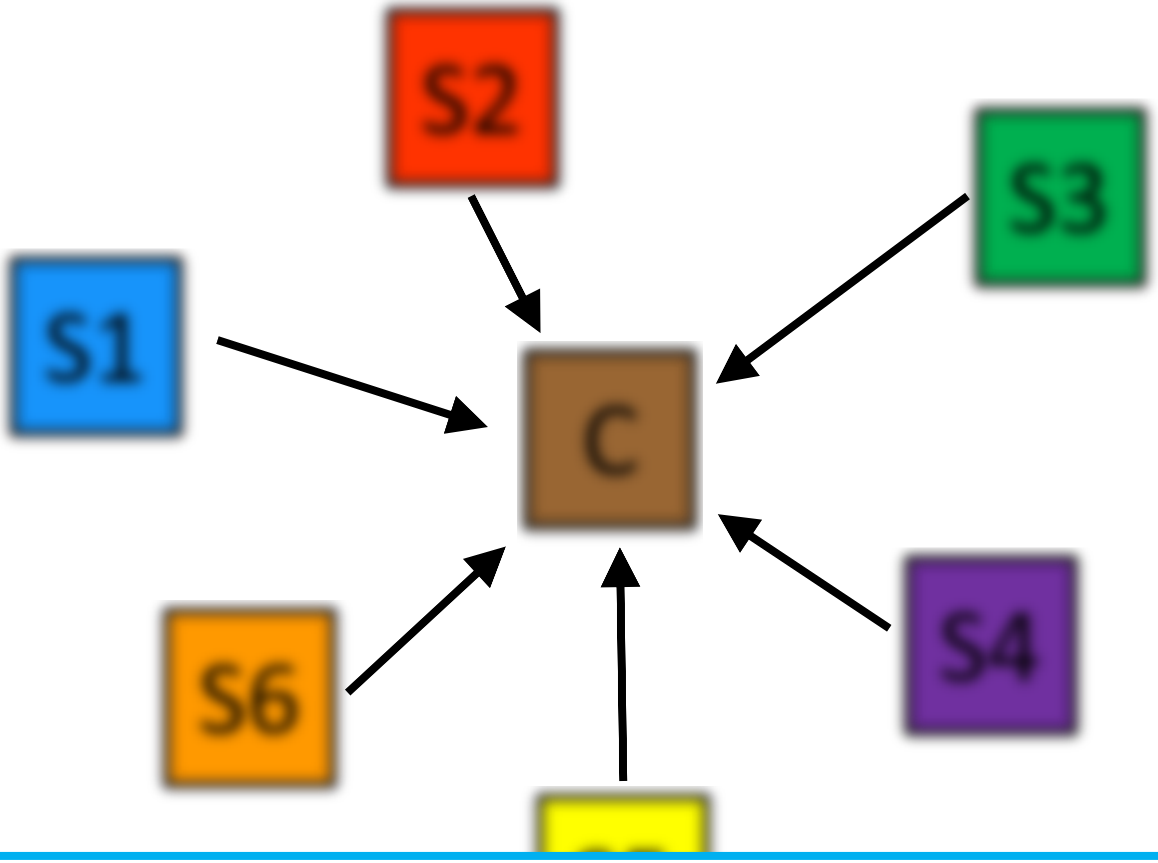
2 main sources of uncertainty: natural variability (holds ecological info - we want to keep it!) and analytical error (we aim to minimise it, but we have to deal with it anyway)



Real-world food webs

Isotopic compositions of consumers and food items are uncertain

2 main sources of uncertainty: natural variability (holds ecological info - we want to keep it!) and analytical error (we aim to minimise it, but we have to deal with it anyway)



To build more realistic mixing models, we need to take uncertainty into account!

A new family of mixing models


Ecology Letters, (2008) **11**: 470–480 doi: 10.1111/j.1461-0248.2008.01163.x

LETTER

Jonathan W. Moore[1,2*,†] and Brice X. Semmens[1,†]

Incorporating uncertainty and prior information into stable isotope mixing models

MixSIR (https://conserver.iugo-cafe.org/user/brice.semmens/MixSIR)


March 2010 | Volume 5 | Issue 3 | e9672 PLoS one

Source Partitioning Using Stable Isotopes: Coping with Too Much Variation

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

SIAR (https://github.com/AndrewLJackson/siar)

Models based on Bayesian inference

Research Article Environmetrics

Bayesian stable isotope mixing models

Andrew C. Parnell[a*], Donald L. Phillips[b], Stuart Bearhop[c], Brice X. Semmens[d], Eric J. Ward[e], Jonathan W. Moore[f], Andrew L. Jackson[g], Jonathan Grey[h], David J. Kelly[g] and Richard Inger[i]

Environmetrics 2013; **24**: 387–399

Bayesian inference

Method of estimating the probability of an event based on prior knowledge of conditions related to this event.

What’s the probability of Standard de Liège winning the Belgian championship this year?

Bayesian inference

Method of estimating the probability of an event based on prior knowledge of conditions related to this event.

What's the probability of Standard de Liège winning the Belgian championship this year?

What's the probability of Standard de Liège winning the Belgian championship this year, knowing that they won 10 times in 119 championships?

Bayesian inference

Method of estimating the probability of an event based on prior knowledge of conditions related to this event.

What’s the probability of Standard de Liège winning the Belgian championship this year?

What’s the probability of Standard de Liège winning the Belgian championship this year, knowing that they won 10 times in 119 championships?

What’s the probability of Standard de Liège winning the Belgian championship this year, knowing that they won 10 times in 119 championships and currently rank 13th out of 18?

Bayesian inference

Method of estimating the probability of an event based on prior knowledge of conditions related to this event.

$$P(A|B) = \frac{P(B|A).P(A)}{P(B)}$$

P(A|B) : Likelihood of event A occurring given that event B occurred
P(B|A) : Likelihood of event B occurring given that event A occurred
P(A) : Probability that event A happens independently of B
P(B) : Probability that event B happens independently of A

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

Intuitive answer: 99%

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

$$P(A|B) = \frac{P(B|A).P(A)}{P(B)}$$

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

$$P(User|+) = \frac{P(+|User).P(User)}{P(+)}$$

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

$$P(\text{User}|+) = \frac{P(+|\text{User}).P(\text{User})}{P(+)}$$

$$P(+) = P(+|\text{User}).P(\text{User}) + P(+|\text{Non-user}).P(\text{Non-user})$$

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

$$P(\text{User}|+) = \frac{P(+|\text{User}).P(\text{User})}{P(+|\text{User}).P(\text{User}) + P(+|\text{Non-user}).P(\text{Non-user})}$$

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

$$P(\text{User}|+) = \frac{P(+|\text{User}).P(\text{User})}{P(+|\text{User}).P(\text{User}) + P(+|\text{Non-user}).P(\text{Non-user})}$$

$$P(\text{User}|+) = \frac{0.99 \,.\, 0.01}{0.99 \,.\, 0.01 + 0.01.\, 0.99}$$

Bayesian inference

You take a drug test.
This test has 99% sensitivity (i.e. 99% of drug users test positive).
This test has 99% reliability (i.e. 99% of non-drug users test negative).
The tested drug is used by 1% of the population.

Your test comes up positive. What's the probability that you use the drug?

$$P(User|+) = \frac{P(+|User).P(User)}{P(+|User).P(User) + P(+|Non\text{-}user).P(Non\text{-}user)}$$

$$P(User|+) = \frac{0.99 \,.\, 0.01}{0.99 \,.\, 0.01 + 0.01 .\, 0.99}$$

$$P(User|+) = 0.5$$

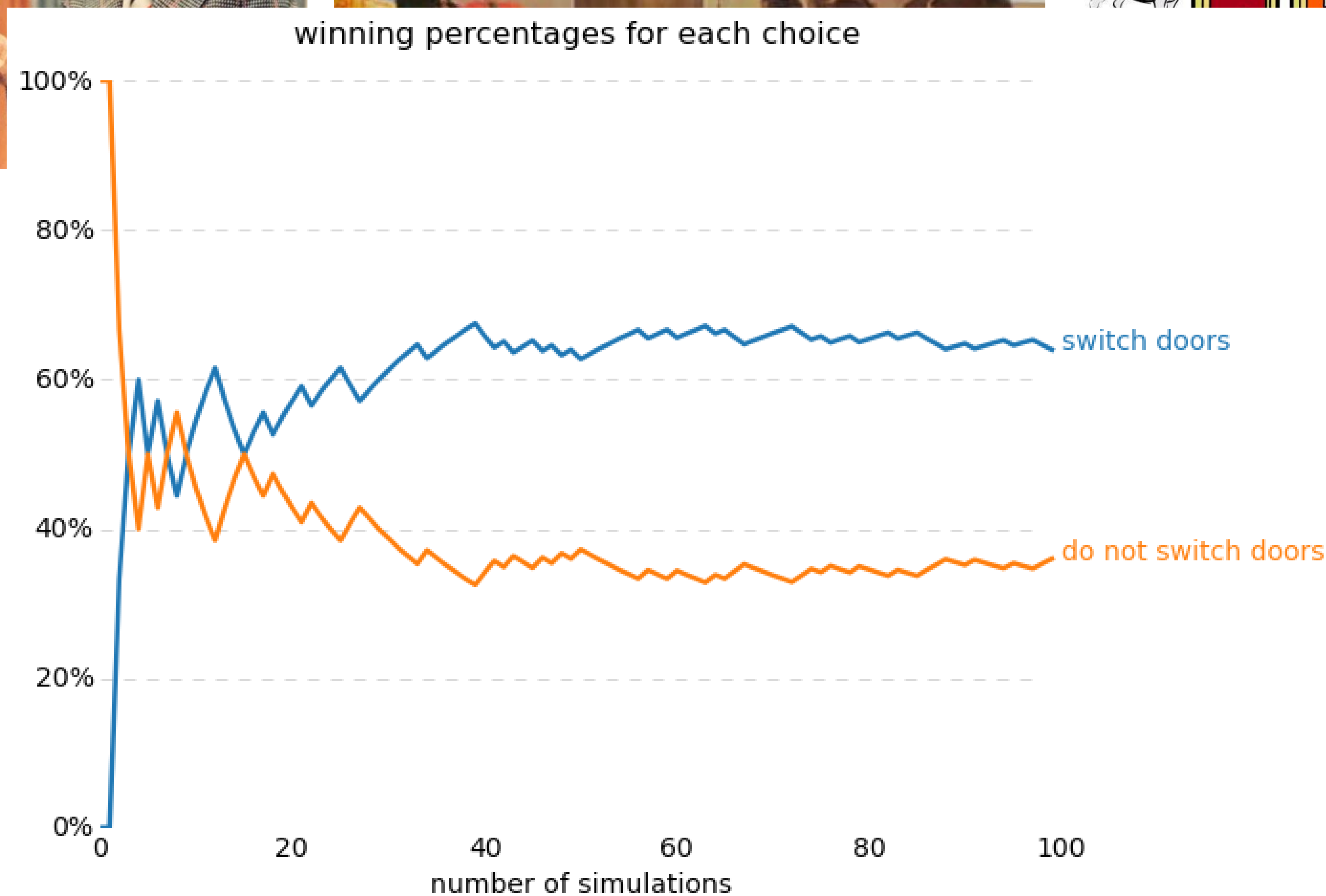
The Monty Hall problem

You have three doors to choose from. Behind one, there is a car. Behind the others, there is a goat.

After you picked one, Monty Hall opens one of the two remaining doors, and shows you that it leads to a goat.

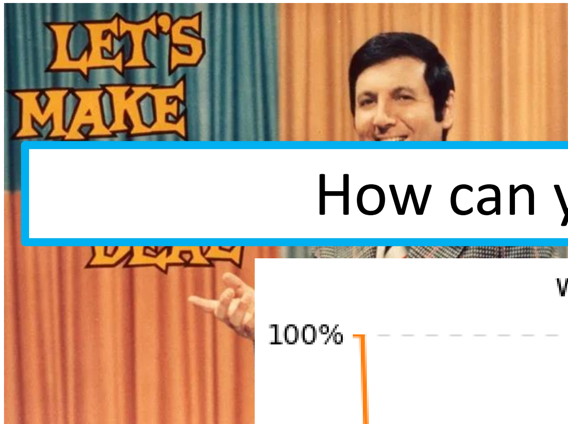
You have two doors remaining: the one you initially picked, and another one. Should you stick to your door or switch?

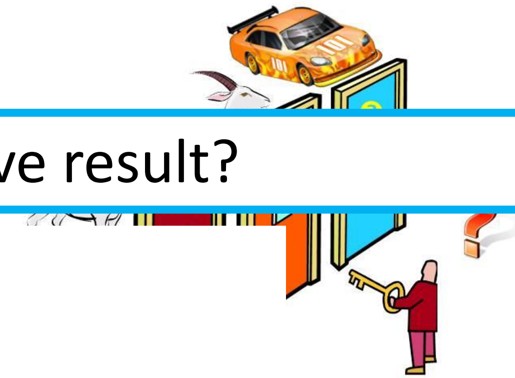
The Monty Hall problem



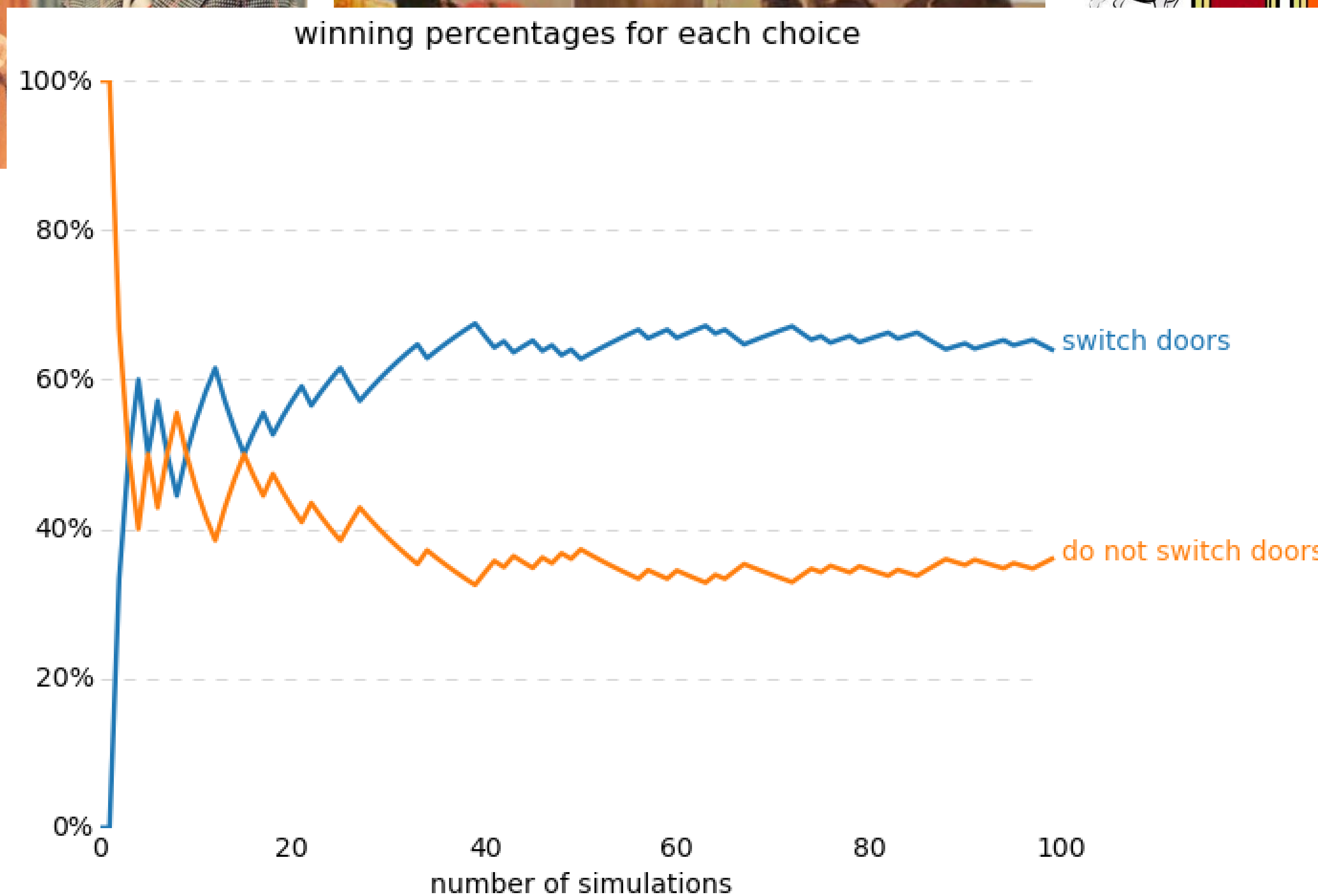
Source: https://medium.com/@NickDoesData/applying-bayes-theorem-simulating-the-monty-hall-problem-with-python-5054976d1fb5

The Monty Hall problem



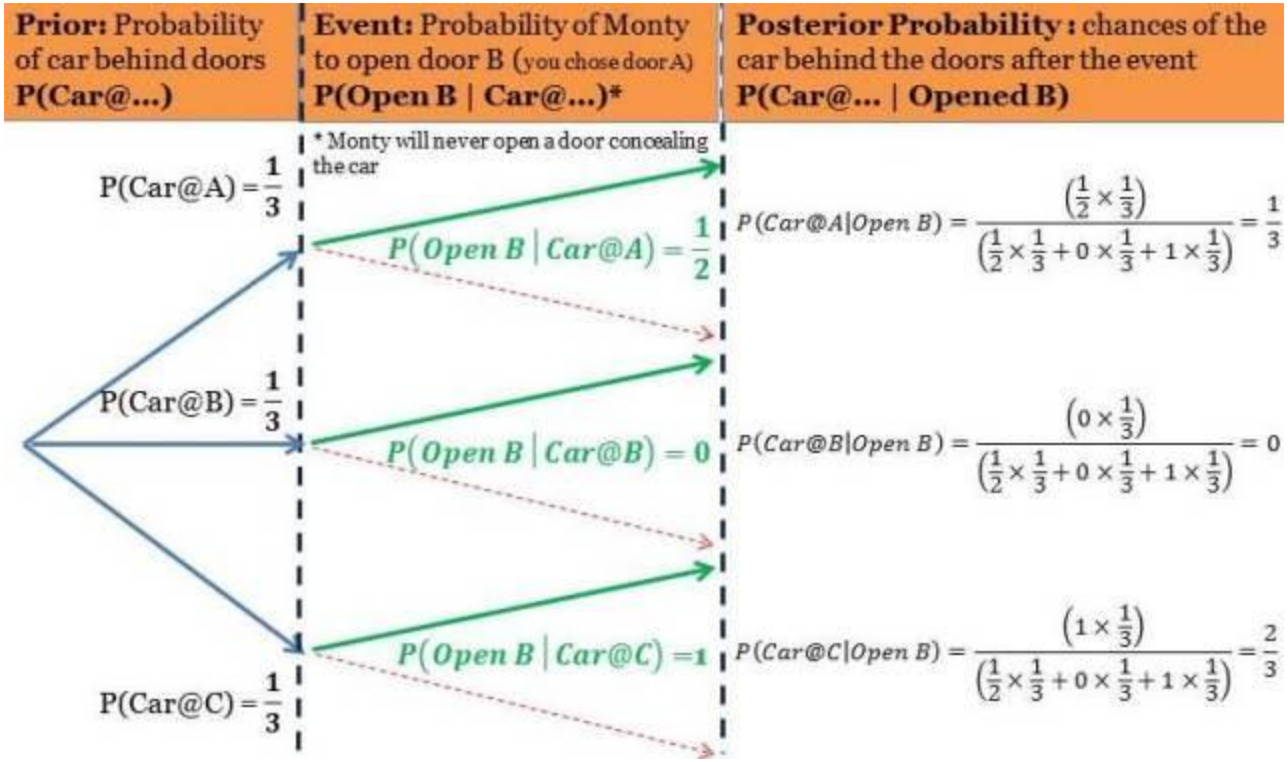


How can you explain this counter-intuitive result?



Source: https://medium.com/@NickDoesData/applying-bayes-theorem-simulating-the-monty-hall-problem-with-python-5054976d1fb5

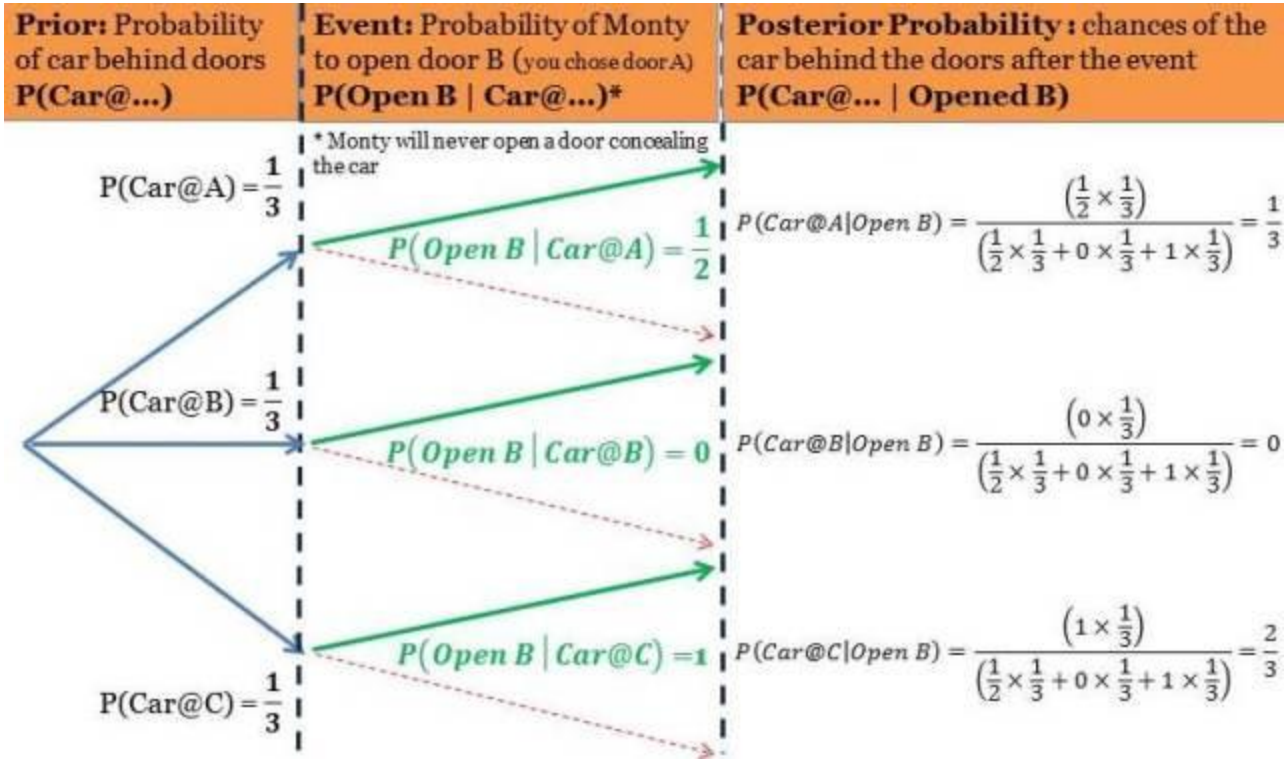
The Monty Hall problem



Source: http://ucanalytics.com/blogs/bayes-theorem-monty-hall-problem/. Assuming you initially picked door A, and Monty opened door B.

The Monty Hall problem

To maximise your chances to win, you should always switch doors

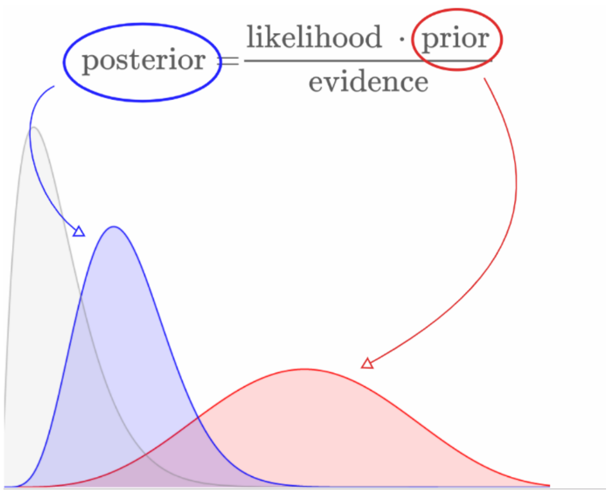


Source: http://ucanalytics.com/blogs/bayes-theorem-monty-hall-problem/. Assuming you initially picked door A, and Monty opened door B.

Bayesian mixing models: why?

1) Bayesian methods allow incorporation of prior information

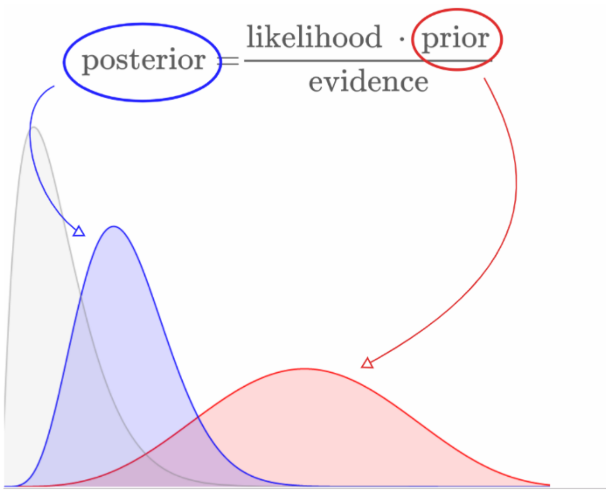
    If you have any info about you consumer's diet (gut contents, functional traits), you can include it as a prior.



Bayesian mixing models: why?

1) Bayesian methods allow incorporation of prior information
   If you have any info about you consumer's diet (gut contents, functional traits), you can include it as a prior.

2) Bayesian methods can integrate uncertainty from various sources
   Variability in sources and consumers isotopic ratios, but also in TEFs, can be taken into account in your model



Bayesian mixing models: why?

1) Bayesian methods allow incorporation of prior information

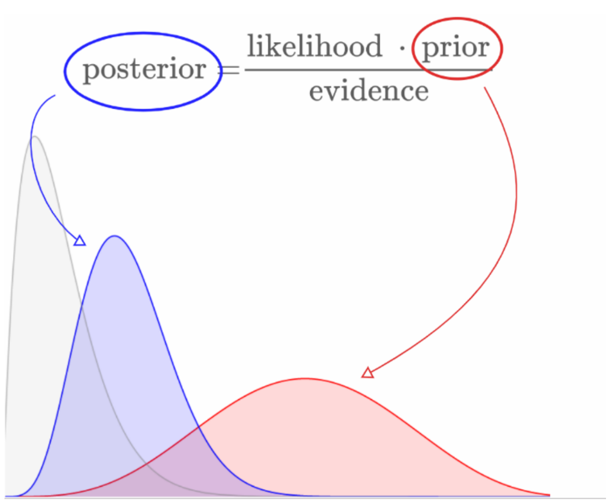
    If you have any info about you consumer's diet (gut contents, functional traits), you can include it as a prior.

2) Bayesian methods can integrate uncertainty from various sources

    Variability in sources and consumers isotopic ratios, but also in TEFs, can be taken into account in your model

3) Bayesian methods explicitly compare the strength of support for competing models or parameter values

    It is straightforward to compare model solutions (posterior probability distributions), as well as to estimate model performance (using diagnostics)



Bayesian mixing model: an example

Mascart et al. 2018 Food webs 16: e00086

b: *Ectinosoma dentatum*
d: *Clausocalanus arcuicornis*

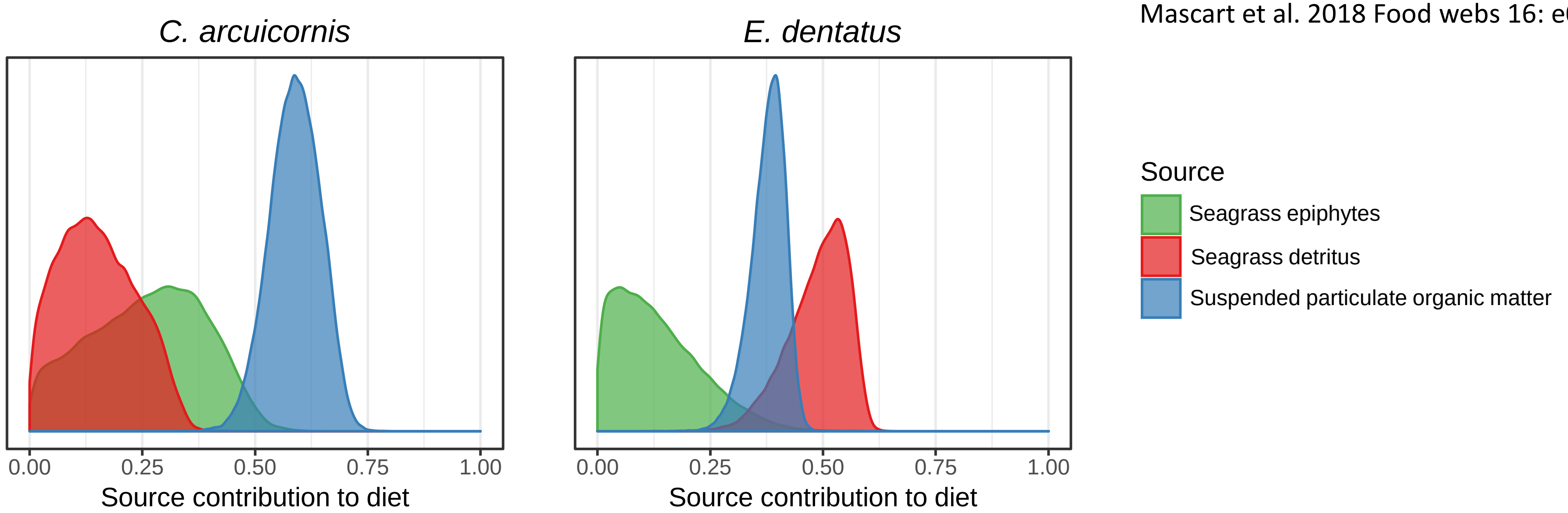
Context: These two species of copepods live together in *Posidonia oceanica* litter.

Question: Do they rely on the same resources?

Hypothesis: Differences in their morphology could be linked with different feeding behaviour, and therefore resource partitioning.

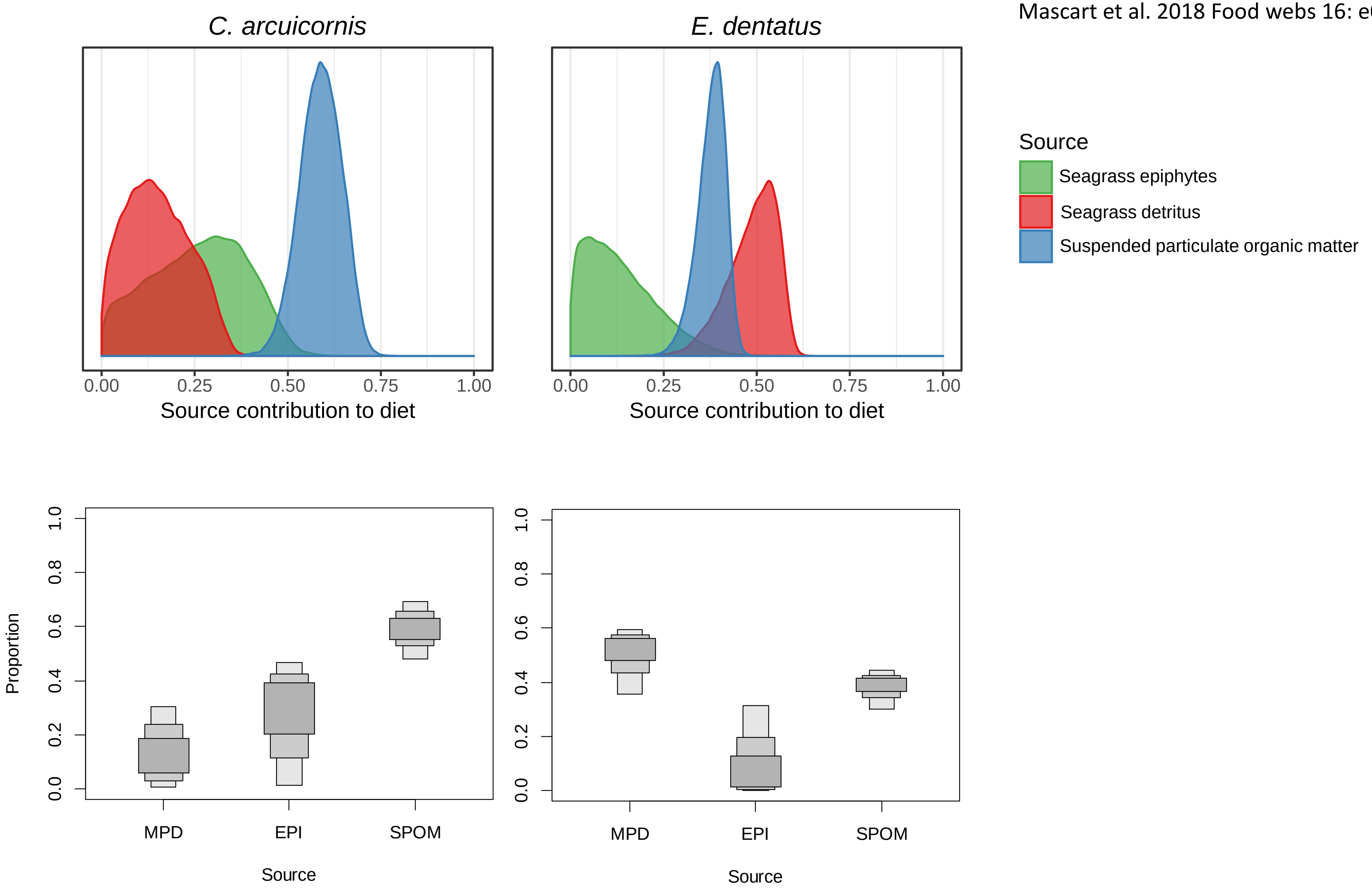
Bayesian mixing model: an example

Mascart et al. 2018 Food webs 16: e00086



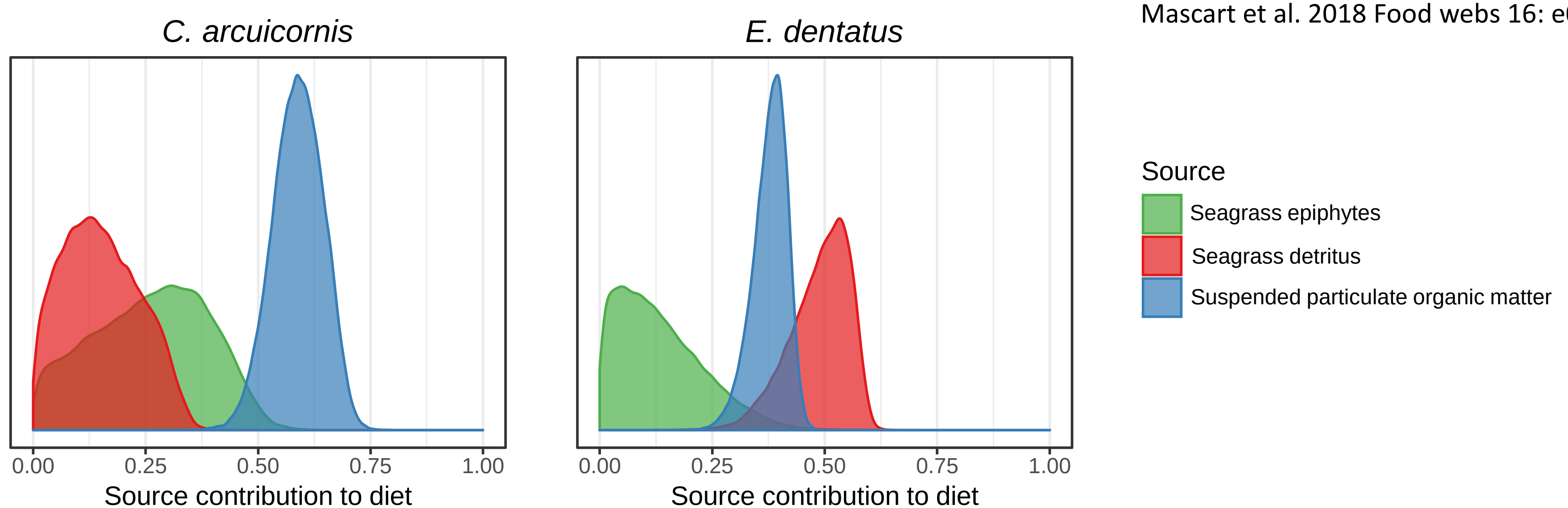
Bayesian mixing model: an example

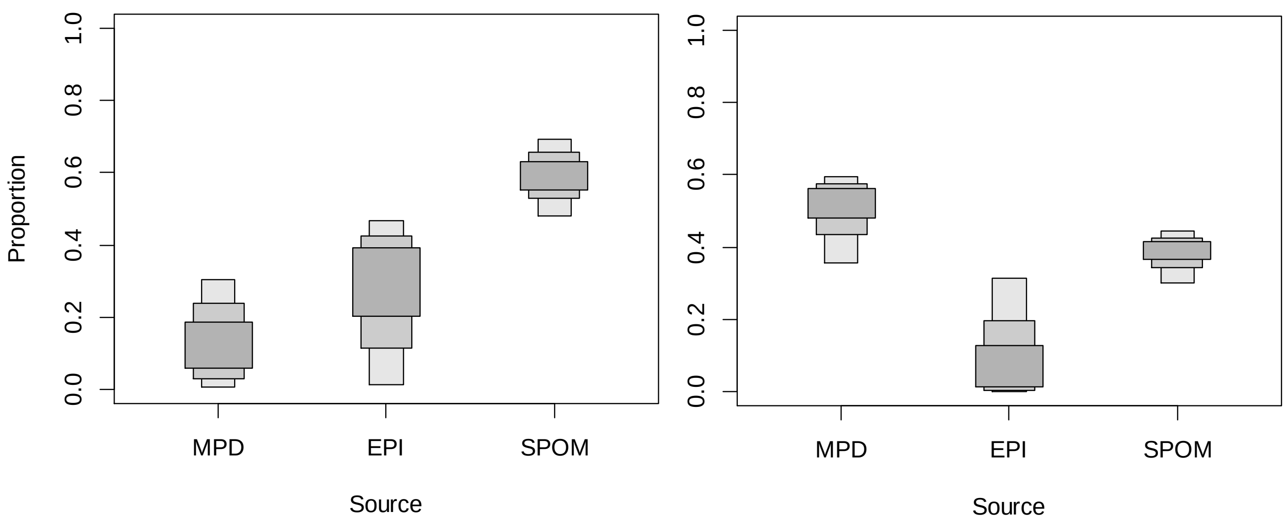
Mascart et al. 2018 Food webs 16: e00086



Bayesian mixing model: an example

Mascart et al. 2018 Food webs 16: e00086





How probable is it that contribution of a given source is different in the two species?

Epiphytes: 78.16%
Detritus: 99.86%
SPOM: 99.99%

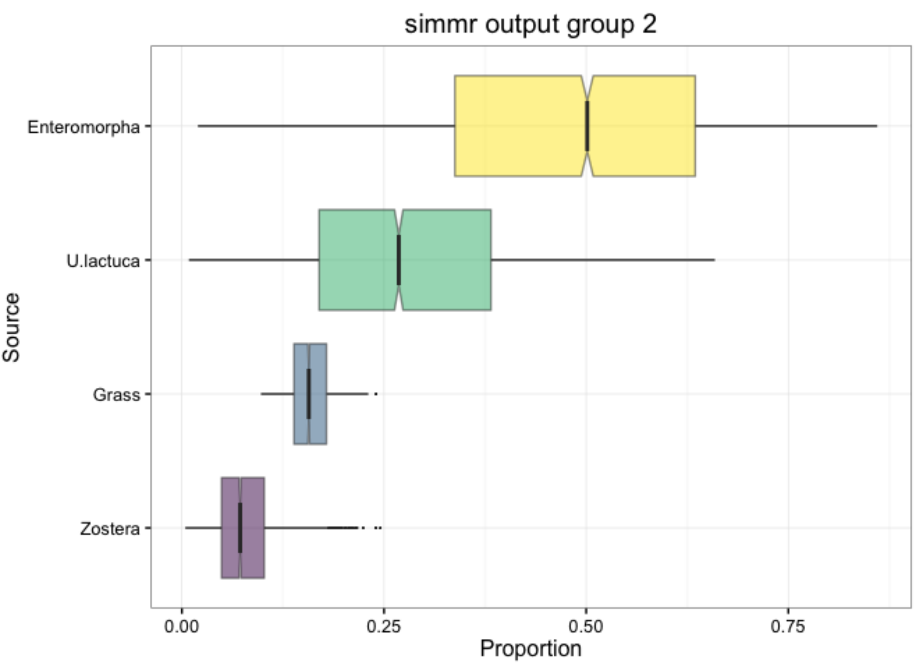
A simple Bayesian mixing model: simmr

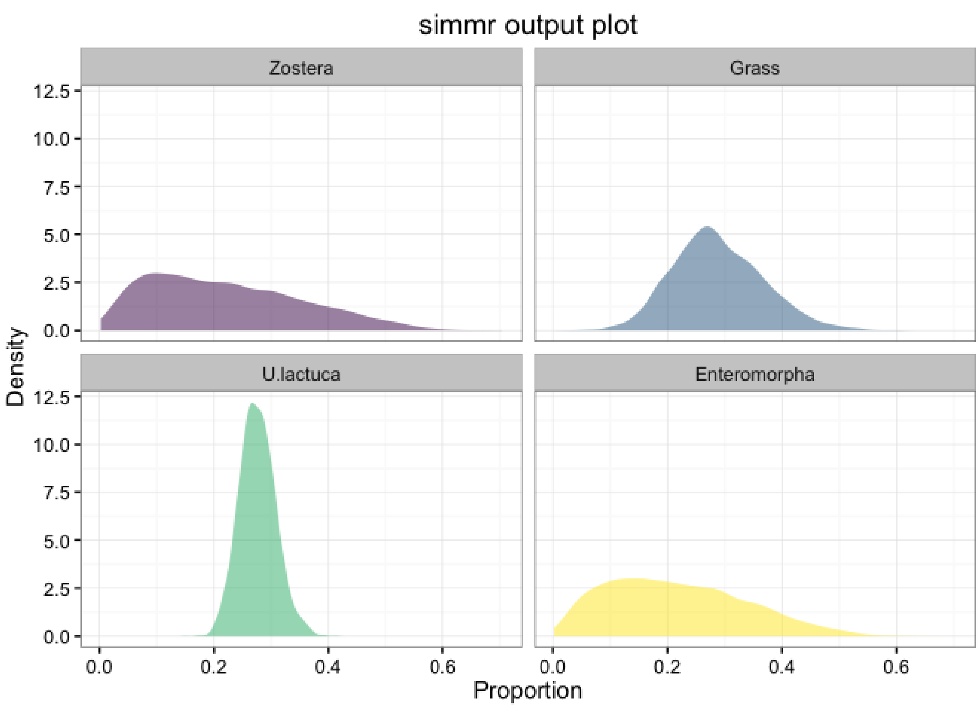
Stable Isotope Mixing Models in R with simmr

Andrew Parnell and Richard Inger

https://github.com/andrewcparnell/simmr

Upgrade of SIAR: many common features, plus a few improvements





A complex Bayesian mixing model: MixSIAR

Analyzing mixing systems using a new generation of Bayesian tracer mixing models

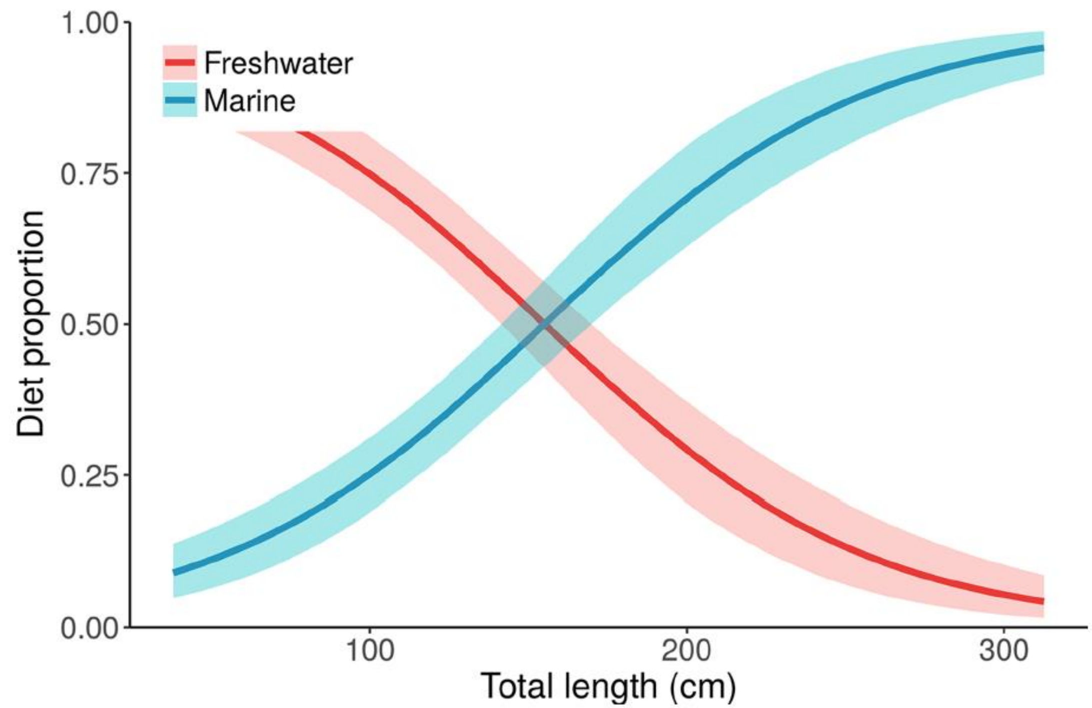
Brian C. Stock[1], Andrew L. Jackson[2], Eric J. Ward[3], Andrew C. Parnell[4], Donald L. Phillips[5] and Brice X. Semmens[1]

PeerJ

Stock et al. (2018), *PeerJ*, DOI 10.7717/peerj.5096

MixSIAR capabilities:

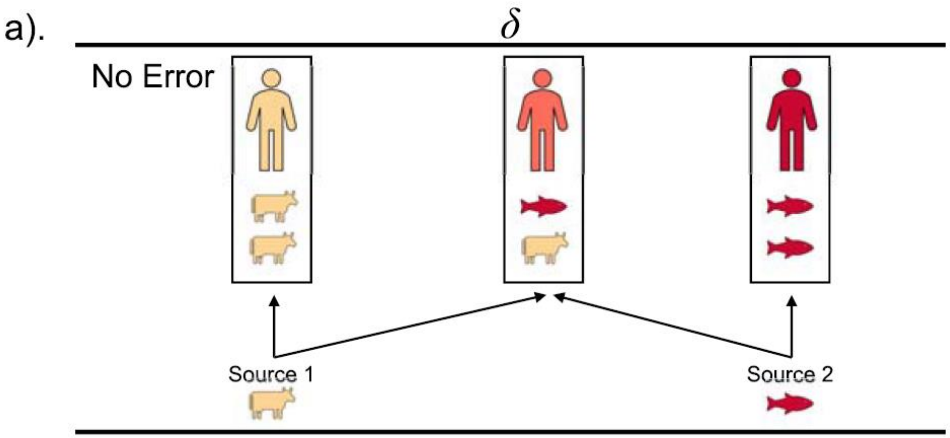
- Any number of tracers
- Categorical or continuous covariates
- Multiple error structures



Ontogenic shift in resource use in *Alligator mississippiensis*

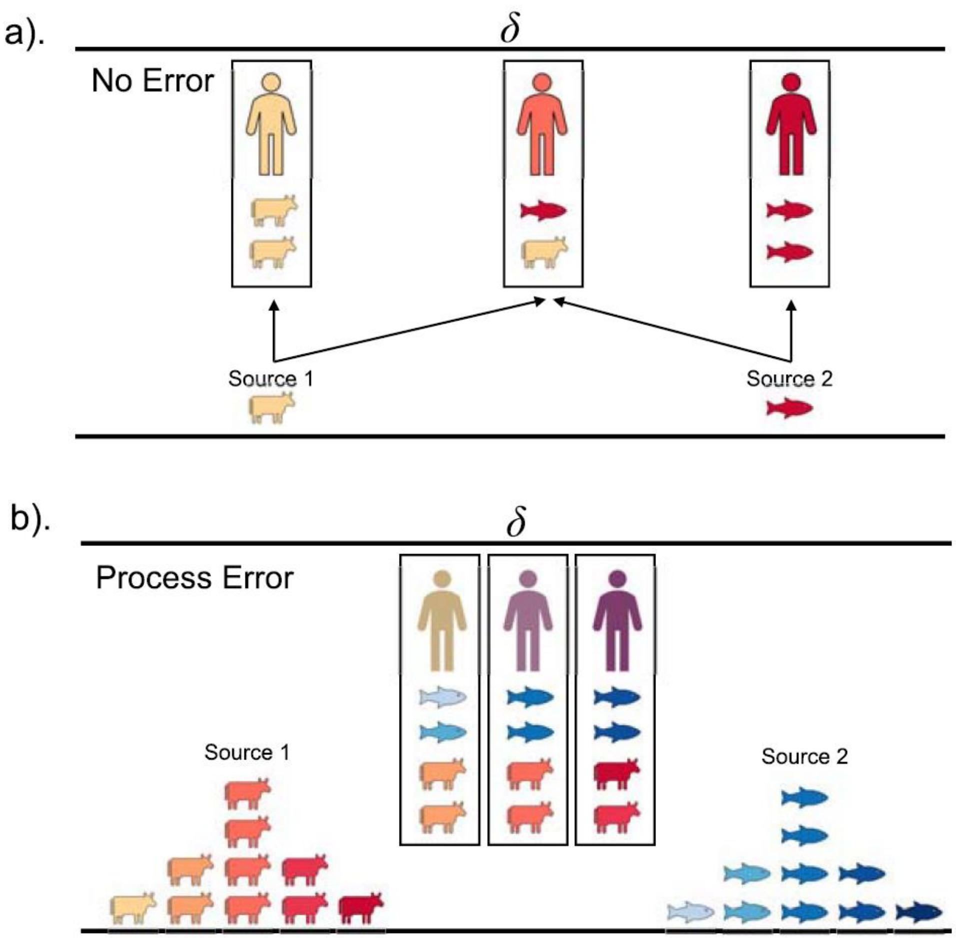
MixSIAR error structure

Cheung & Szpak 2020 J. Archaeo Method Theory - https://doi.org/10.1007/s10816-020-09492-5



MixSIAR error structure

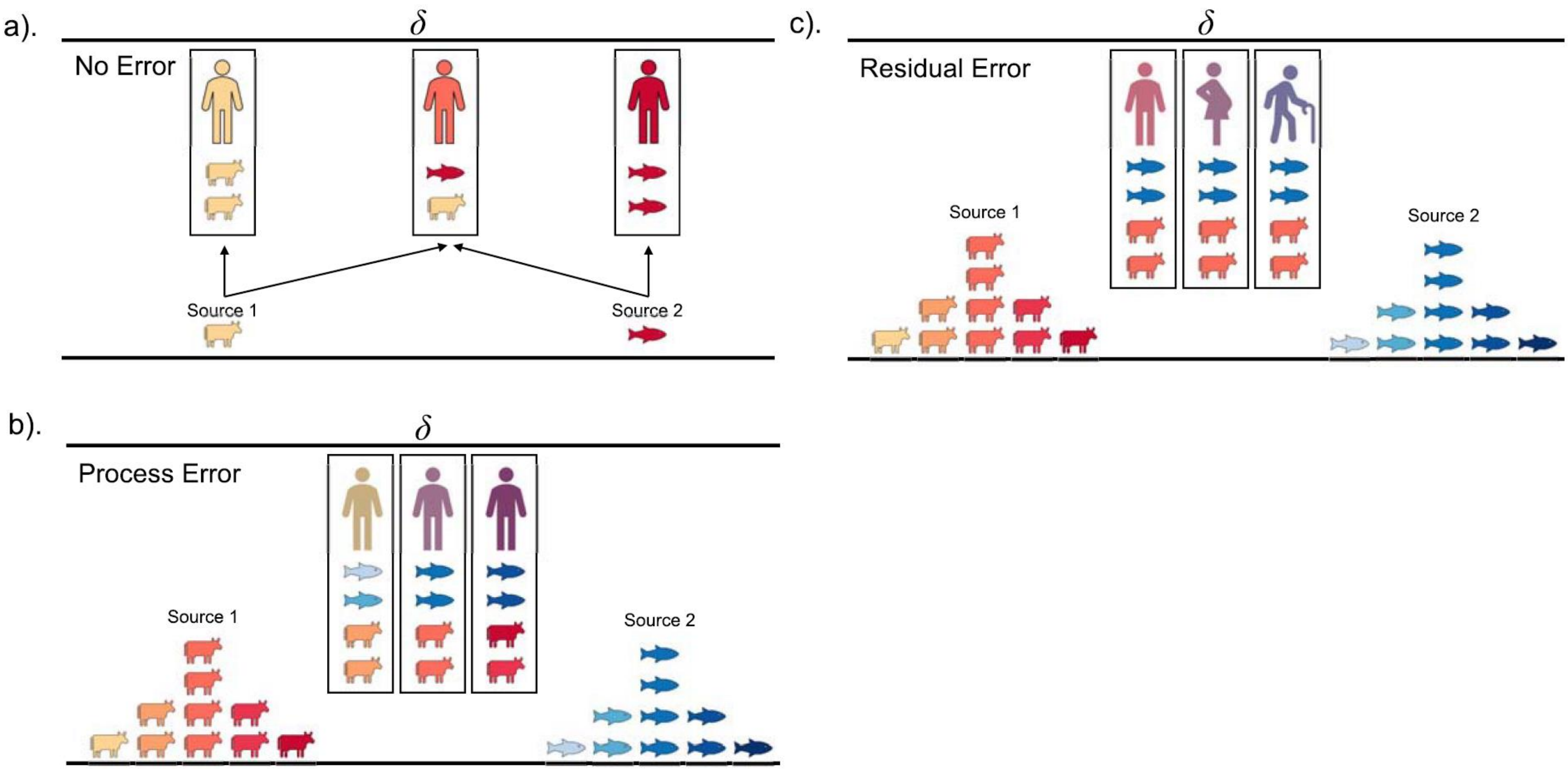
Cheung & Szpak 2020 J. Archaeo Method Theory - https://doi.org/10.1007/s10816-020-09492-5



Process error: sources are isotopically variable, and consumers subsample.

MixSIAR error structure

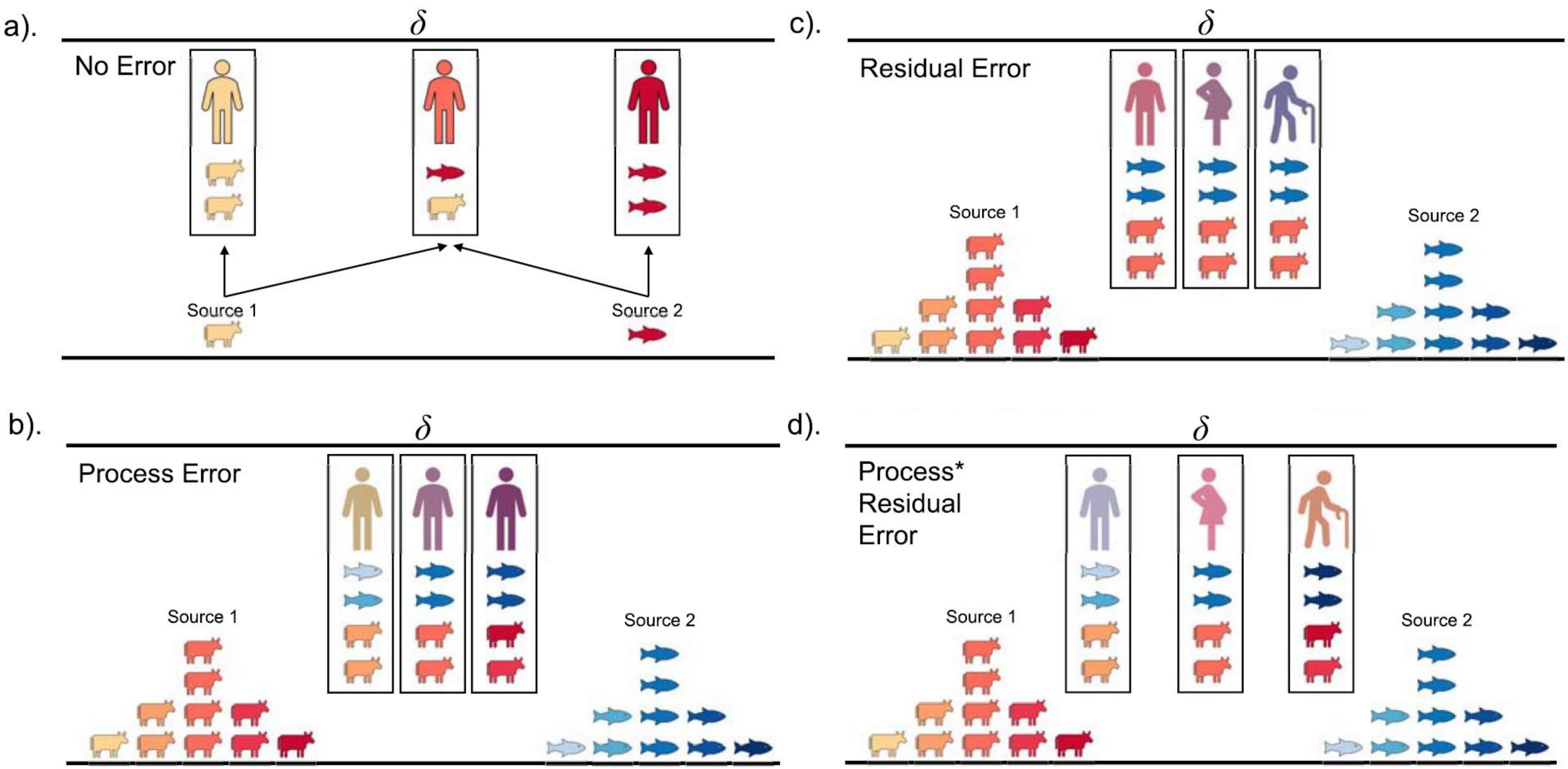
Cheung & Szpak 2020 J. Archaeo Method Theory - https://doi.org/10.1007/s10816-020-09492-5



Process error: sources are isotopically variable, and consumers subsample.
Residual error: inter-consumer differences in physiology influence their isotopic ratios

MixSIAR error structure

Cheung & Szpak 2020 J. Archaeo Method Theory - https://doi.org/10.1007/s10816-020-09492-5



Process error: sources are isotopically variable, and consumers subsample.
Residual error: inter-consumer differences in physiology influence their isotopic ratios

Note the impact on consumer δ without any diet change!

A complex Bayesian mixing model: MixSIAR

Analyzing mixing systems using a new generation of Bayesian tracer mixing models

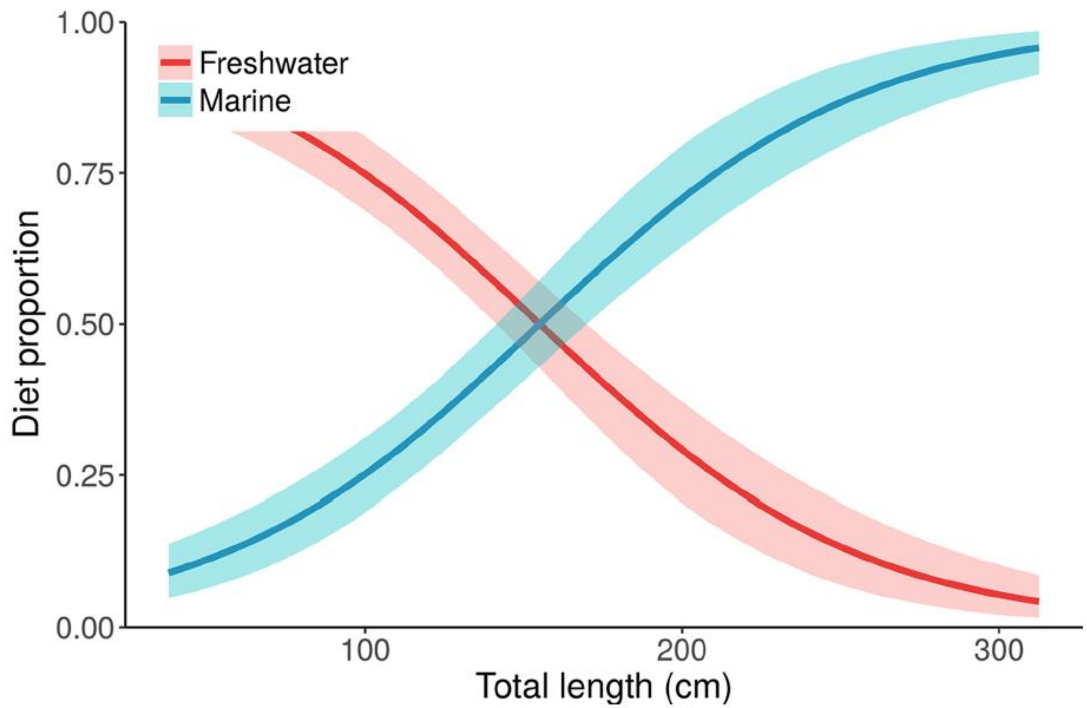
Brian C. Stock[1], Andrew L. Jackson[2], Eric J. Ward[3], Andrew C. Parnell[4], Donald L. Phillips[5] and Brice X. Semmens[1]

Stock et al. (2018), *PeerJ*, DOI 10.7717/peerj.5096

MixSIAR capabilities:

- Any number of tracers
- Categorical or continuous covariates
- Multiple error structures
- …

Drawback: computationally intensive



Ontogenic shift in resource use in *Alligator mississippiensis*

A complex Bayesian mixing model: MixSIAR

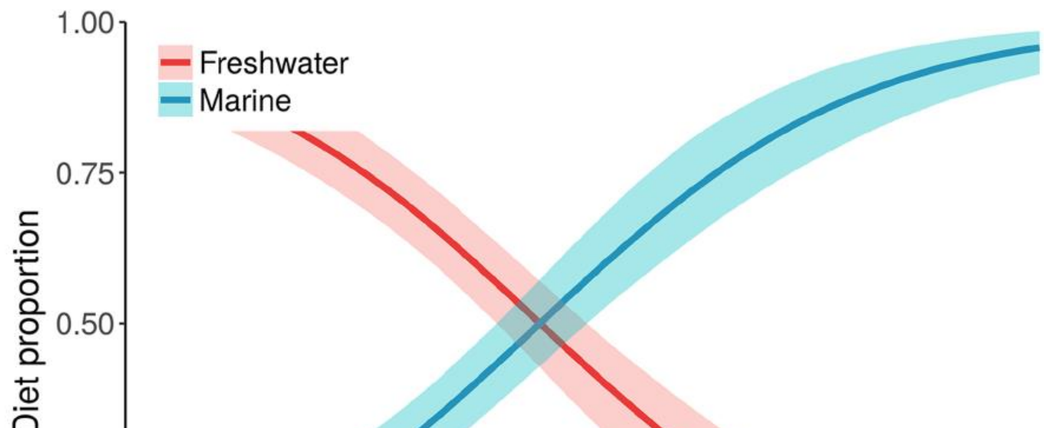
Analyzing mixing systems using a new generation of Bayesian tracer mixing models

Brian C. Stock[1], Andrew L. Jackson[2], Eric J. Ward[3], Andrew C. Parnell[4], Donald L. Phillips[5] and Brice X. Semmens[1]

Stock et al. (2018), *PeerJ*, DOI 10.7717/peerj.5096

MixSIAR capabilities:

- Any number of tracers
- Categorical or continuous covariates
- Multiple error structures
- …



Practical course: get the files at
doi.org/10.5281/zenodo.3903263

Ontogenic shift in resource use in *Alligator mississippiensis*

Mixing models: beyond diet analysis

Mixing models can be used to answer many questions in biogeochemistry, hydrology, ecology, …

Mixing models: beyond diet analysis

Mixing models can be used to answer many questions in biogeochemistry, hydrology, ecology, …

Zostera marina: most wide-ranging angiosperm of the Northern Hemisphere

Present in most of the Baltic Sea, including along the Polish coasts

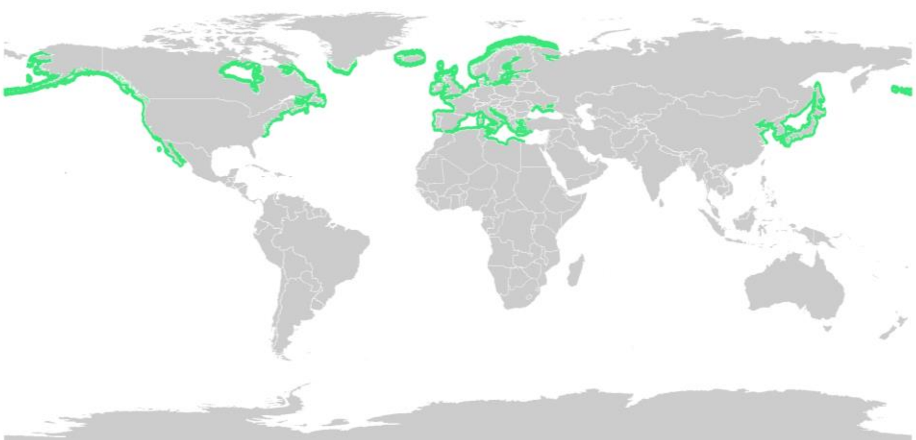
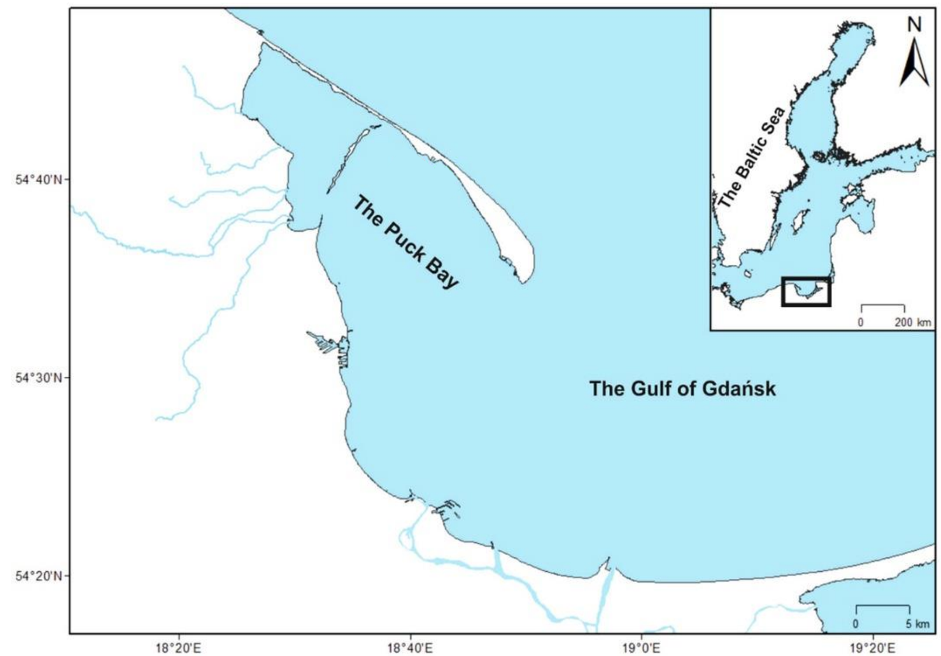


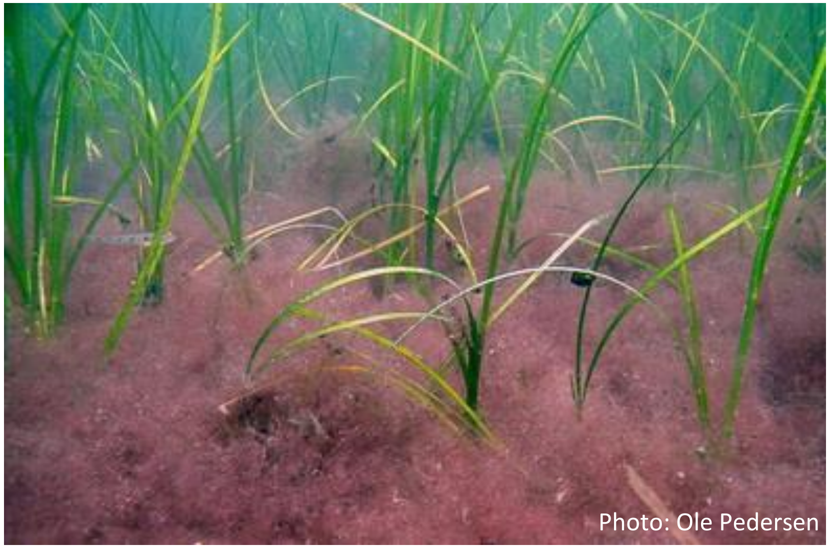
Image: Wikimedia Commons

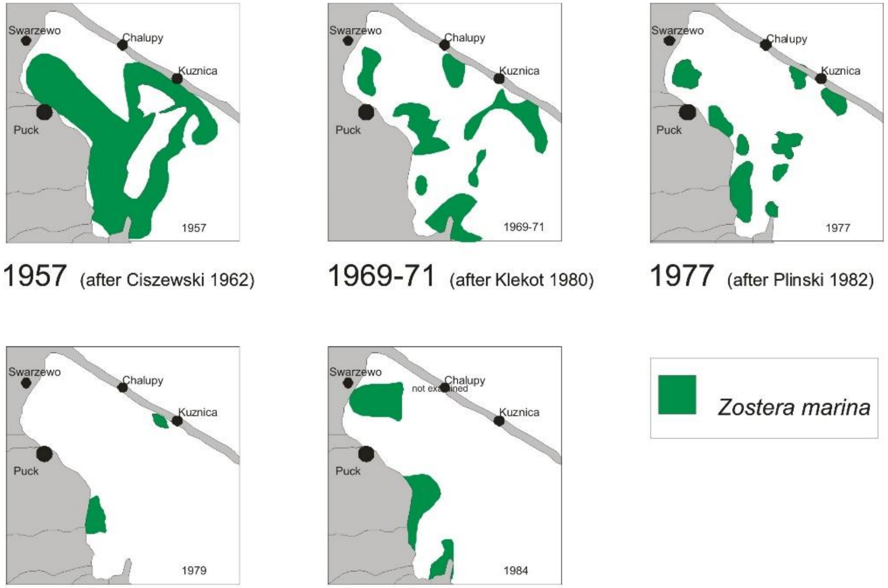


Mixing models: beyond diet analysis

Mixing models can be used to answer many questions in biogeochemistry, hydrology, ecology, …

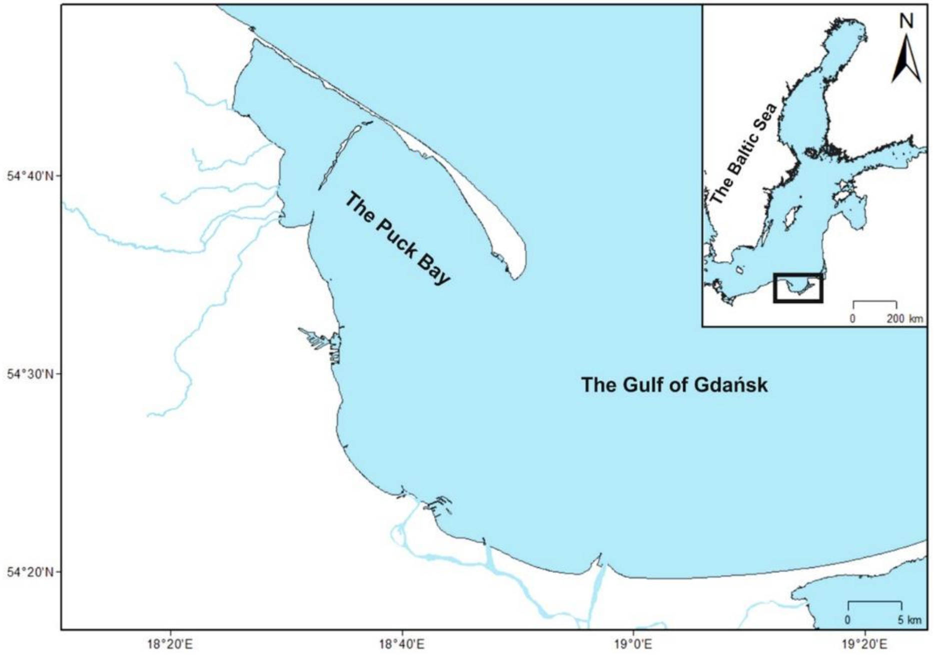
Puck Bay (Gulf of Gdansk): strong regression of meadow extent from the 50's to the 90's: eutrophication





1957 (after Ciszewski 1962) 1969-71 (after Klekot 1980) 1977 (after Plinski 1982)

1979 (after Ciszewski *et al.* 1992) 1984 (after Plinski 1986)



Mixing models: beyond diet analysis

Mixing models can be used to answer many questions in biogeochemistry, hydrology, ecology, …

In recent years: natural recovery of meadows, but density and biomass low compared to other meadows

Question: Are these recovering meadows capable of sustaining ecosystem services, notably blue carbon storage?

Image: Piotr Bałazy - iopan.pl/projects/Zostera

Mixing models: beyond diet analysis

Mixing models can be used to answer many questions in biogeochemistry, hydrology, ecology, …

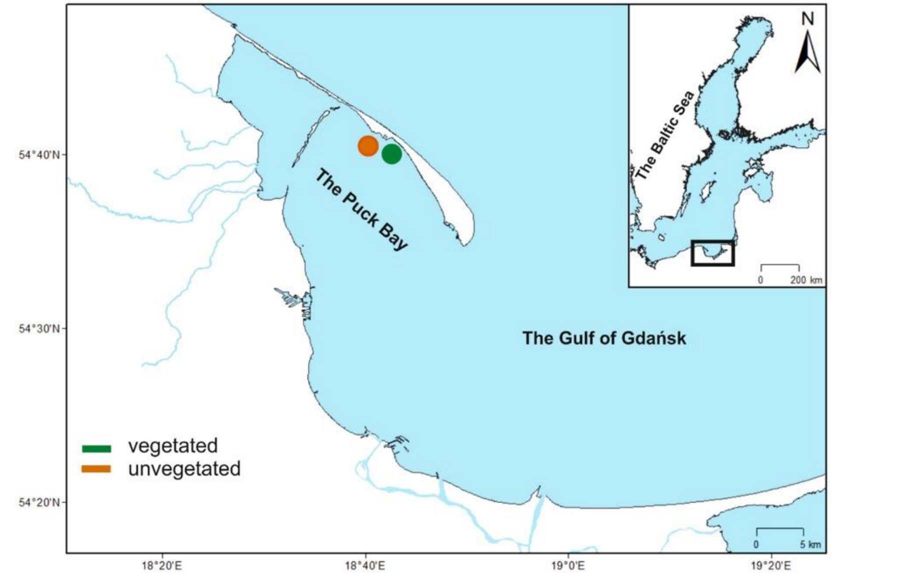
What is the composition of sediment-associated organic matter?

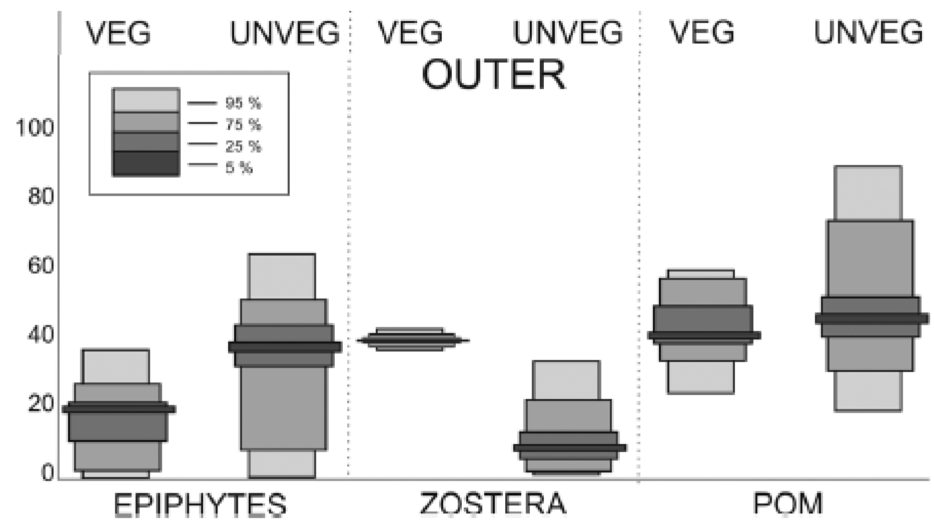
Comparison of vegetated and unvegetated zones

↓

Important contribution of seagrass production to sediments in the vegetated zone

Even at low density, presence of seagrass influences blue carbon storage



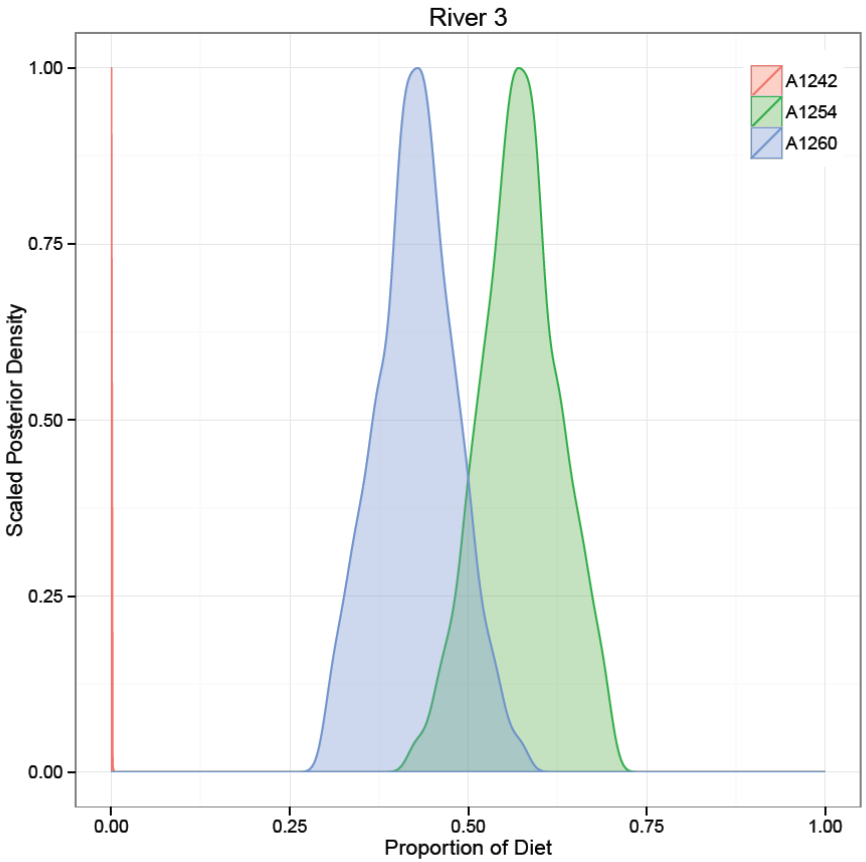


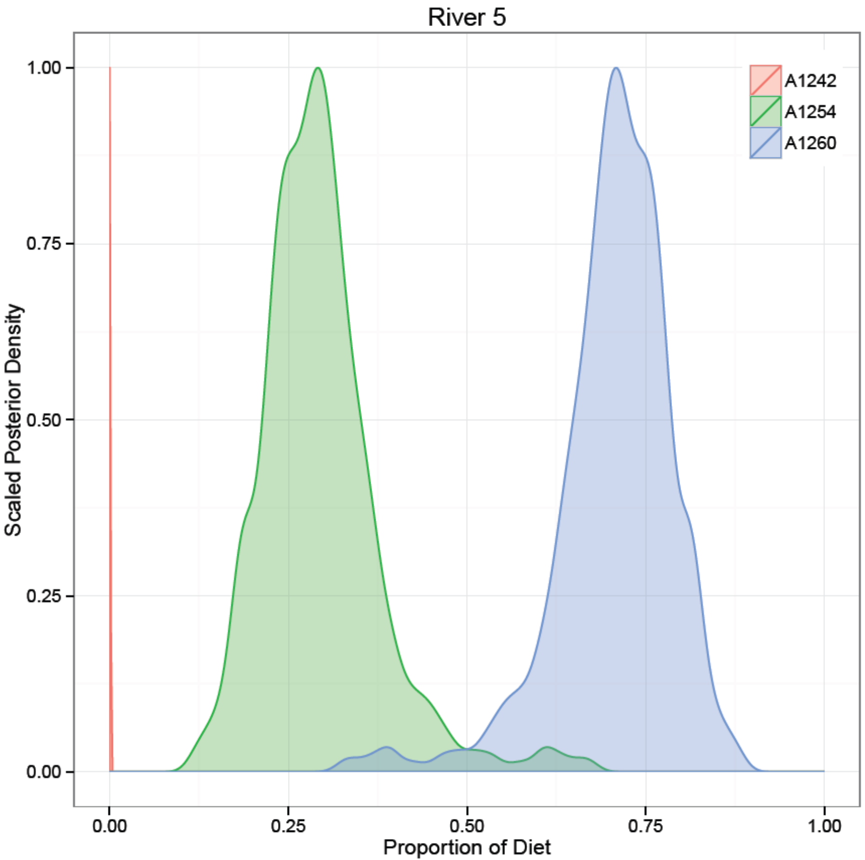
Jankowska et al. 2016 J Geophys Res Biogeosci. 121: 2918-2934

Mixing models: beyond diet analysis

Mixing models can be used to answer many questions in biogeochemistry, hydrology, ecology, …

Which commercial mixtures lead to seabass contamination by PCBs?





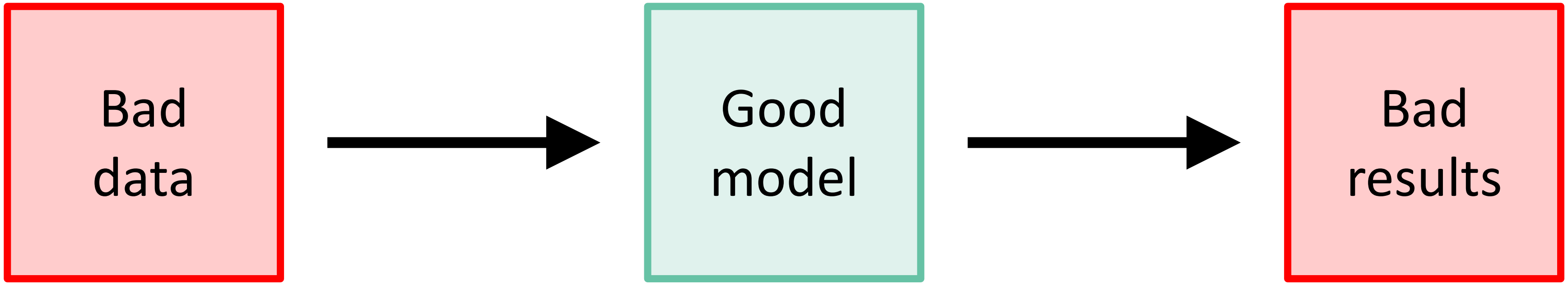
Schnitzler & Michel, Unpubl.

Building sensible mixing models

"Junk in, junk out" paradigm

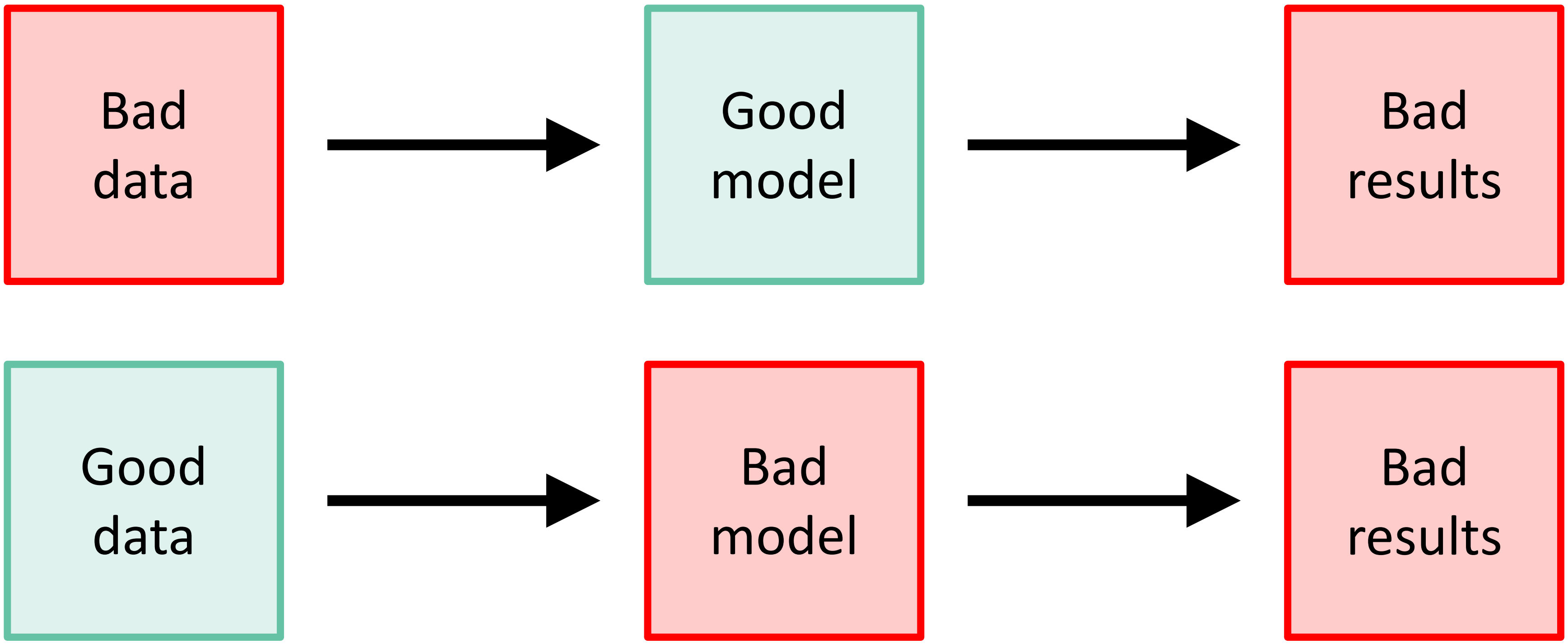
Building sensible mixing models

"Junk in, junk out" paradigm



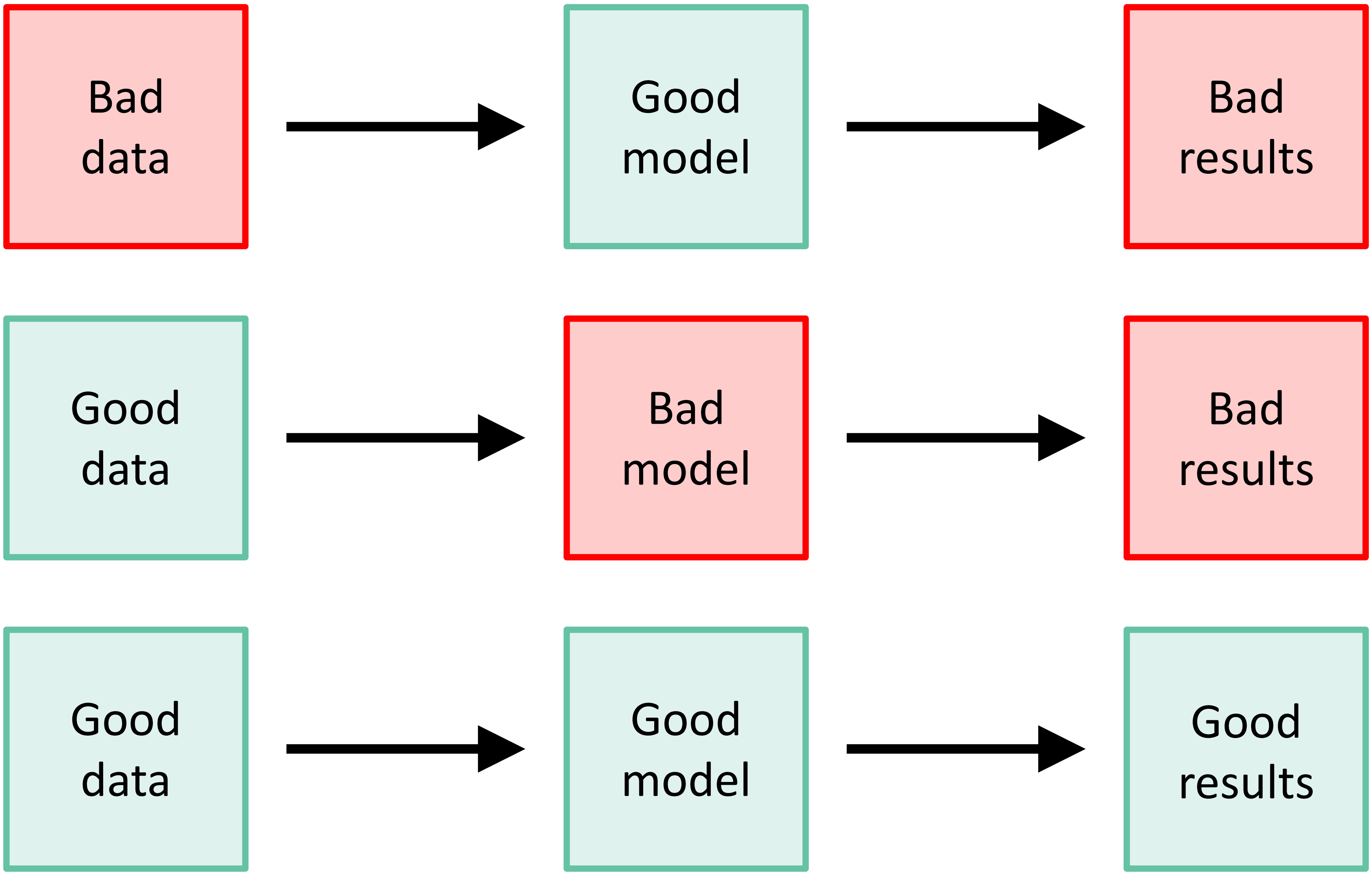
Building sensible mixing models

"Junk in, junk out" paradigm



Building sensible mixing models

"Junk in, junk out" paradigm



Building sensible mixing models

Good data

- Good characterization of food items (as important as consumers!)
- Sufficient replication (robust error estimates)
- Suitable TEFs (as close as your studied species as possible)

Building sensible mixing models

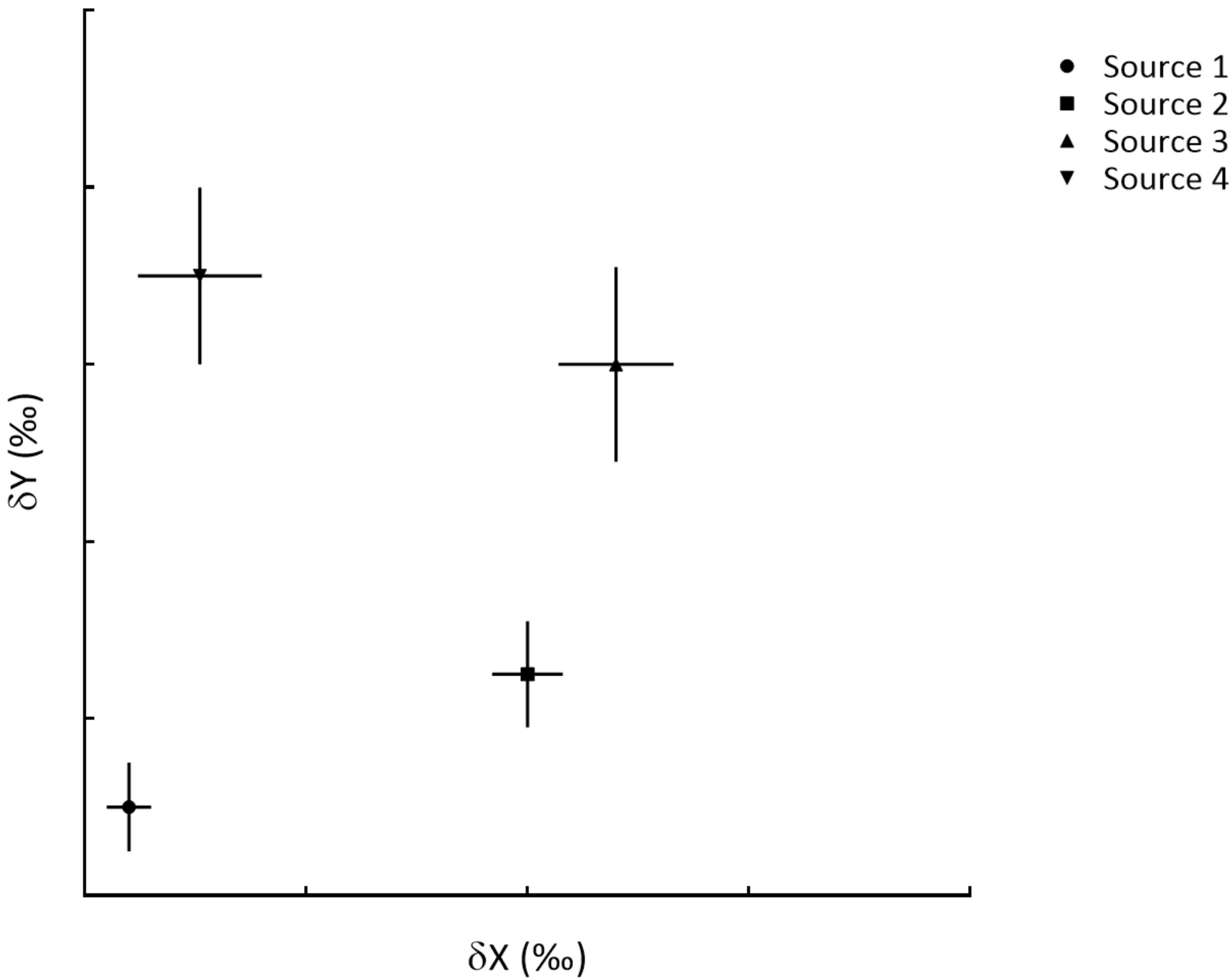
Good data

- Good characterization of food items (as important as consumers!)
- Sufficient replication (robust error estimates)
- Suitable TEFs (as close as your studied species as possible)

Good model

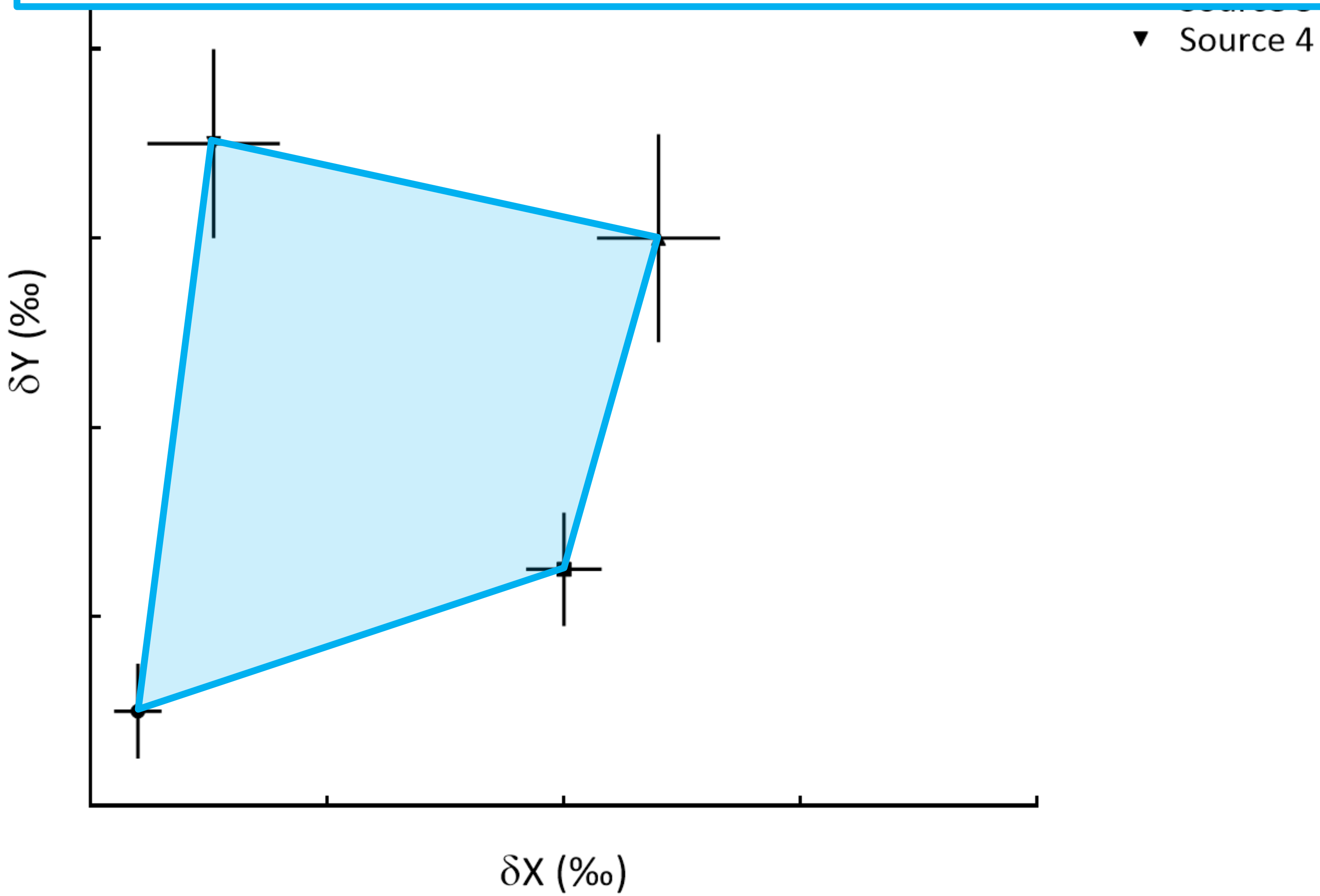
- Set the models parameters sensibly, and assess model performance
- Include all relevant food items (and only them)
- Make sure your model assumptions are met: plot your data

Mixing polygons

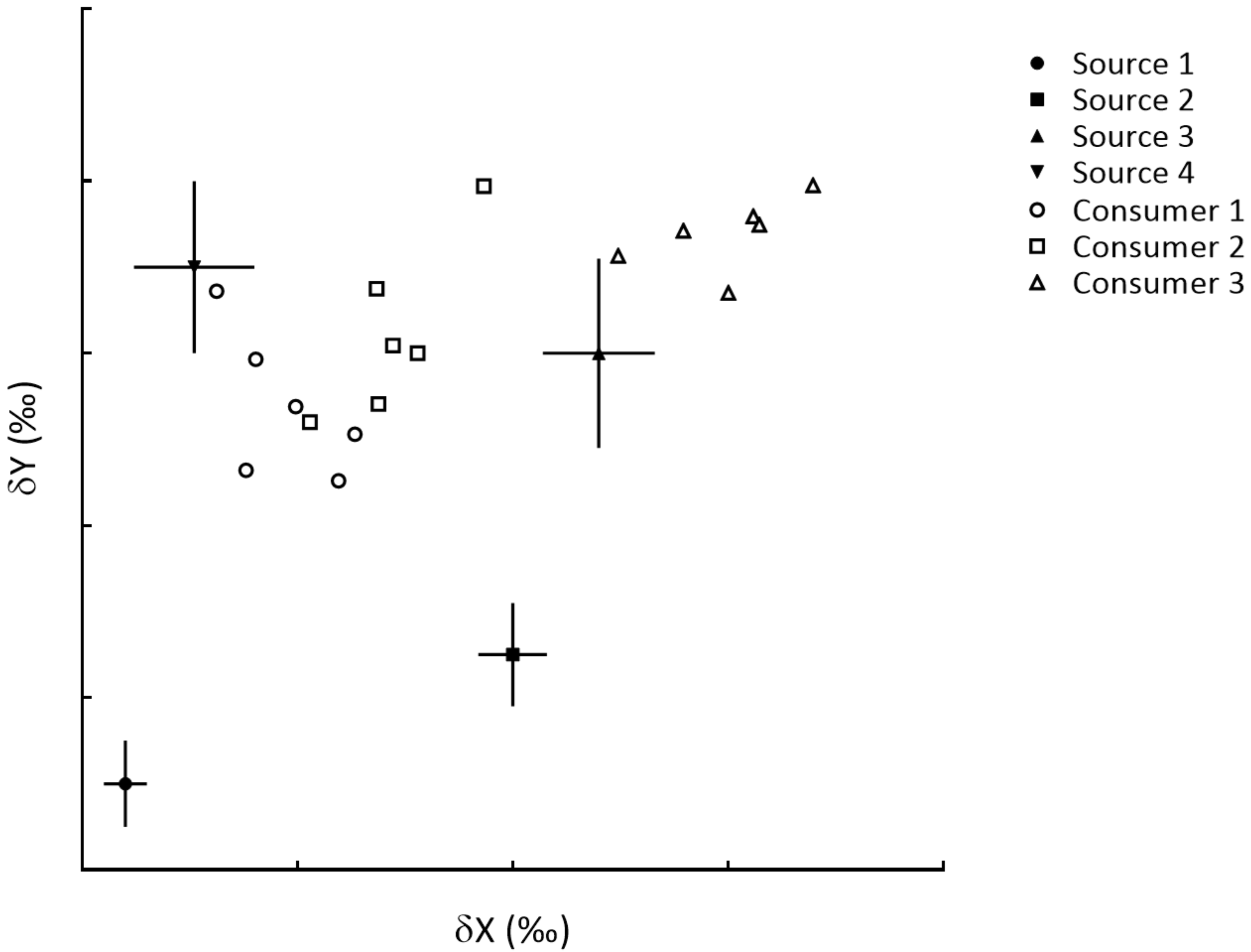


Mixing polygons

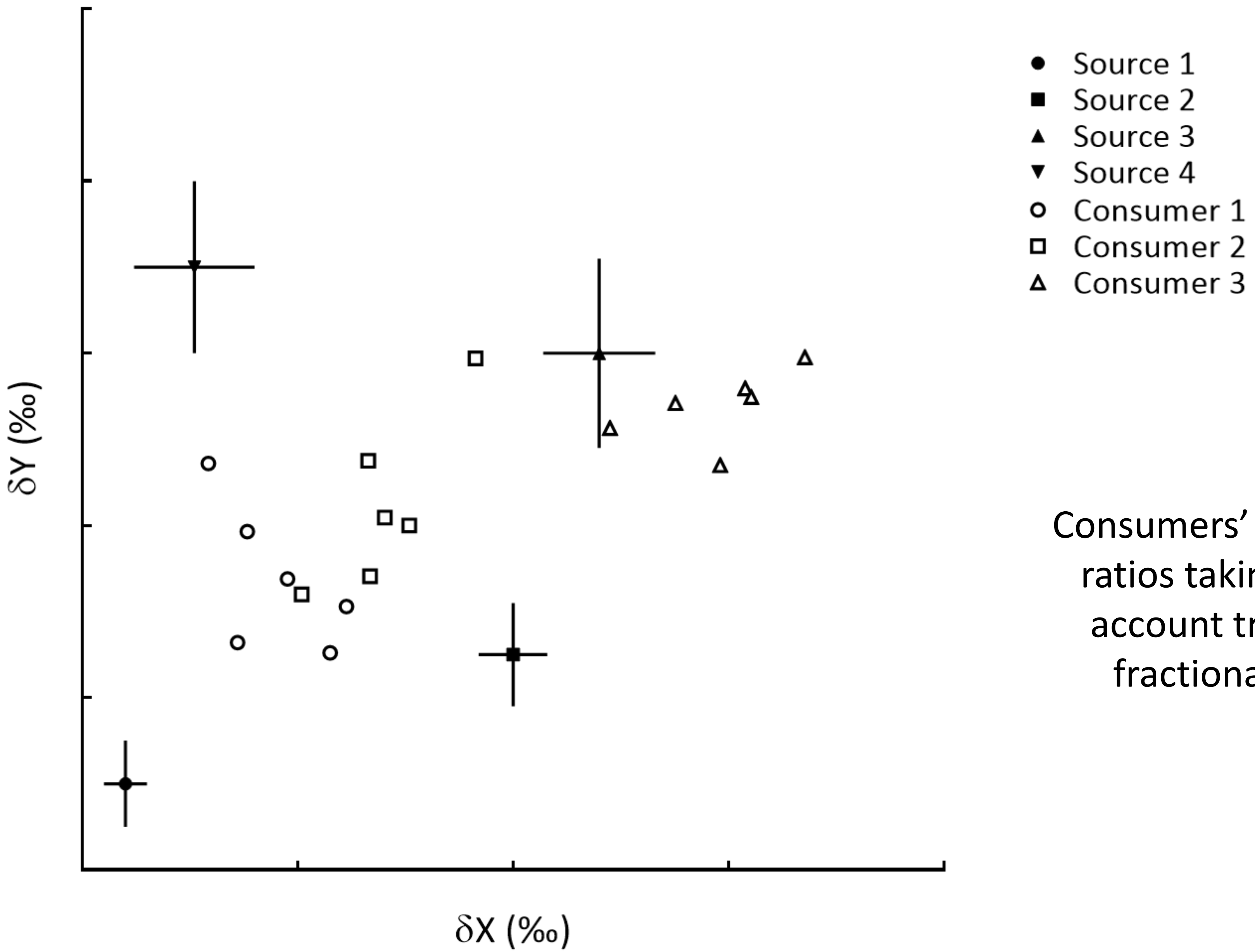
Your mixing model can only work with consumers that are within the "mixing polygon" defined by the sources' isotopic values



Mixing polygons

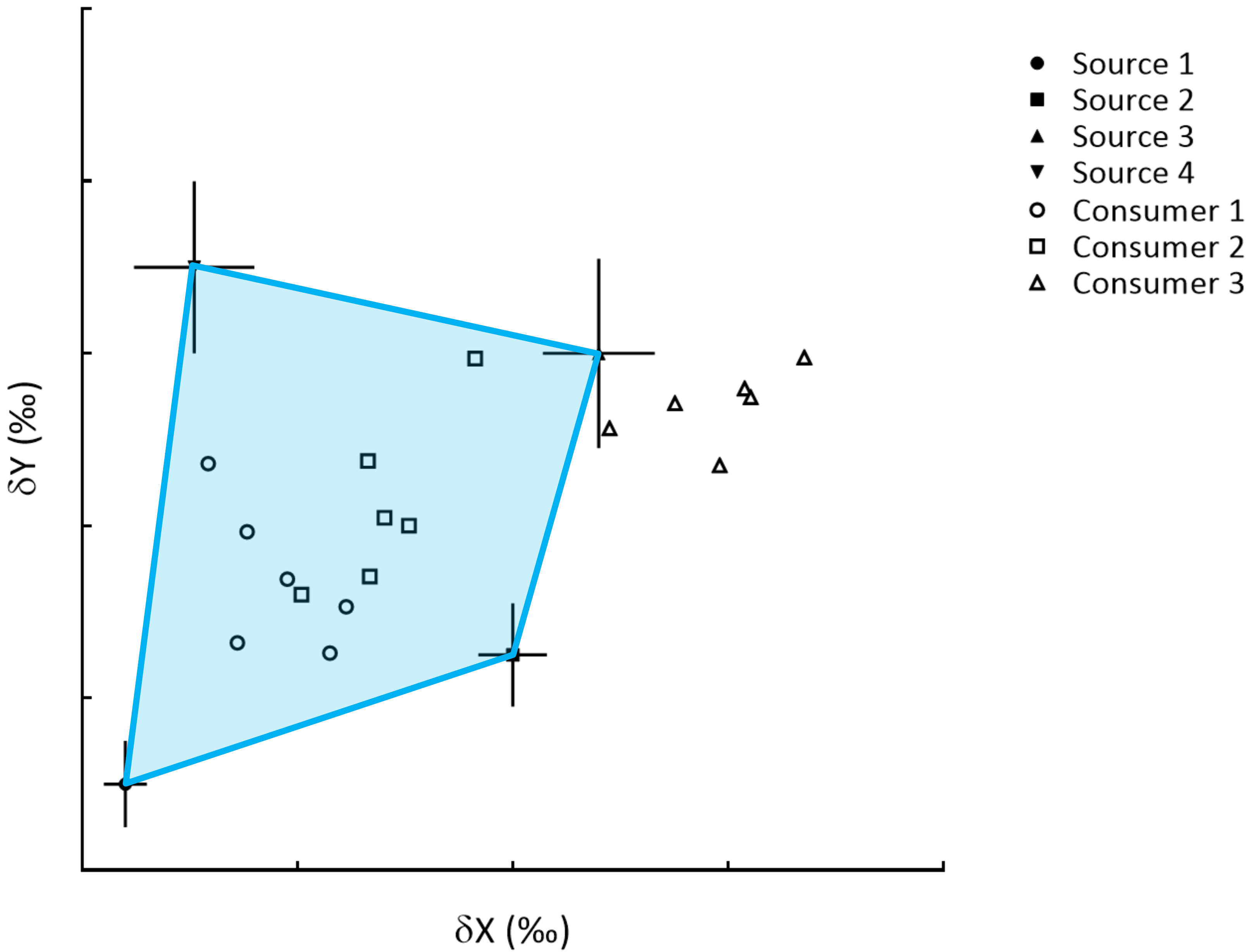


Mixing polygons



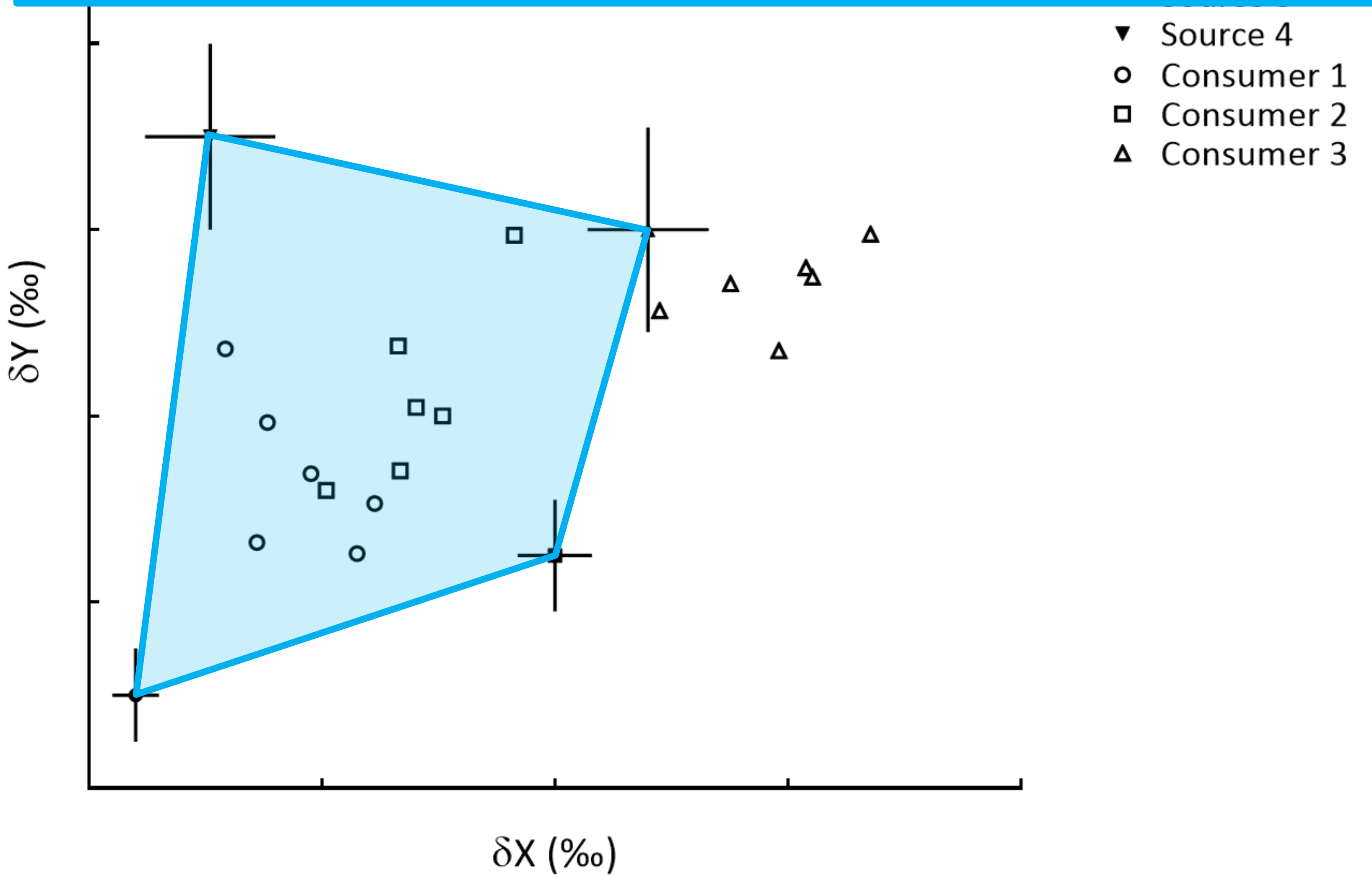
Consumers' isotopic ratios taking into account trophic fractionation

Mixing polygons

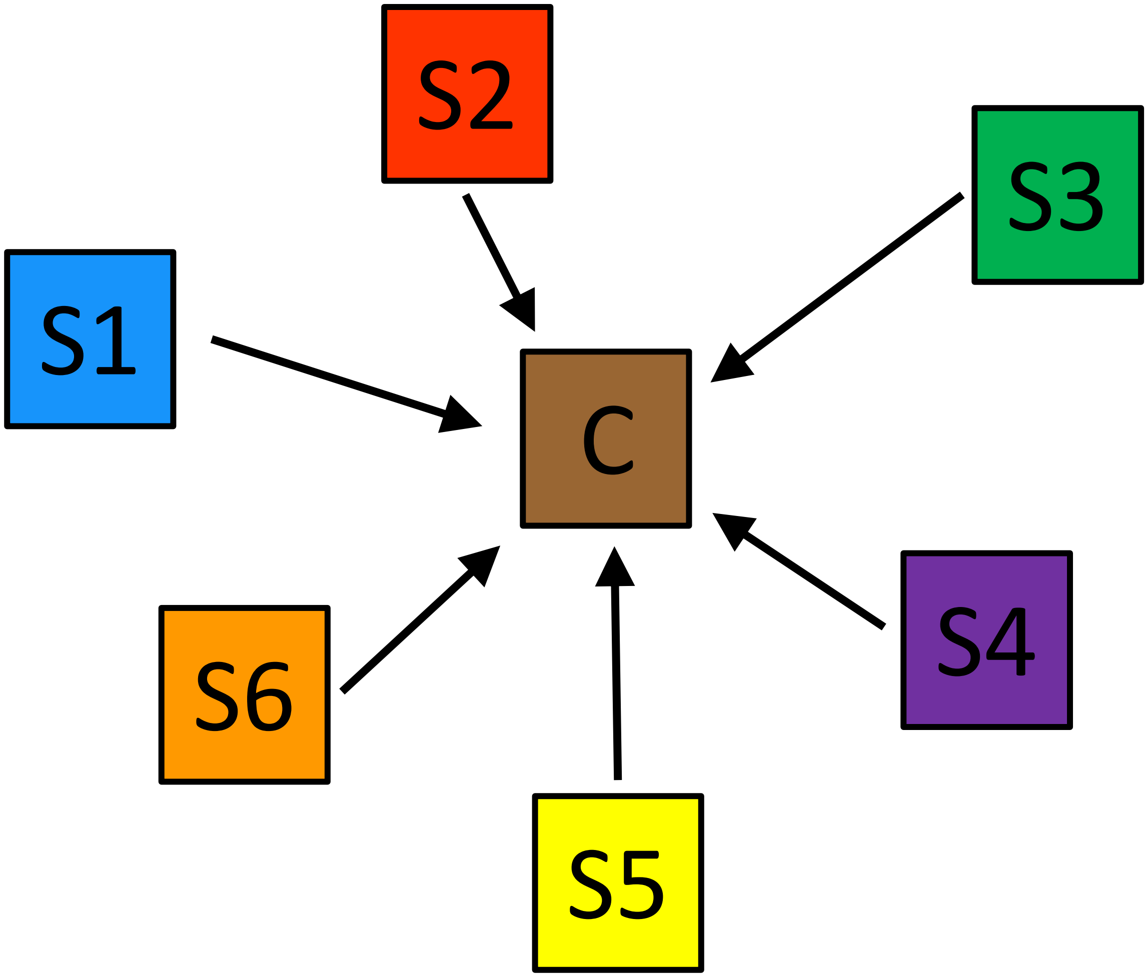


Mixing polygons

If, after correcting for fractionation, your consumers do not fit in the mixing polygon, it makes no sense to run a mixing model

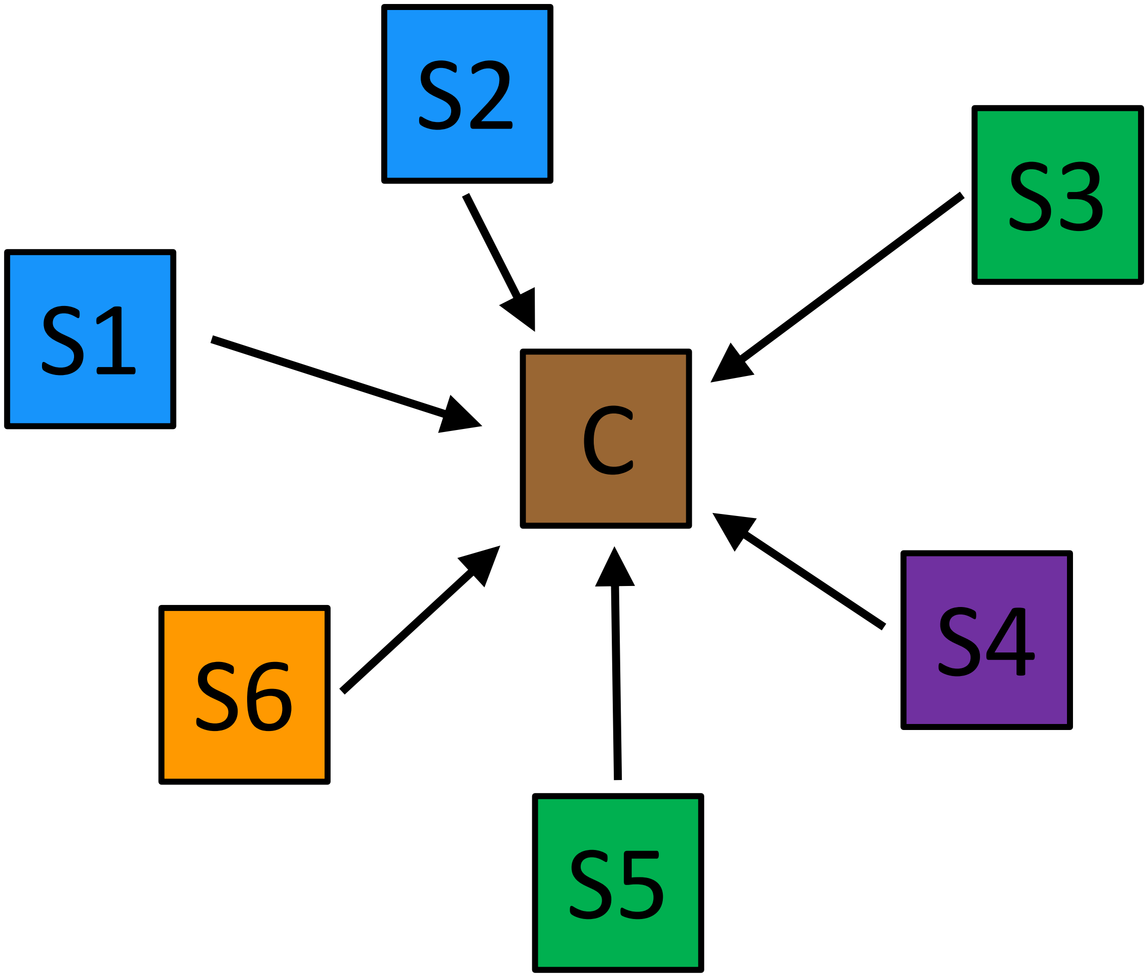


Isotopic similarity of food items

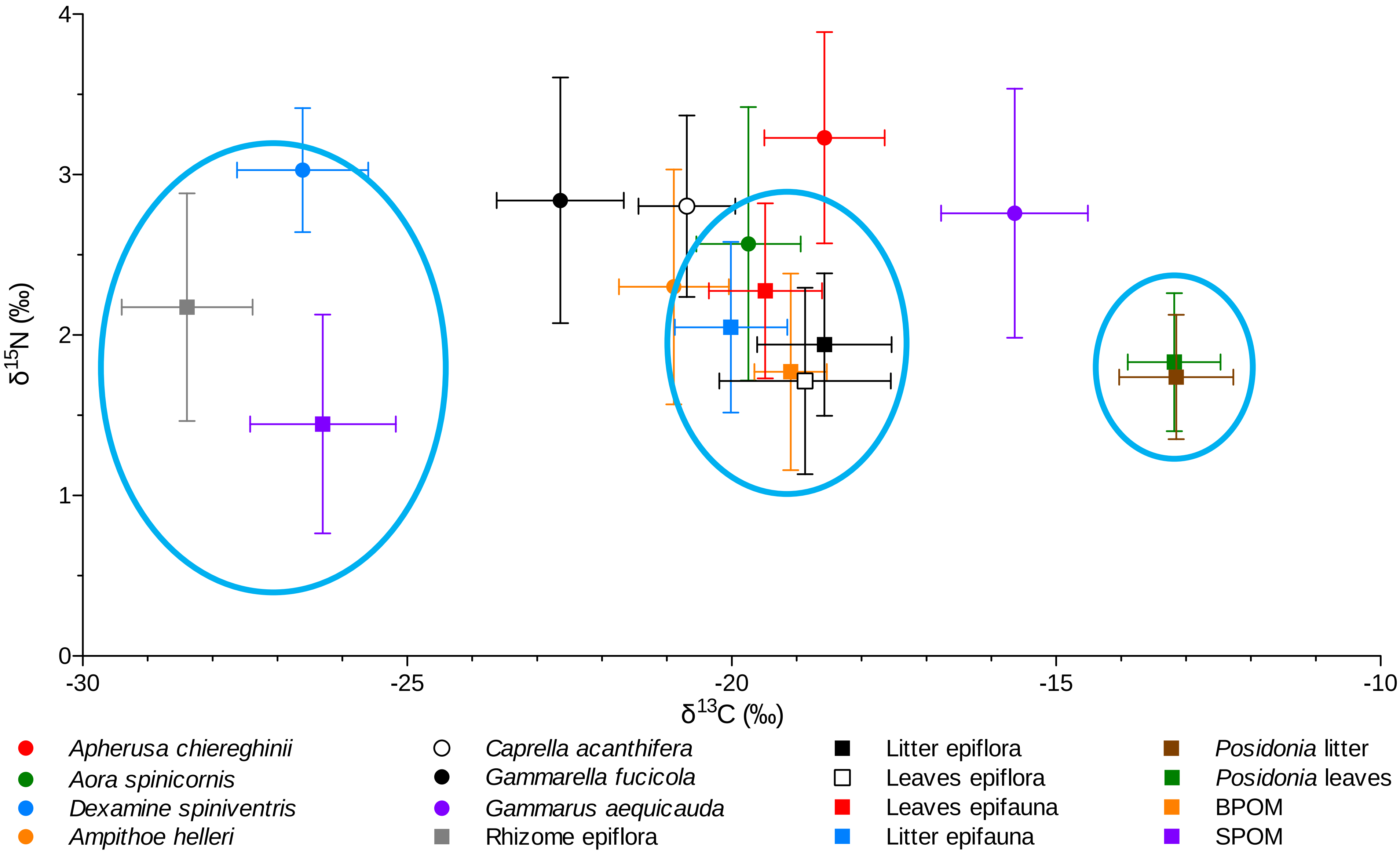


Isotopic similarity of food items

Common problem: some potential food items have the same isotopic composition…

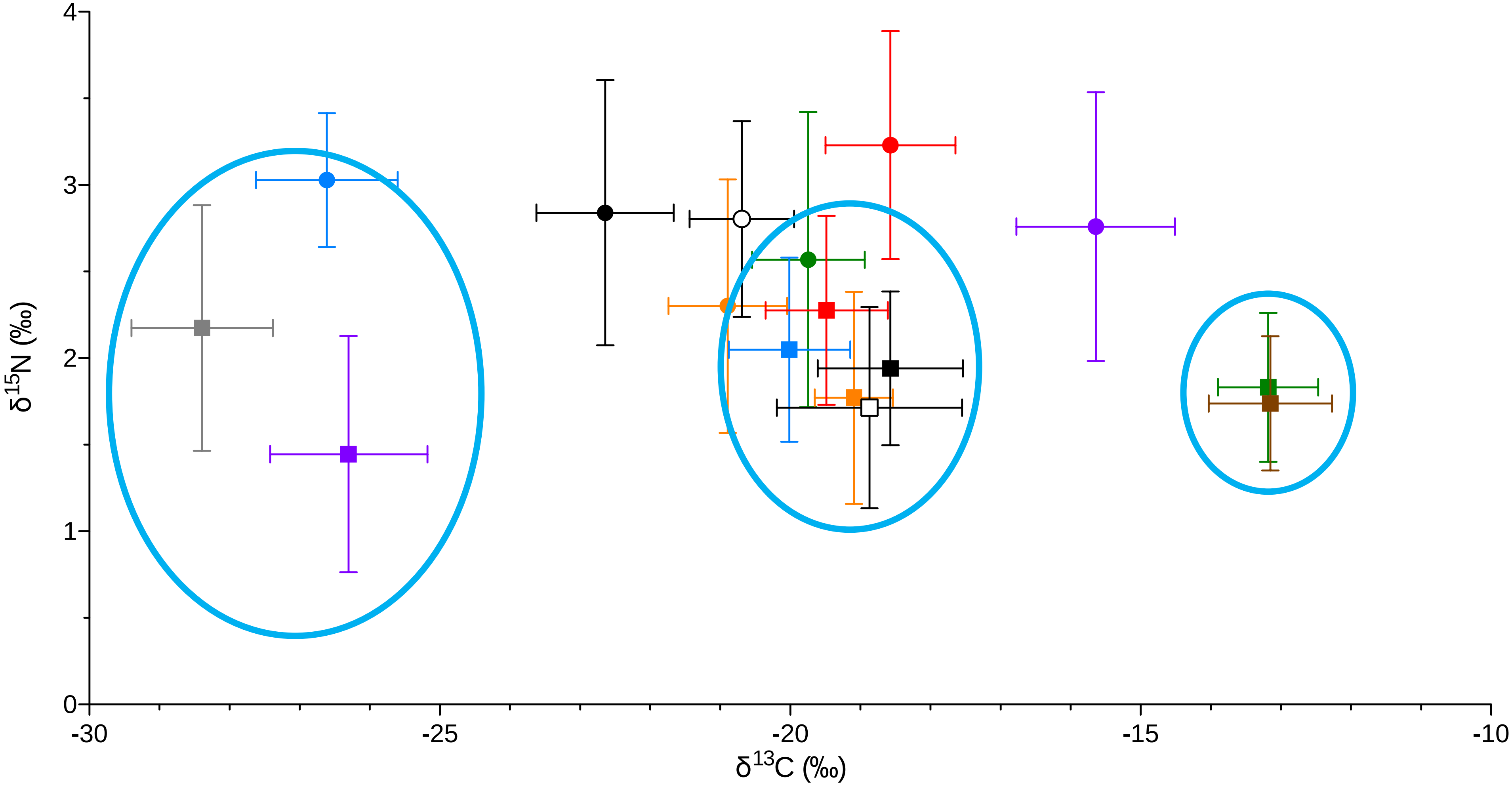


Isotopic similarity of food items



Michel et al 2015 Mar Ecol 36: 969-981

Isotopic similarity of food items



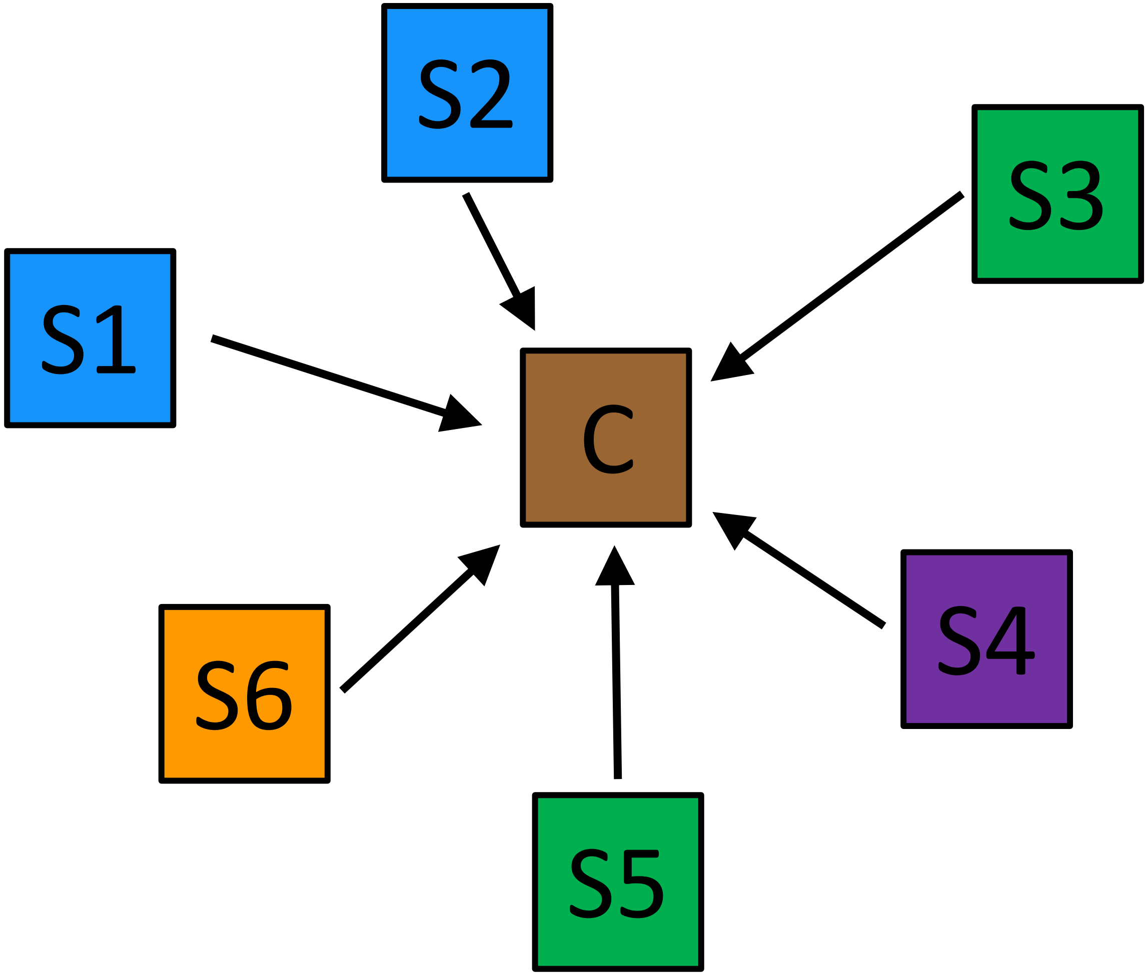
If isotopic compositions of food items are too similar, the model will not be able to tell them apart from one another

s

Michel et al 2015 Mar Ecol 36: 969-981

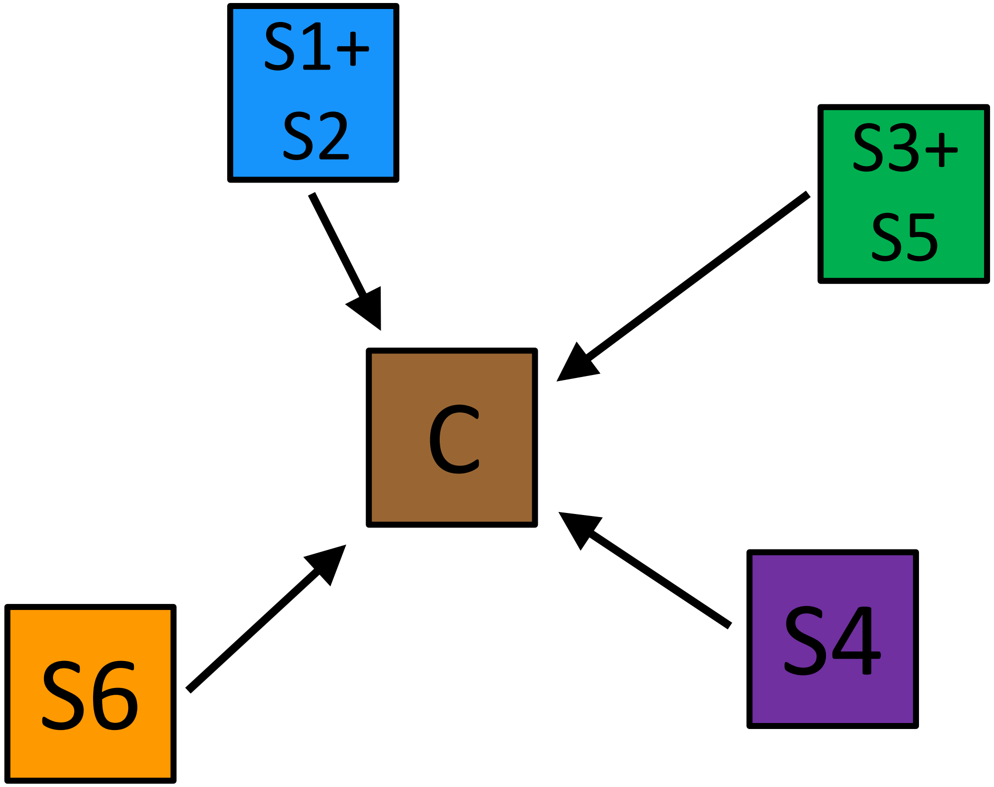
Isotopic similarity of food items

Solution 1: Aggregate the similar sources



Isotopic similarity of food items

Solution 1: Aggregate the similar sources

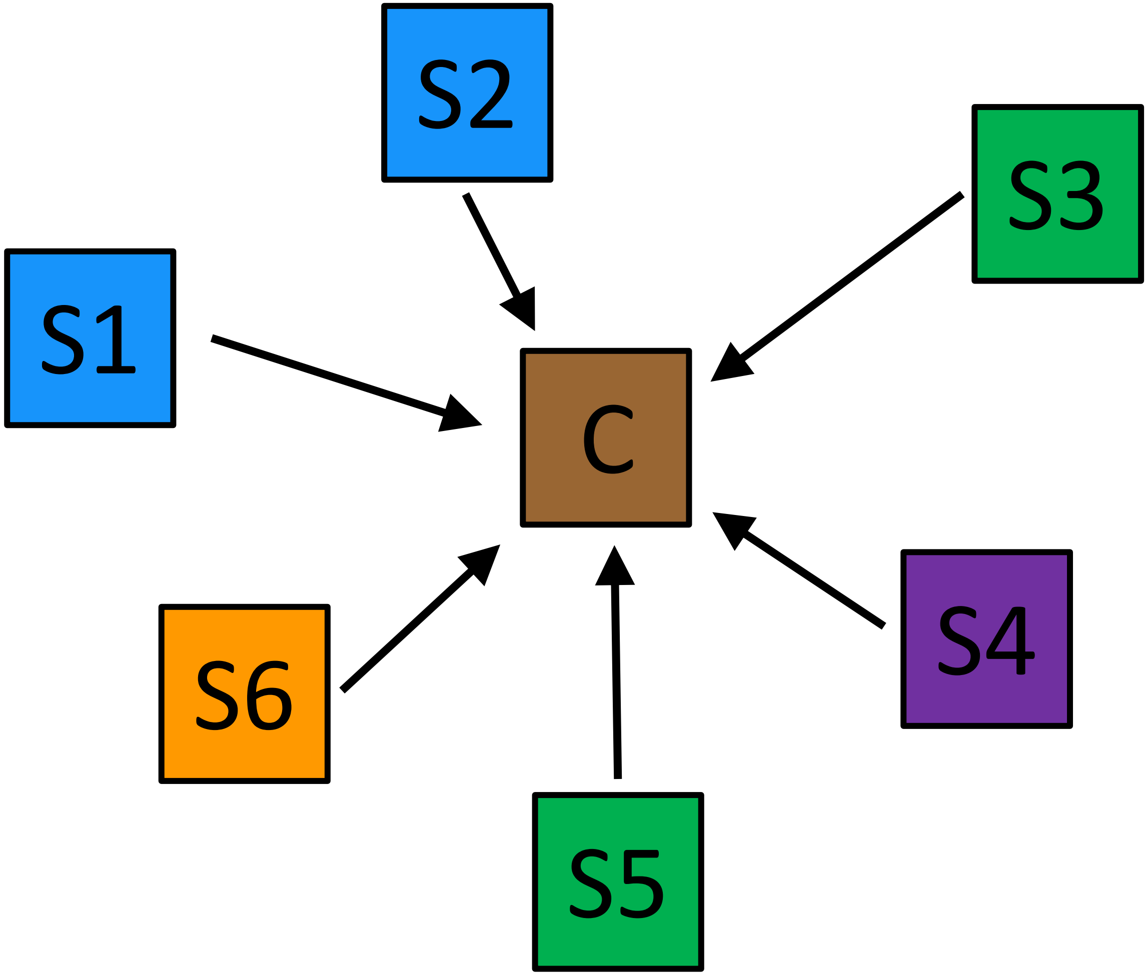


Realistic from a modeling point of view, but can lead to loss of ecological information

Isotopic similarity of food items

Solution 1: Aggregate the similar sources

Solution 2: Combine SI with other tracers that can discriminate the food items



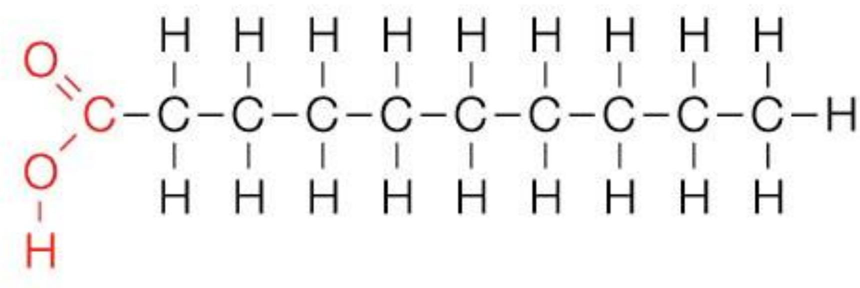
Alternative tracers: fatty acids

"Building blocks" of lipids

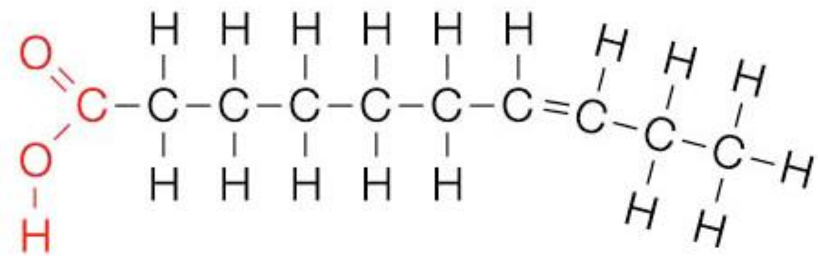
Long carbon chain with a final acid group

Source : www.nature.com

Saturated



Unsaturated



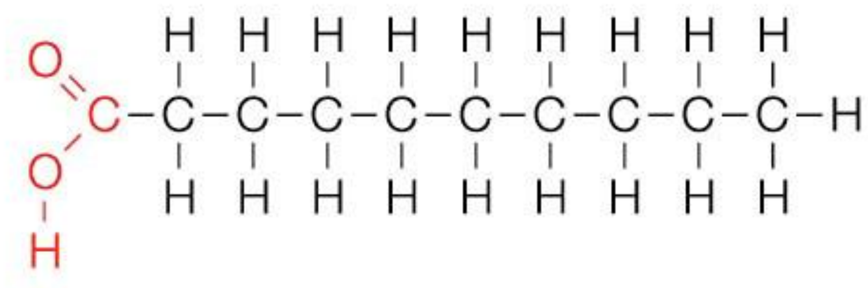
Alternative tracers: fatty acids

"Building blocks" of lipids

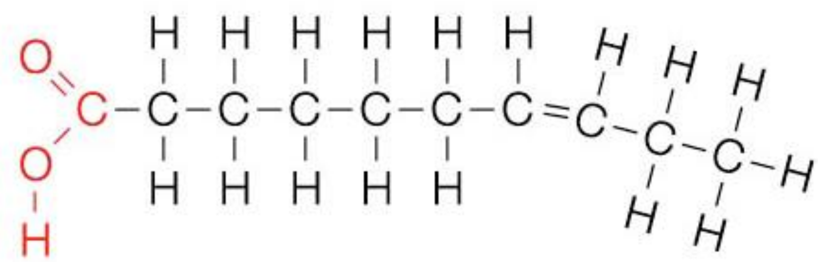
Long carbon chain with a final acid group

Source : www.nature.com

Saturated



Unsaturated



During digestion, lipids are degraded but fatty acids are incorporated in the consumer's tissues in a conservative way

➔ A consumer's fatty acid composition is similar to the one of its food sources

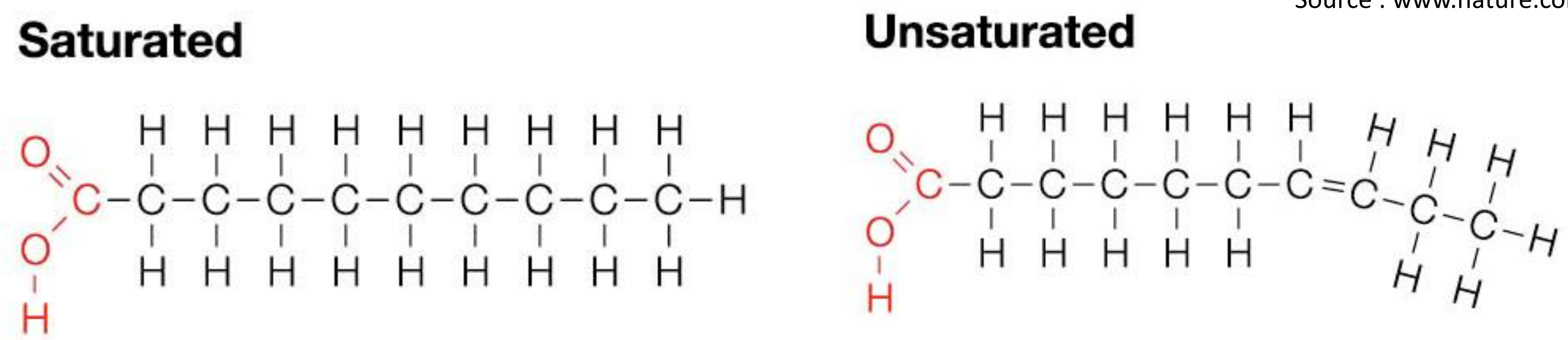
➔ Fatty acids can be used as trophic markers and combined to stable isotopes to build mixing models

Alternative tracers: fatty acids

"Building blocks" of lipids

Long carbon chain with a final acid group

Source : www.nature.com



+: Limits loss of ecological info

-: More assumptions (what about fatty acid biosynthesis?)

➔ A consumer's fatty acid composition is similar to the one of its food sources

➔ Fatty acids can be used as trophic markers and combined to stable isotopes to build mixing models

Building sensible mixing models

NRC Research Press

REVIEW

Best practices for use of stable isotope mixing models in food-web studies

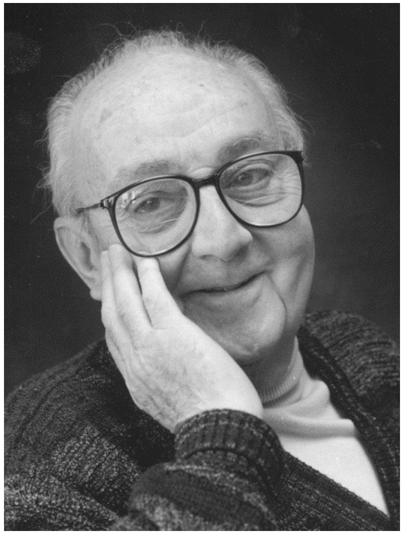
Donald L. Phillips, Richard Inger, Stuart Bearhop, Andrew L. Jackson, Jonathan W. Moore, Andrew C. Parnell, Brice X. Semmens, and Eric J. Ward

Can. J. Zool. 92: 823–835 (2014) dx.doi.org/10.1139/cjz-2014-0127 Published at www.nrcresearchpress.com/cjz on 27 August 2014.

1. Use prior knowledge to identify relevant questions
2. Consider what's known about the animal's diet
3. Plan your sampling design well
4. Use appropriate trophic fractionation factors
5. Plot your data before running your model
6. Include all relevant food items, in an informed way
7. Group your sources when isotopically and/or ecologically relevant
8. Don't forget about concentration dependence and isotopic routing
9. Consider and incorporate uncertainties
10. Report distribution of results
11. Your model will always be an oversimplification of a complex ecological reality. Assess its performance. Remember its limitations!

Building sensible mixing models

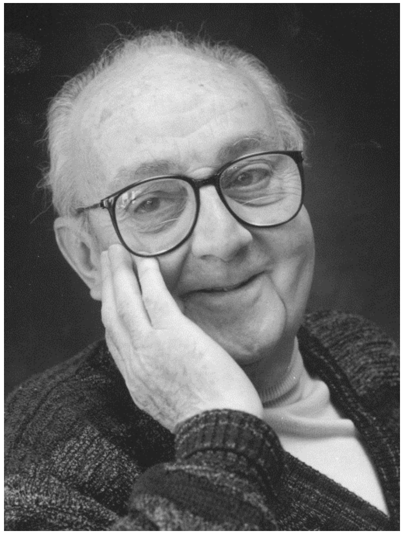
"Remember that all models are wrong; the practical question is how wrong do they have to be to not be useful."



George E.P. Box
1919-2013

Building sensible mixing models

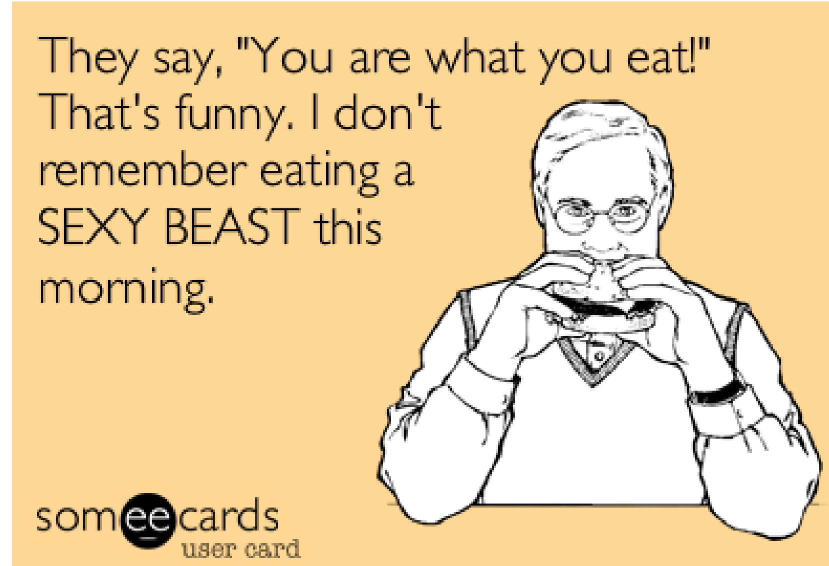
"Remember that all models are wrong; the practical question is how wrong do they have to be to not be useful."

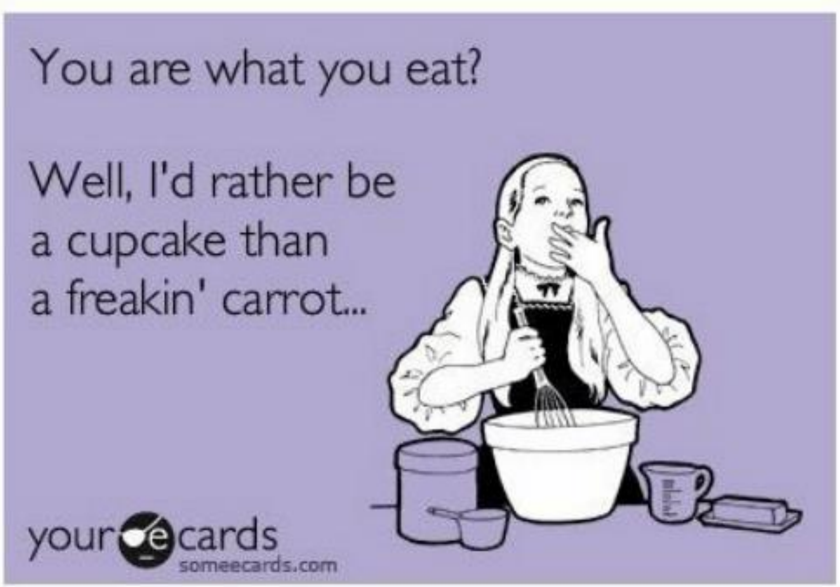


George E.P. Box
1919-2013

Mixing models are not "magic wands", nor perfect tools

However, when used sensibly, they offer an efficient way to assess animal diet and explore ecological patterns





Thanks for your attention

References & further reading

Cheung, C. & Szpak, P. 2020. Interpreting Past Human Diets Using Stable Isotope Mixing Models. *Journal of Archaeological Method and Theory*, 37, https://doi.org/10.1007/s10816-020-09492-5.

Fry, B. 2006. Stable Isotope Ecology. Springer, 308 pp, https://doi.org/10.1007/0-387-33745-8.

Jankowska, E., Michel, L.N., Zaborska, A. & Włodarska-Kowalczuk, M. 2016. Sediment carbon sink in low-density temperate eelgrass meadows (Baltic Sea). *Journal of Geophysical Research: Biogeosciences*, **121**, 1–17, https://doi.org/10.1002/2016JG003424.

Komorita, T., Kajihara, R., Tsutsumi, H., Shibanuma, S., Yamada, T. & Montani, S. 2014. Food sources for *Ruditapes philippinarum* in a coastal lagoon determined by mass balance and stable isotope approaches. *PLoS One*, **9**, e86732, https://doi.org/10.1371/journal.pone.0086732.

Mascart, T., De Troch, M., Remy, F., Michel, L.N. & Lepoint, G. 2018. Seasonal dependence on seagrass detritus and trophic niche partitioning in four copepod eco-morphotypes. *Food Webs*, **16**, e00086, https://doi.org/10.1016/j.fooweb.2018.e00086.

Michel, L.N., Dauby, P., Gobert, S., Graeve, M., Nyssen, F., Thelen, N. & Lepoint, G. 2015. Dominant amphipods of *Posidonia oceanica* seagrass meadows display considerable trophic diversity. *Marine Ecology*, **36**, 969–981, https://doi.org/10.1111/maec.12194.

Moore, J.W. & Semmens, B.X. 2008. Incorporating uncertainty and prior information into stable isotope mixing models. *Ecology Letters*, **11**, 470–480, https://doi.org/10.1111/j.1461-0248.2008.01163.x.

Newsome, S.D., del Rio, C.M., Bearhop, S. & Phillips, D.L. 2007. A niche for isotopic ecology. *Frontiers in Ecology and the Environment*, **5**, 429–436, https://doi.org/10.1890/060150.01.

Parnell, A.C., Inger, R., Bearhop, S. & Jackson, A.L. 2010. Source partitioning using stable isotopes: Coping with too much variation. *PLoS One*, **5**, e9672, https://doi.org/10.1371/journal.pone.0009672.

References & further reading

Parnell, A.C., Phillips, D.L., Bearhop, S., Semmens, B.X., Ward, E.J., Moore, J.W., Jackson, A.L., Grey, J., Kelly, D.J. & Inger, R. 2013. Bayesian stable isotope mixing models. *Environmetrics*, **24**, 387–399, https://doi.org/10.1002/env.2221.

Phillips, D., Inger, R., Bearhop, S., Jackson, A., Moore, J., Parnell, A., Semmens, B. & Ward, E. 2014. Best practices for use of stable isotope mixing models in food-web studies. *Canadian Journal of Zoology*, **92**, 823–835, https://doi.org/10.1139/cjz-2014-0127.

Phillips, D.L. & Gregg, J.W. 2003. Source partitioning using stable isotopes: coping with too many sources. *Oecologia*, **136**, 169–261, https://doi.org/10.1007/s00442-003-1218-3.

Stock, B.C., Jackson, A., Ward, E.J., Parnell, A.C., Phillips, D.L. & Semmens, B.X. 2018. Analyzing mixing systems using a new generation of Bayesian tracer mixing models. *PeerJ*, e5096, https://doi.org/10.7717/peerj.5096.