Online Supplemental Material

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Effects of sex, tissue type, and dietary components on stable isotope discrimination factors ($\Delta^{15}N$ and $\Delta^{13}C$) in mammalian omnivores

Equations for calculating the fraction of carbon routed to body tissues from dietary lipids and carbohydrates.

Tables S1-S6

Supplemental Information: Equations to determine the fraction of carbon routed to body tissues from dietary lipids and carbohydrates.

Equations

1. Start with the % by weight of lipids and carbohydrates and calculate the decimal % dry weight for each.

$$Decimal\ \%\ dry\ weight\ of\ lipid = \frac{\%\ by\ weight\ of\ lipid}{(\%\ by\ weight\ of\ lipid) + (\%\ by\ weight\ of\ carbohydrates)} \quad (1)$$

The % by weight of lipid and carbohydrate values are in Table 1 in the manuscript.

2. Calculate the grams of carbon from lipid in 100 grams of dry weight:

$$g\ C\ from\ lipid\ per\ 100\ g\ dry\ weight = [(decimal\ \%\ dry\ weight\ of\ lipid)(100)]x\ (0.75)\ (2)$$

The 0.75 value comes from [1].

3. Do the same for grams of carbon from carbohydrates per 100 grams of dry weight:

$$g\ C\ from\ carbohydrates\ per\ 100\ g\ dry\ weight = [(decimal\ \%\ dry\ weight\ of\ carbs)(100)]x\ (0.45)\ (3)$$

The 0.45 value comes from [1].

4. Calculate the fraction of lipid derived carbon and the fraction of carbohydrate derived carbon:

$$\%\ carbon\ from\ lipid = \frac{lipid\ per\ 100\ g\ dry\ weight}{(lipid\ per\ 100\ g\ dry\ weight) + (carbohydrate\ per\ 100\ g\ dry\ weight)} \quad (4)$$

5. Determine the $\delta^{13}C$ value for the entire dietary energy budget:

$$\partial^{13}C_{energy} = [(\partial^{13}C_{lipid})(\%\ C\ from\ lipid)] + [(\partial^{13}C_{carbohydrate})(\%\ C\ from\ carbohydrate)] \quad (5)$$

The $\delta^{13}C$ values for lipids and carbohydrates are in Table 1 in manuscript.

6. Calculate 25% of the $\delta^{13}C_{energy}$ value to account for the amount of dietary carbon incorporated into the body from energy (lipids and carbohydrates). This is modified from MacAvoy et al. 2005 [2].

$$\partial^{13}C\ of\ dietary\ energy\ incorporated\ into\ body\ tissues = (\partial^{13}C_{energy})(0.25)\ (6)$$

7. This value would be added to the $\delta^{13}C$ value that reflects the proportion of carbon incorporated into the body from the protein components of the diet [($\delta^{13}C_{protein}$)(0.75)] to arrive at an adjusted bulk diet $\delta^{13}C$ value that accounts for the routing of carbon from dietary protein, carbohydrate, and lipid into the consumer carbon pool.

Example using the values from the wheat gluten diet in this study

1. Start with the % by weight lipids and carbohydrates and calculate the decimal % dry weight for each.

$$Decimal\ \%\ dry\ weight\ of\ lipid = \frac{8.20}{(8.20) + (53.40)} = 0.1331 \ \ (1)$$

$$Decimal\ \%\ dry\ weight\ of\ lipid = \frac{53.40}{(8.20) + (53.40)} = 0.0.8669 \ \ (1)$$

The % by weight of lipid and carbohydrate values are in Table 1 in the manuscript.

2. Calculate the grams of carbon from lipid in 100 grams of dry weight:

$$g\ C\ from\ lipid\ per\ 100\ g\ dry\ weight = [(0.1331)(100)]x\ (0.75) = 9.98\ (2)$$

The 0.75 value comes from [1].

3. Do the same for grams of carbon from carbohydrates per 100 grams of dry weight:

$$g\ C\ from\ carbohydrates\ per\ 100\ g\ dry\ weight = [(0.8669)(100)]x\ (0.45) = 39.01\ (3)$$

The 0.45 value comes from [1].

4. Calculate the fraction of lipid derived carbon and the fraction of carbohydrate derived carbon:

$$\%\ carbon\ from\ lipid = \frac{9.98}{(9.98) + (39.01)} = 0.20 \ \ (4)$$

$$\%\ carbon\ from\ carbohydrates = \frac{39.01}{(9.98) + (39.01)} = 0.80 \ \ (4)$$

5. Determine the $\delta^{13}C$ value for the entire dietary energy budget:

$$\partial^{13}C_{energy} = [(-28.5‰)(0.20)] + [(-24.9‰)(0.80)] = -25.62‰ \ \ (5)$$

6. Calculate 25% of the $\delta^{13}C_{energy}$ value to account for the amount of dietary carbon incorporated into the body from energy (lipids and carbohydrates). This is modified from MacAvoy et al. 2005 [2].

$$\partial^{13}C\ of\ dietary\ energy\ incorporated\ into\ body\ tissues = (-25.62‰)(0.25) = -6.405‰\ (6)$$

7. This value (-6.045‰) would be added to the $\delta^{13}C$ value that reflects the proportion of carbon incorporated into the body from the protein components of the diet to arrive at an adjusted bulk diet $\delta^{13}C$ value that accounts for the routing of carbon from dietary protein, carbohydrate, and lipid into the consumer carbon pool.
To calculate that value for the wheat gluten diet:

$(\delta^{13}C_{protein})(0.75)$ = -19.35‰

-6.045‰ + -19.35‰ = -25.8‰

Conclusion:

The adjusted bulk diet $\delta^{13}C$ value used in the manuscript for the wheat diet, following the protocol described in MacAvoy et al. 2005 [2], was -25.6‰, only 0.2‰ larger than the value calculated above. The same was true for all diets; the $\delta^{13}C$ value for the adjusted bulk diet accounting for lipid routing and using the above equations would have resulted in $\delta^{13}C$ values that were only 0.2‰ larger than what were reported and used in the manuscript, an extremely minimal amount. As accounting for lipid routing this way has not been described or used before, we accounted for carbohydrate routing only (as per MacAvoy et al. 2005 [2]), but are including the above calculations for reference.

Table S1. Essential amino acid contents of the four experimental diets and the minimum amino acid requirements for maintenance and growth of an adult rat (measured in g/kg of diet)[1] [3, 4]. Amino acids marked with an asterisk are only essential in certain cases. All other dietary components are listed in Table 1.

Amino acid	Diet Type				Maintenance	Growth
	Wheat gluten	Fish	Wheat/ fish	Wheat/fish/ casein/egg		
Arginine*	9.6	16.1	11.8	12.6	NA	4.3
Cystine*	3.9	3.0	3.7	4.0	1.2	NA
Histidine*	4.8	5.4	5.1	5.4	0.7 to 0.8	2.8
Isoleucine	9.6	10.9	10.3	11.3	2.4 to 3.1	6.2
Leucine	15.6	17.4	16.6	18.4	1.0 to 1.8	10.7
Lysine	9.2	18.1	9.7	12.2	0.5 to 1.1	9.2
Methionine	6.9	8.7	6.9	8.2	1.7	NA
Phenylalanine	11.1	9.1	10.8	11.5	0.8	NA
Threonine	6.4	10.3	7.0	8.7	1.6 to 1.8	6.2
Tryptophan	2.4	2.7	2.6	2.9	0.4 to 0.5	2.0
Tyrosine*	6.3	7.3	6.8	8.1	0.6	NA
Valine	9.6	12.1	10.6	12.7	1.5 to 2.3	7.4
Cystine and Methionine Together[2]	10.8	11.7	10.6	12.2	2.3	9.8

[1][3, 4]
[2]Cystine may supply up to 50% of the methionine plus cystine requirement on a weight basis [3].

Table S2. Carbon trophic discrimination factors ($\Delta^{13}C$, reported in ‰ ± SD) between bulk diet and tissues for rats held on 4 experimental diets (see Table 1 for diet details). Tissues are listed in order of isotope turnover time from fastest (liver) to slowest (fur) [5]. Significant differences in $\Delta^{13}C$ values between sexes held on the same diets are denoted by an asterisk (*). N = 3 females and 3 males per diet. The $\Delta^{13}C$ values reported in the manuscript are based on adjusted dietary $\delta^{13}C$ values that consider differential routing of carbon from dietary protein and carbohydrates sources (see Methods).

Tissue	Wheat Diet		Fish Diet	
	Female $\Delta^{13}C$	Male $\Delta^{13}C$	Female $\Delta^{13}C$	Male $\Delta^{13}C$
Liver	+1.0±0.3	+0.8±0.8	-0.7±0.1	-1.2±0.4
Serum	+1.3±0.1	+1.1±0.0	-1.0±0.2	-0.8±0.2
Kidney	+1.0±0.1	+1.1±0.1	-0.8±0.2	-0.8±0.1
RBC	+1.0±0.1	+0.9±0.1	-0.9±0.1	-1.0±0.1
Muscle	+1.4±0.1	+1.4±0.2	-0.4±0.1	-0.4±0.1
Fur	+3.2±0.2	+2.9±0.1	+0.6±0.1	+0.4±0.2
	Wheat /Fish Diet		Wheat/Fish/Casein/Egg White Diet	
Liver	+1.1±0.5	+0.4±1.1	-0.7±0.0	-0.6±0.1
Serum	+1.9±0.1	+1.9±0.2	-1.7±0.0	-1.6±0.1
Kidney	+1.9±0.2	+1.5±0.3	-1.4±0.1	-1.6±0.1
RBC	+2.0±0.1	+1.6±0.3	-2.2±0.1	-2.3±0.1
Muscle	+2.3±0.1	+1.8±0.4	-1.4±0.0	-1.3±0.1
Fur	+3.6±0.1*	+3.1±0.2*	-0.5±0.1*	-0.6±0.1*

Table S3. Stable isotope values ($\delta^{15}N$ and $\delta^{13}C$, reported in ‰ ± SD) from rat tissues held on four experimental diets for 276-278 days. Significant differences ($p \leq 0.05$) in $\delta^{15}N$ and $\delta^{13}C$ values (as shown by t-tests, $-2.4 \leq t \leq 11.7$, all n=3, all df = 4, $0.01 > p \leq 0.90$) between sexes held on the same diets are denoted by an asterisk (*).

Tissue	Wheat Diet				Fish Diet			
	Female		Male		Female		Male	
	$\delta^{15}N$	$\delta^{13}C$	$\delta^{15}N$	$\delta^{13}C$	$\delta^{15}N$	$\delta^{13}C$	$\delta^{15}N$	$\delta^{13}C$
Liver	8.0 ± 0.2	-24.3 ± 0.0	7.8 ± 0.5	-24.5 ± 0.1	15.7 ± 0.1*	-16.9 ± 0.1	15.1 ± 0.1*	-17.4 ± 0.1
Serum	8.5 ± 0.1*	-24.0 ± 0.1	8.3 ± 0.0*	-24.2 ± 0.0	16.2 ± 0.1*	-17.2 ± 0.1	15.7 ± 0.2*	-17.0 ± 0.1
Kidney	7.6 ± 0.0*	-24.3 ± 0.1	7.0 ± 0.1*	-24.2 ± 0.1	15.7 ± 0.1*	-17.0 ± 0.1	15.0 ± 0.1*	-17.0 ± 0.0
RBC	7.0 ± 0.1*	-24.4 ± 0.1	6.5 ± 0.1*	-24.3 ± 0.1	15.5 ± 0.1*	-17.1 ± 0.0	15.0 ± 0.1*	-17.2 ± 0.0
Muscle	7.3 ± 0.0*	-23.9 ± 0.0	6.8 ± 0.1*	-23.8 ± 0.1	15.7 ± 0.1*	-16.6 ± 0.0	15.0 ± 0.0*	-16.6 ± 0.0
Fur	7.3 ± 0.1	-22.1 ± 0.1	7.1 ± 0.0	-22.4 ± 0.1	16.4 ± 0.1	-15.6 ± 0.1	16.4 ± 0.1	-15.6 ± 0.1
	Wheat/Fish Diet				Wheat/Fish/Casein/Egg Diet			
Liver	10.5 ± 0.1*	-23.4 ± 0.3	10.0 ± 0.1*	-24.1 ± 0.6	10.6 ± 0.1*	-17.4 ± 0.0	10.1 ± 0.0*	-17.3 ± 0.1
Serum	11.0 ± 0.1*	-22.6 ± 0.1	10.6 ± 0.1*	-22.6 ± 0.1	11.2 ± 0.1*	-18.4 ± 0.0	10.6 ± 0.0*	-18.3 ± 0.1
Kidney	10.1 ± 0.1*	-22.6 ± 0.1	9.4 ± 0.1*	-23.0 ± 0.2	9.9 ± 0.1*	-18.2 ± 0.1	9.3 ± 0.0*	-18.3 ± 0.1
RBC	9.6 ± 0.0*	-22.5 ± 0.1	9.2 ± 0.0*	-22.9 ± 0.2	9.8 ± 0.1	-19.0 ± 0.1	9.4 ± 0.0	-19.0 ± 0.0
Muscle	10.1 ± 0.1*	-22.2 ± 0.1	9.3 ± 0.2*	-22.7 ± 0.0	9.9 ± 0.2	-18.1 ± 0.0	9.5 ± 0.1	-18.1 ± 0.0
Fur	10.0 ± 0.1	-20.9 ± 0.1*	9.7 ± 0.1	-21.4 ± 0.1*	10.4 ± 0.1	-17.2 ± 0.0*	10.2 ± 0.0	-17.4 ± 0.0*

Table S4. F-values from ANOVAs and *p*-values from Post-hoc Tukey's Honestly Significant Difference tests demonstrating the differences in the $\Delta^{15}N$ and $\Delta^{13}C$ values among tissue types from rats held on 1 of 4 diets: Wheat (wheat gluten, beet sugar, cottonseed oil (sugar and oil from C_3 plants)), Fish (fish meal, cane sugar, corn oil (sugar and oil from C_4 plants)), Wheat/Fish (wheat gluten, fish meal, beet sugar, cottonseed oil), and Wheat/Fish/Egg/Milk (wheat gluten, fish meal, cow casein, chicken egg whites). All ANOVA *p*-values < 0.05.

Diet	Sex	Isotope	$F_{5,12}$	Tissues	Tukey *p*-values				
Wheat	Female	N	88.4		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.00				
				Kidney	0.00	0.00			
				RBC	0.00	0.00	0.00		
				Muscle	0.00	0.00	0.05	0.01	
				Fur	0.00	0.00	0.05	0.01	1.00
Wheat	Female	C	90.0		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.29				
				Kidney	1.00	0.20			
				RBC	1.00	0.42	0.99		
				Muscle	0.00	0.98	0.07	0.16	
				Fur	0.00	0.00	0.00	0.00	0.00
Wheat	Male	N	46.7		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.03				
				Kidney	0.00	0.00			
				RBC	0.00	0.00	0.04		
				Muscle	0.00	0.00	0.81	0.30	
				Fur	0.00	0.00	0.97	0.01	0.40
Wheat	Male	C	16.4		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.79				
				Kidney	0.81	1.00			
				RBC	0.99	0.98	0.99		
				Muscle	0.22	0.85	0.83	0.47	
				Fur	0.00	0.00	0.00	0.00	0.00
Fish	Female	N	11.2		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.05				
				Kidney	1.00	0.05			
				RBC	0.67	0.01	0.74		
				Muscle	1.00	0.04	0.00	0.84	
				Fur	0.01	0.77	0.00	0.00	0.00
Fish	Female	C	58.4		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.26				
				Kidney	0.96	0.66			
				RBC	0.51	0.99	0.91		
				Muscle	0.16	0.00	0.04	0.01	
				Fur	0.00	0.00	0.00	0.00	0.00
Fish	Male	N	21.7		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.00				
				Kidney	1.00	0.00			
				RBC	1.00	0.00	1.00		
				Muscle	1.00	0.00	1.00	1.00	
				Fur	0.00	0.33	0.00	0.00	0.00

Table S4. F-values from ANOVAs and *p*-values from Post-hoc Tukey's Honestly Significant Difference tests demonstrating the differences in the $\Delta^{15}N$ and $\Delta^{13}C$ values among tissue types from rats held on 1 of 4 diets: Wheat (wheat gluten, beet sugar, cottonseed oil (sugar and oil from C_3 plants)), Fish (fish meal, cane sugar, corn oil (sugar and oil from C_4 plants)), Wheat/Fish (wheat gluten, fish meal, beet sugar, cottonseed oil), and Wheat/Fish/Egg/Milk (wheat gluten, fish meal, cow casein, chicken egg whites). All ANOVA *p*-values < 0.05.

Diet	Sex	Isotope	$F_{5,12}$	Tissues	Tukey *p*-values				
Fish	Male	C	30.6		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.23				
				Kidney	0.29	1.00			
				RBC	0.85	0.80	0.88		
				Muscle	0.00	0.15	0.12	0.02	
				Fur	0.00	0.00	0.00	0.00	0.00
Wheat/Fish	Female	N	29.7		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.02				
				Kidney	0.04	0.00			
				RBC	0.00	0.00	0.05		
				Muscle	0.02	0.00	0.00	0.08	
				Fur	0.01	0.00	0.95	0.19	0.99
Wheat/Fish	Female	C	39.6		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.01				
				Kidney	0.02	1.00			
				RBC	0.01	1.00	0.96		
				Muscle	0.00	0.53	0.32	0.75	
				Fur	0.00	0.00	0.00	0.00	0.00
Wheat/Fish	Male	N	76.0		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.00				
				Kidney	0.00	0.00			
				RBC	0.00	0.00	0.47		
				Muscle	0.00	0.00	0.97	0.87	
				Fur	0.05	0.00	0.02	0.00	0.01
Wheat/Fish	Male	C	8.5		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.04				
				Kidney	0.16	0.94			
				RBC	0.13	0.97	1.00		
				Muscle	0.05	1.00	0.98	0.99	
				Fur	0.00	0.11	0.02	0.03	0.07
Wheat/Fish/Egg/Milk	Female	N	21.0		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.02				
				Kidney	0.02	0.00			
				RBC	0.01	0.00	0.98		
				Muscle	0.02	0.00	1.00	0.98	
				Fur	0.95	0.00	0.10	0.03	0.10
Wheat/Fish/Egg/Milk	Female	C	310.3		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.00				
				Kidney	0.00	0.00			
				RBC	0.00	0.00	0.00		
				Muscle	0.00	0.00	0.96	0.00	
				Fur	0.00	0.00	0.01	0.00	0.00

Table S4. F-values from ANOVAs and *p*-values from Post-hoc Tukey's Honestly Significant Difference tests demonstrating the differences in the $\Delta^{15}N$ and $\Delta^{13}C$ values among tissue types from rats held on 1 of 4 diets: Wheat (wheat gluten, beet sugar, cottonseed oil (sugar and oil from C_3 plants)), Fish (fish meal, cane sugar, corn oil (sugar and oil from C_4 plants)), Wheat/Fish (wheat gluten, fish meal, beet sugar, cottonseed oil), and Wheat/Fish/Egg/Milk (wheat gluten, fish meal, cow casein, chicken egg whites). All ANOVA *p*-values < 0.05.

Diet	Sex	Isotope	$F_{5,12}$	Tissues					
Wheat/Fish/Egg/Milk	Male	N	97.6		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.00				
				Kidney	0.00	0.00			
				RBC	0.00	0.00	0.47		
				Muscle	0.00	0.00	0.16	0.96	
				Fur	0.57	0.00	0.00	0.00	0.00
Wheat/Fish/Egg/Milk	Male	C	198.2		Liver	Serum	Kidney	RBC	Muscle
				Serum	0.00				
				Kidney	0.00	1.00			
				RBC	0.00	0.01	0.00		
				Muscle	0.00	0.03	0.02	0.00	
				Fur	0.96	0.00	0.00	0.00	0.00

Table S5. The *p*-values from ANOVA and Tukey post-hoc tests for differences in discrimination factors ($\Delta^{15}N$ and $\Delta^{13}C$) between diet types for 6 tissues from rats held on 1 of 4 diets: Plant (wheat gluten, beet sugar, cottonseed oil (sugar and oil from C_3 plants)), Fish (fish meal, cane sugar, corn oil (sugar and oil from C_4 plants)), Plant/Fish (wheat gluten, fish meal, beet sugar, cottonseed oil), and Plant/Fish/Egg/Milk (wheat gluten, fish meal, cow casein, chicken egg whites, cane sugar, corn oil).

Tissue	Sex	Isotope	$F_{3,8}$	ANOVA *p*-value	Diet	Tukey *p*-values		
Fur	Female	N	68.1	0.00		Wheat	Fish	Wheat/Fish
					Fish	0.00		
					Wheat/Fish	0.05	0.00	
					Wheat/Fish/Egg/Milk	0.01	0.00	0.67
						Wheat	Fish	Wheat/Fish
Fur	Female	C	246.4	0.00	Fish	0.00		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.00	0.56	0.00
						Wheat	Fish	Wheat/Fish
Fur	Male	N	42.7	0.00	Fish	0.00		
					Wheat/Fish	0.06	0.00	
					Wheat/Fish/Egg/Milk	0.01	0.00	0.71
						Wheat	Fish	Wheat/Fish
Fur	Male	C	140.2	0.00	Fish	0.00		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.00	0.50	0.00
						Wheat	Fish	Wheat/Fish
Kidney	Female	N	9.8	0.01	Fish	0.03		
					Wheat/Fish	0.66	0.15	
					Wheat/Fish/Egg/Milk	0.40	0.00	0.08
						Wheat	Fish	Wheat/Fish
Kidney	Female	C	92.0	0.00	Fish	0.02		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.69	0.00	0.00
						Wheat	Fish	Wheat/Fish
Kidney	Male	N	27.0	0.00	Fish	0.00		
					Wheat/Fish	0.36	0.02	
					Wheat/Fish/Egg/Milk	0.07	0.00	0.01
						Wheat	Fish	Wheat/Fish
Kidney	Male	C	34.0	0.00	Fish	0.01		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.52	0.07	0.00
						Wheat	Fish	Wheat/Fish
Liver	Female	N	3.5	0.07	Fish	0.25		
					Wheat/Fish	0.23	1.00	
					Wheat/Fish/Egg/Milk	0.98	0.15	0.13
						Wheat	Fish	Wheat/Fish
Liver	Female	C	11.9	0.00	Fish	0.33		
					Wheat/Fish	0.14	0.01	
					Wheat/Fish/Egg/Milk	0.03	0.00	0.70
						Wheat	Fish	Wheat/Fish
Liver	Male	N	2.4	0.15	Fish	0.42		
					Wheat/Fish	1.00	0.51	
					Wheat/Fish/Egg/Milk	0.20	0.93	0.25
						Wheat	Fish	Wheat/Fish
Liver	Male	C	3.4	0.07	Fish	0.69		
					Wheat/Fish	1.00	0.66	
					Wheat/Fish/Egg/Milk	0.23	0.05	0.27

Table S5. The *p*-values from ANOVA and Tukey post-hoc tests for differences in discrimination factors ($\Delta^{15}N$ and $\Delta^{13}C$) between diet types for 6 tissues from rats held on 1 of 4 diets: Plant (wheat gluten, beet sugar, cottonseed oil (sugar and oil from C_3 plants)), Fish (fish meal, cane sugar, corn oil (sugar and oil from C_4 plants)), Plant/Fish (wheat gluten, fish meal, beet sugar, cottonseed oil), and Plant/Fish/Egg/Milk (wheat gluten, fish meal, cow casein, chicken egg whites, cane sugar, corn oil).

Tissue	Sex	Isotope	$F_{3,8}$	ANOVA *p*-value	Diet	Tukey *p*-values		
						Wheat	Fish	Wheat/Fish
Muscle	Female	N	9.4	0.01	Fish	0.01		
					Wheat/Fish	0.16	0.21	
					Wheat/Fish/Egg/Milk	1.00	0.01	0.18
						Wheat	Fish	Wheat/Fish
Muscle	Female	C	259.2	0.00	Fish	0.00		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.03	0.03	0.00
						Wheat	Fish	Wheat/Fish
Muscle	Male	N	20.7	0.00	Fish	0.00		
					Wheat/Fish	0.10	0.01	
					Wheat/Fish/Egg/Milk	0.81	0.00	0.34
						Wheat	Fish	Wheat/Fish
Muscle	Male	C	19.1	0.00	Fish	0.06		
					Wheat/Fish	0.02	0.00	
					Wheat/Fish/Egg/Milk	0.44	0.48	0.00
						Wheat	Fish	Wheat/Fish
Serum	Female	N	1.69	0.25	Fish	0.34		
					Wheat/Fish	0.24	0.99	
					Wheat/Fish/Egg/Milk	0.61	0.95	0.85
						Wheat	Fish	Wheat/Fish
Serum	Female	C	103.0	0.00	Fish	0.00		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.02	0.02	0.00
						Wheat	Fish	Wheat/Fish
Serum	Male	N	3.7	0.06	Fish	0.93		
					Wheat/Fish	0.91	0.62	
					Wheat/Fish/Egg/Milk	0.13	0.29	0.06
						Wheat	Fish	Wheat/Fish
Serum	Male	C	79.9	0.00	Fish	0.00		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.39	0.03	0.00
						Wheat	Fish	Wheat/Fish
RBC	Female	N	28.3	0.00	Fish	0.00		
					Wheat/Fish	0.03	0.00	
					Wheat/Fish/Egg/Milk	0.21	0.00	0.50
						Wheat	Fish	Wheat/Fish
RBC	Female	C	324.2	0.00	Fish	0.00		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.00	0.37	0.00
						Wheat	Fish	Wheat/Fish
RBC	Male	N	45.5	0.00	Fish	0.00		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.02	0.00	0.41
						Wheat	Fish	Wheat/Fish
RBC	Male	C	81.0	0.00	Fish	0.01		
					Wheat/Fish	0.00	0.00	
					Wheat/Fish/Egg/Milk	0.00	0.86	0.00

Table S6. Nitrogen and carbon trophic discrimination factors ($\Delta^{15}N$ and $\Delta^{13}C$, reported in ‰ ± SD) between diet (adjusted diet for C; see methods) and tissues for all rats (females and males together) held on 4 experimental diets (see Table 1 for diet details). Tissues are listed in order of isotope turnover time from fastest (liver) to slowest (fur)[1]. N = 6 animals (3 females and 3 males) per diet. The $\Delta^{13}C$ values between the $\delta^{13}C_{bulk\ diet}$ and $\delta^{13}C_{rat\ tissues}$ are reported in Table S2.

Tissue	Wheat Diet			Fish Diet	
	All Rats			All Rats	
	$\Delta^{15}N$	$\Delta^{13}C$		$\Delta^{15}N$	$\Delta^{13}C$
Liver	+3.1±0.2	+1.2±0.5		+3.1±0.4	+0.7±0.3
Serum	+3.6±0.2	+1.5±0.1		+3.7±0.3	+0.7±0.2
Kidney	+2.5±0.4	+1.3±0.1		+3.1±0.4	+0.8±0.1
RBC	+1.9±0.3	+1.3±0.1		+3.0±0.3	+0.6±0.1
Muscle	+2.3±0.3	+1.7±0.1		+3.1±0.4	+1.2±0.1
Fur	+2.4±0.2	+3.4±0.2		+3.9±0.3	+2.1±0.1
	Wheat /Fish Diet			Wheat/Fish/Casein/ Egg White Diet	
Liver	+3.2±0.3	+1.5±0.9		+2.9±0.3	+2.2±0.1
Serum	+3.7±0.2	+2.6±0.2		+3.5±0.4	+1.2±0.1
Kidney	+2.7±0.4	+2.4±0.3		+2.2±0.4	+1.3±0.1
RBC	+2.4±0.2	+2.5±0.3		+2.2±0.2	+0.5±0.1
Muscle	+2.6±0.4	+2.7±0.4		+2.3±0.4	+1.4±0.1
Fur	+2.8±0.2	+4.1±0.3		+2.9±0.2	+2.2±0.1

[1] [50]

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