Overview of Tools to Analyse Stable Isotopes.

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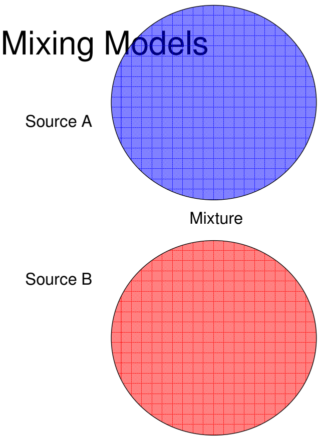
Stable Isotopes & Predator Prey Meeting. University of Tromsø, Norway

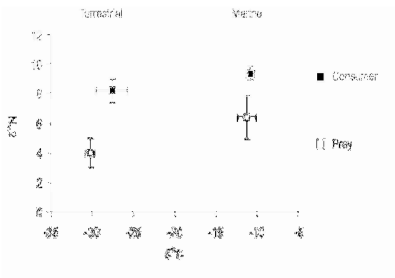
Stable Isotope Mixing Models

- Mixing problem
- A mixture is made up of two sources
- What proportions of each source are found in the mixture?



You are what you eat (almost)

- Consumer stable isotope ratios reflect diet
- Need to adjust for trophic enrichment
- Trophic Enrichment Factor (TEF)
- Δ C = 0 - 1‰
- Δ N = 2 - 4‰



TEFs are variable

Geometric Procedures

- •Until 2001 proportional contribution of sources to a mixture determined by measuring Euclidean distances of sources form mixture
- •However, assumes all food sources are utilised.
- •Leads to;
- •Underestimation of common food sources.
- •Overestimation of rare food sources
- •Demonstrated by Phillips 2001 using other authors work.

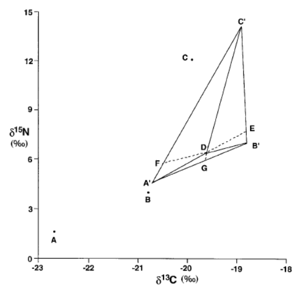
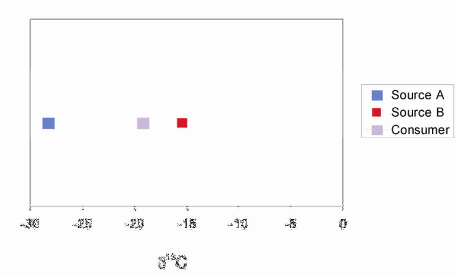


Fig. 1 Plot of dual isotopic compositions of food sources *A*, *B*, and *C*, and predator *D*. *A′*, *B′*, and *C′* represent the food source isotopic composition after adjustment for trophic fractionation. As in Table 1, numerical values for this example were taken from mean isotopic ratios reported by Szepanski et al. (1999) for *A* moose, *B* caribou, *C* salmon, and *D* interior Alaska wolf

From Szepanski Ben-David & Van Ballenberghe (1999)

Simple 1 Isotope 2 Source Mixing Model

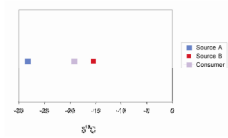


Linear Mixing Model

$\delta^{13}C_{consumer} = f_{source\ A}\ \delta^{13}C_{sourceA} + f_{source\ B}\ \delta^{13}C_{sourceB}$

$f_{source\ A} + f_{source\ B} = 1$

$$f_{source\ A} = \frac{\delta^{13}C_{consumer} - \delta^{13}C_{sourceB}}{\delta^{13}C_{sourceA} - \delta^{13}C_{sourceB}}$$

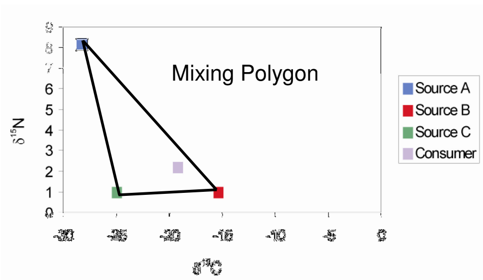


$$f_{source\ A} = \frac{-19.2\ -\ -15.4}{-28.3\ -\ -15.4} = \frac{-3.8}{-12.9}\quad 0.29$$

So $f_{source\ B} = 0.71$

Phillips (2001)

2 Isotope 3 Source Mixing Model



2 Isotope 3 Source Mixing Model

$\delta^{13}C_{consumer} = f_{source\ A}\ \delta^{13}C_{sourceA} + f_{source\ B}\ \delta^{13}C_{sourceB} + f_{source\ C}\ \delta^{13}C_{sourceC}$

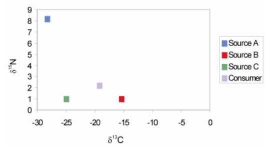
$\delta^{15}N_{consumer} = f_{source\ A}\ \delta^{15}N_{sourceA} + f_{source\ B}\ \delta^{15}N_{sourceB} + f_{source\ C}\ \delta^{15}N_{sourceC}$

$f_{source\ A} + f_{source\ B} + f_{source\ C} = 1$

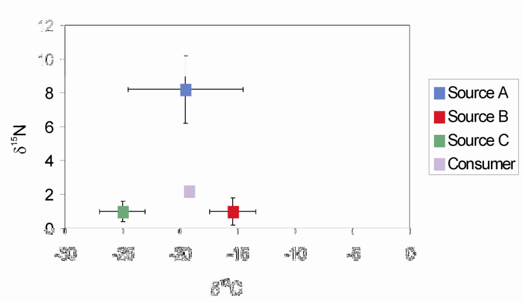
$f_{source\ A} = 0.16$

$f_{source\ B} = 0.67$

$f_{source\ C} = 0.17$



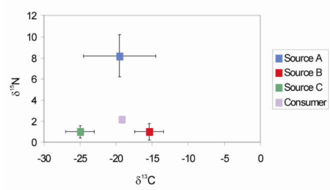
2 Isotope 3 Source Mixing Model With Source Variation



2 Isotope 3 Source Mixing Model With Source Variation

IsoError – Phillips & Gregg (2001)

	Mean	95% CI
Source A	0.17	0.0 – 0.40
Source B	0.50	0.1 – 0.92
Source C	0.32	0.0 – 0.73



2 Isotope 3 Source Mixing Model With Concentration Dependance

Models so far assume that food sources have equal concentrations of each element

High Nitrogen

Low Nitrogen

2 Isotope 3 Source Mixing Model With Concentration Dependance

IsoError – Phillips & Koch (2001)

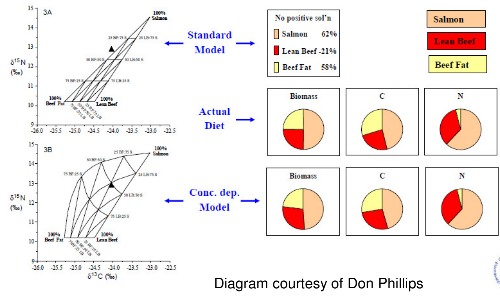
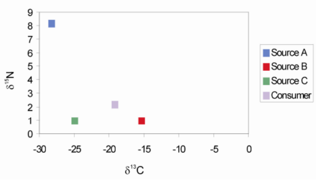
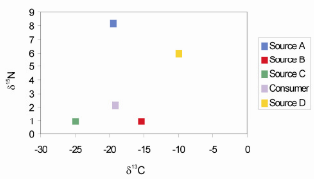


Diagram courtesy of Don Phillips

Determined and Underdetermined Systems





2 Isotopes and 3 Sources

Can be solved with an exact solution

2 Isotopes and 4 Sources

Underdetermined system

Cannot be solved for a unique solution

Maybe many thousands of feasible solutions!!!!

Multiple Source Mixing Models

IsoSource – Phillips & Gregg (2003)

- All is not lost
- Uses iterative approach, using every possible combination of sources in user defined steps.
- Creates a *DISTRIBUTION* of *POSSIBLE* feasible solutions to the mass balance equations.

$A_nB_nC_nD_n$

$A_nB_nC_nD_n$

$A_nB_nC_nD_n$

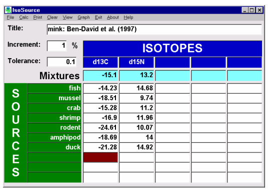
Source Proportions

Calculate Isotopic Signature of Mixture

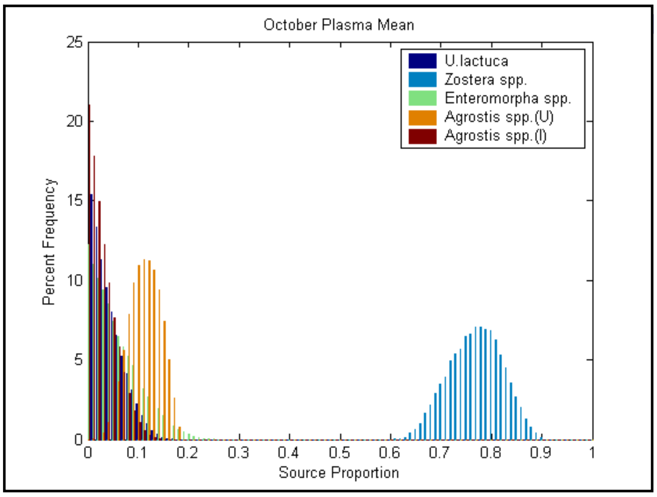
Does it match the actual signature?

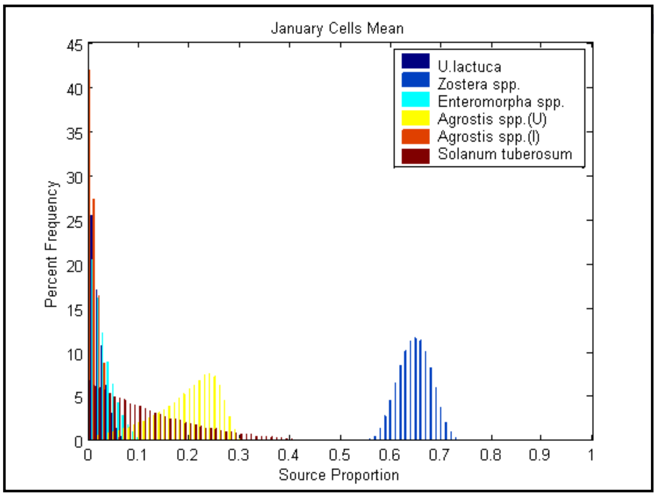
No → Discard

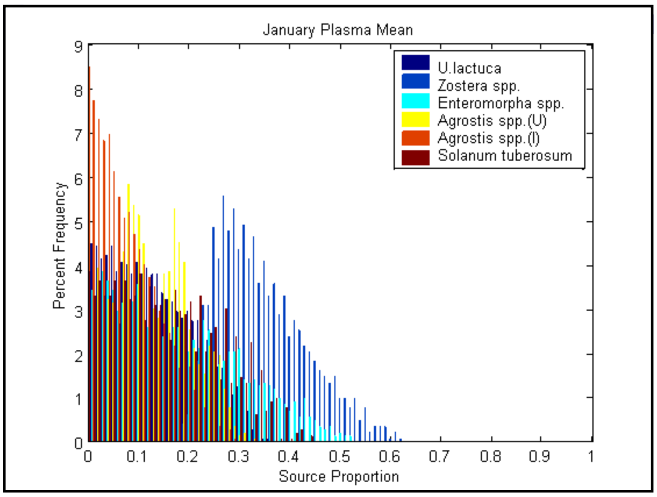
Yes → Add to distribution of possible solutions

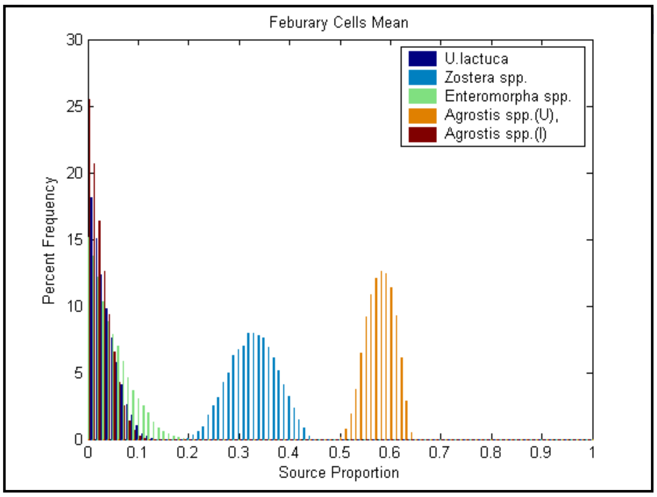


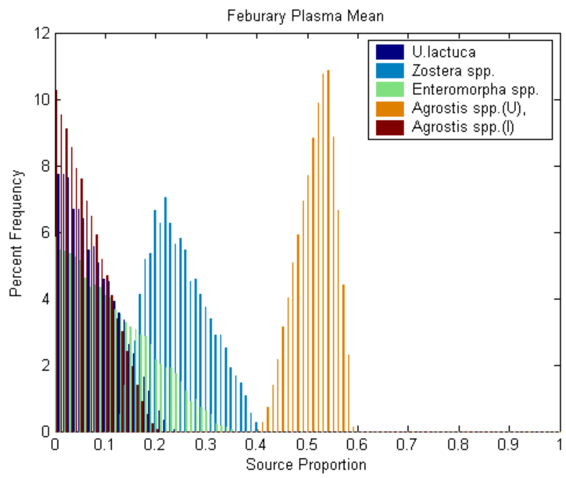
Light-bellied Brent Geese Wintering in Ireland

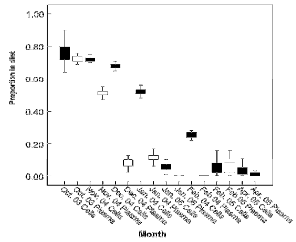

October Plasma Mean
U.lactuca
Zostera spp.
Enteromorpha spp.
Agrostis spp.(U)
Agrostis spp.(I)
Percent Frequency
Source Proportion


January Cells Mean
U.lactuca
Zostera spp.
Enteromorpha spp.
Agrostis spp.(U)
Agrostis spp.(I)
Solanum tuberosum
Percent Frequency
Source Proportion

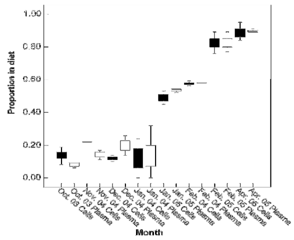

January Plasma Mean
U.lactuca
Zostera spp.
Enteromorpha spp.
Agrostis spp.(U)
Agrostis spp.(I)
Solanum tuberosum
Percent Frequency
Source Proportion


Feburary Cells Mean
U.lactuca
Zostera spp.
Enteromorpha spp.
Agrostis spp.(U),
Agrostis spp.(I)
Percent Frequency
Source Proportion





Zostera



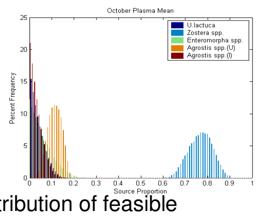
Grass

Problems with Isosource

Cannot Include Variation

Cannot Include Conc Dependance

Output is not a
Probability Density Function



MUST consider the whole of the distribution of feasible soultions.

ACTUAL solution is somewhere within the distribution, but we have NOT IDEA where!!!

Makes further analysis very difficult

Can be done will very large mixed effect GLMS – Statisticians HATE this

Summary So Far

Determined systems

Number of Source <= number of isotopes +1

Simple model – Linear Mixing Model (Phillips 2001. 111 Cites)

With Variation – IsoError (Phillips & Gregg 2001. 211 Cites)

Concentration Dependence – IsoConc (Phillips & Kock 2001. 133 Cites)

Underdetermined systems

Number of Source > number of isotopes +1

Isosource (Phillips & Gregg 2003. 217 Cites)

SOURCE (Lubetkin & Simenstad 2004. 11 Cites)

Bayesian Mixing Models – A New Hope?

MixSIR (Moore & Semmens 2008)

SIAR (Jackson, Inger, Bearhop & Parnell 2008)

- Will work with underdetermined systems
- Can incorporate multiple sources of variation and uncertainty with the Bayesian Model Framework

SISUS

- Bayesian version of Isosource.
- Cannot incorporate variability

Differences between MixSIR & SIAR

- Very similar models. Both produce probability density functions describing feasible proportions of sources
- SIAR has an additional residual error term to account for unquantifiable or unknown variation
- SIAR works on populations or individuals
- SIAR can incorporate concentration dependence
- Model fit;
  - SIAR -Markov chain Monte Carlo (MCMC)
  - MixSIR – Sampling Importance Resampling (SIR)
- SIAR runs in R
- MixSIR runs in Matlab.

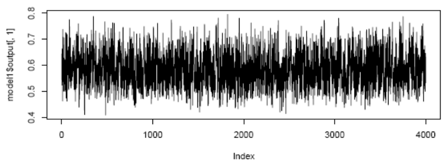
SIAR Stable Isotope Analysis in R

- Consumer Data
  - Multiple individuals with grouping variable
    - Default
  - Single individuals -SIARSOLO
- Source Data
  - Mean and SD – Any number of sources
- Trophic Enrichment Factors (TEFs)
  - Mean and SD
- Elemental Concentrations
  - Mean and SD (Optional)

SIAR Stable Isotope Analysis in R

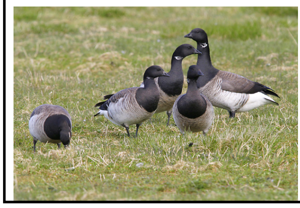
$$P(A|B) = \frac{P(B|A)P(A)}{P(B)}$$

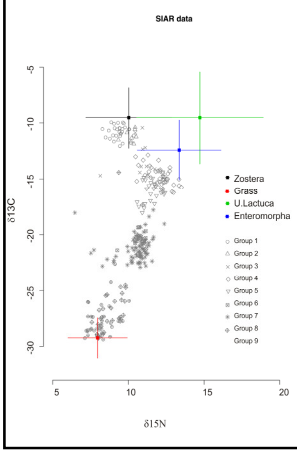
Rev. T. Bayes

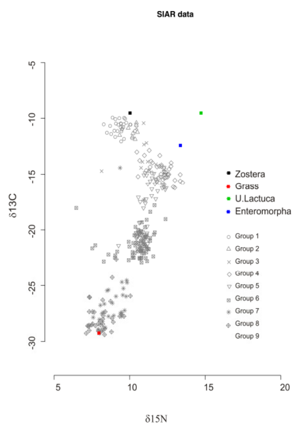


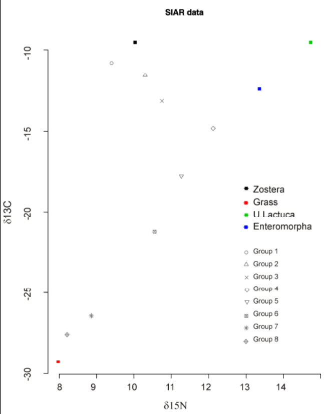
Light-bellied Brent Geese

- Wintering in Ireland
- Blood plasma samples
- 281 Individuals over 8 months
- How does diet vary with time and other factors





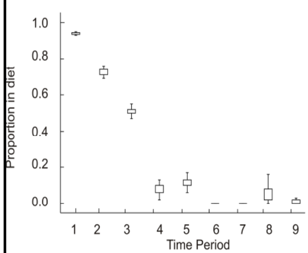


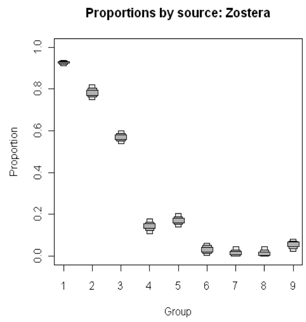


Use of means results in huge loss of information

Isosource

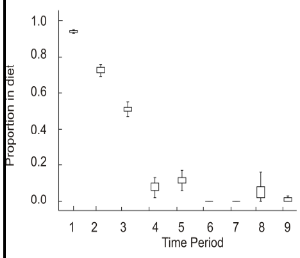
SIAR
No Variation

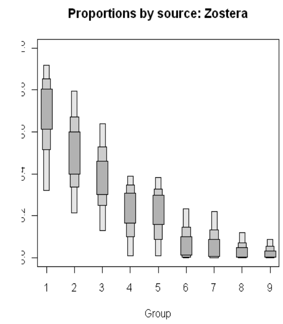




Isosource

SIAR
Variation in Sources

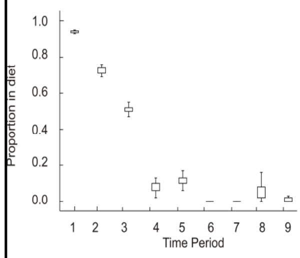


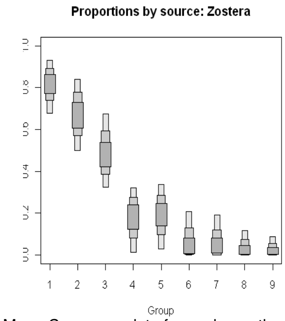


Mean Consumer data for each month

Isosource

SIAR
Variation in TEFs

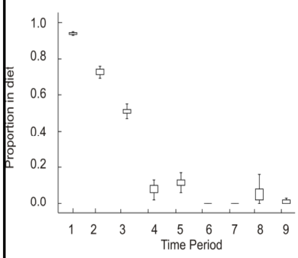


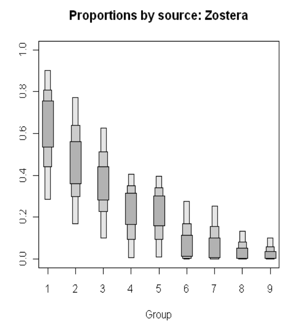


Mean Consumer data for each month

Isosource

SIAR
Variation in Sources & TEFs

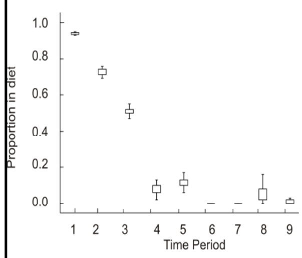


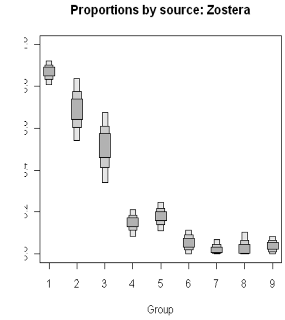


Mean Consumer data for each month

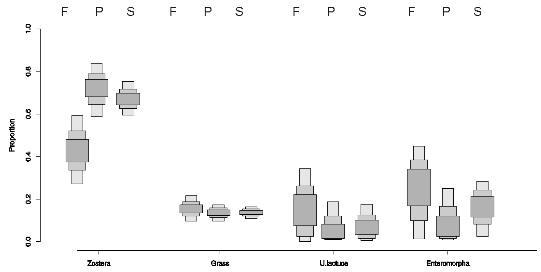
Isosource

SIAR
Variation in Sources & TEFs
and between individuals





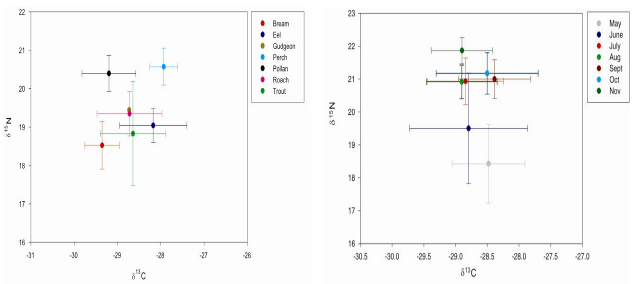
Comparison between social groups



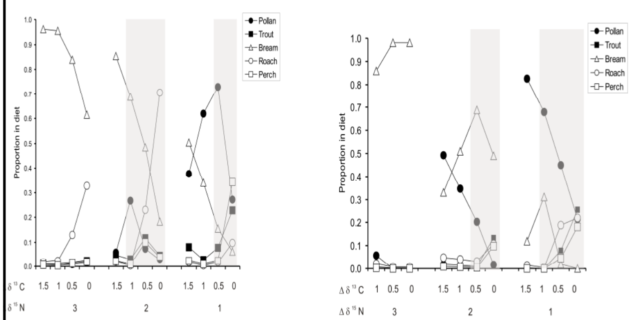
- •Outputs ARE true probability density functions
- •Suitable for further analysis
- •Ideally in a full Bayesian framework
- •Can be done with standard techniques

River Lamprey

River Lamprey



River Lamprey – Different TEFs in Isosource



River Lamprey

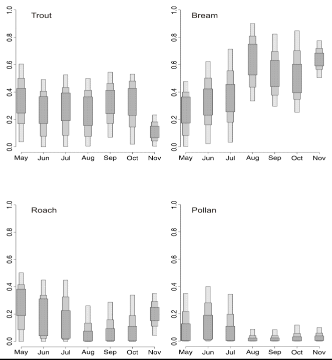
SIAR output

Multiple Individuals / Month

Multiple Tissues / Individuals

Mean TEFs from literature

Large S.D.s on TEFs



Future Developments

- SIAR V5
- New formulation for TEFs
- Normal distribution
  - Mean drawn from uniform distribution
  - Variance normal drawn from normal
  - Fitted based on the available data
- Beyond V5
  - Built up Bayesian GLM
-

Summary

- Bayesian mixing models
  - Can include many forms of variation
  - Dietary proportions are true PDFs
    - Suitable for further analysis
  - SIAR
    - Works on individuals and populations
    - Contains residual error term
    - Can incorporate Elemental Concentrations
    - Runs in R
      - Open Source / Can be customised

Acknowledgements

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- Andrew Parnell
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