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"The Stoichiometry of Animal Energetics"

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The stoichiometry of animal energetics

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Abstract

Models for energy uptake and use by animals implicitly specify all mass transformations, given homeostasis assumptions. This is because mass fluxes can be written as weighted sums of energy fluxes. This paper presents mass transformations on the basis of the Dynamic Energy Budget model. A theoretical foundation is given for indirect calorimetry, i.e. empirical rules for heat dissipation in terms of oxygen consumption, carbon dioxide production and nitrogen outflux. The foundation implies a method to obtain the chemical potentials of structural biomass and reserves. Conditions on the composition of structural biomass, reserves and nitrogen wastes are derived that ensure a constant respiration quotient. The specific dynamic action turns out to be a simple function of the parameters of the energy budget model. A simple model for drinking is presented that takes into account all mass transformations, as implicitly specified by the energy budget model.

INTRODUCTION

The formulation of models for mass uptake and use by animals are bound to suffer from the huge amount of different compounds relevant to physiological processes, such as feeding, growth, and reproduction. These complexities may be circumvented to some extent by focussing on energy fluxes rather than mass fluxes. For many problems, however, mass transformations are of interest, directly or indirectly, that is, as a means of measuring energy fluxes. In this paper I will argue that the concept of homeostasis implicitly specifies mass fluxes once rules are established for energy fluxes. In other words, such sets of rules do not allow supplementary assumptions on mass fluxes, such as respiration, without creating inconsistensies. I will illustrate this by means of a particular model for energy fluxes, the Dynamic Energy Budget (DEB) model, which specifies uptake and use of energy for maintenance, growth, development and reproduction in animals that have three life stages: embryo, juvenile and adult. These rules also specify, implicitly, mass fluxes such as the use of oxygen and the production of carbon dioxide.

The purpose of this paper is to evaluate mass fluxes and the consequences of energy flux specification for respiration quotients, specific dynamic action, indirect calorimetry and drinking rates. For the latter purpose, we first assume that water, just like oxygen and carbon dioxide, can freely be taken up or excreted; later we specify these processes in more detail. Although the specification of energy fluxes (known as 'powers' in physics), is specific for the DEB model, the coupling between energy and mass fluxes holds for a broad class of models. Since it is not an easy task to delineate this class of models concisely, the DEB model here just serves as an example.

The animal plus the relevant chemical compounds in the environment will constitute our thermodynamic system, so that the system only exchanges heat with its environment. We will consider a set of coupled irreversible reactions and assume that the temperature

and pressure is constant. Since the arguments rest heavily on the concept of homeostasis, that is where the line of reasoning begins.

HOMEOSTASIS

The term 'homeostasis' is used to indicate the ability of most organisms to keep the chemical composition of their body constant, despite changes in the chemical composition of the environment, including their food. I will here use this term in a rather strict sense, but only apply it to the elemental composition of two components of the animal: structural biomass and reserves. These components may differ in elemental composition, but the compositions are here taken to be constant, which is obviously an idealisation.

The microbiological tradition (see e.g. Roels, 1983, Nielsen and Villadsen, 1994) will be followed to combine all compounds in an 'organic' component into a single abstract 'molecule', only accounting for the frequencies of the elements relative to carbon. These 'molecules' can be counted, corresponding with the number of C-atoms, and we refer to these numbers as C-moles.

Note that the partitioning of the animal into components allows for particular deviations from homeostasis for the animal as a whole when the relative sizes of the components can vary. When the number of components is equal to or larger than the number of elements to be followed, it is in principle possible to describe any change in composition. Food related changes in whole animal compositions could be described realistically for microorganisms on the basis of the two components structural biomass and reserves. See Kooijman (1993) for tests with experimental data on bacteria and yeasts.

The DEB model exploits an even stronger homeostasis assumption: the composition of the entire juvenile body does not change when food density does not change. This translates into constraints on reserve dynamics. The embryonic body composition does change because reserves decrease during incubation and the adult female body composition does change because of reserves which accumulate prior to reproduction events. The foetal mode of embryo development is not considered in this paper because the combination of mother plus foetus has to be considered, see Kooijman (1993) for a description of the kinetics.

Size measures for the total body, such as wet weights, dry weights, total carbon, volumes or length measures, comprise both components of the body. One reason to assume homeostasis is that deviations from homeostasis directly affect size measures in a rather problematic way. This is important in relation to rules for energy uptake and use that involve size measures as well as tests of theoretical predictions against experimental data. A deeper reason to assume homeostasis is to avoid inconsistencies in rules for energy uptake and use. In the DEB model growth is assumed to cost a certain fixed amount of energy per unit of structural biovolume and a unit of reserves is assumed to represent a certain fixed amount of energy. When homeostasis would not apply, the logic of such assumptions will be hard to underpin. Many energy budget models will have similar assumptions.

POWERS

The term 'energy' stands for the capacity to do work, but the more precise thermodynamic definition for living systems still awaits further clarification. The chemical potentials (Gibbs free energies) of all compounds are here taken to be constant (i.e. independent of the concentrations of the compounds), which means that the concentrations of the 'minerals', have to be low for this approximation to hold. One problem of the chemical potential of compounds in living cells, is that the cell is highly compartimentalized, which results in a small number of molecules per compartment. (A bacterium with a volume of 0.25 μm^3 and pH 7 has only 15 free protons, for instance; cell compartments have similar volumes.) Moreover, many reactions require involvement of membrane-bound enzymes, which complicates the notion of 'concentration'. A third source of problems is cells' solution to the problem that the chemical potential depends on concentration: polymerization; this also solves their osmotic problems. Reserves, such as carbohydrates, lipids, proteins, are polymers, while the concentration of monomers is usually low.

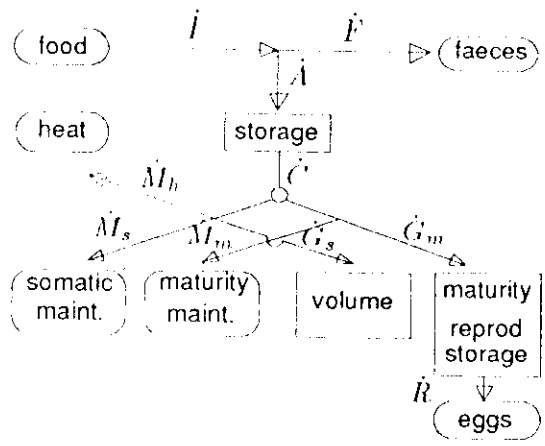
The DEB model describes the fate of energy derived from food upon arrival in the animal, that is when crossing the gut membrane (or outer membrane, in case of ciliates, for instance). The arguments will be illustrated for isomorphs, i.e. animals that do not change shape during growth. The surface area of isomorphs is proportional to l^2, where the state variable 'scaled length' l is defined as $l \equiv (V/V_m)^{1/3}$ where V is the structural biovolume and V_m is the maximum structural biovolume, which is ultimately reached at abundant food availability. The second state variable of the animal is the 'scaled energy reserve density' e, defined as $e \equiv [E]/[E_m]$, where $[E]$ denotes the reserve per unit of structural biovolume (in Gibbs free energy per volume) and $[E_m]$ the maximum value of $[E]$ that can be reached in juveniles and adults after prolonged exposure to abundant food.

The flows of food and faeces are conceived as mass fluxes, food being converted to faeces, under extraction of assimilation energy with a fixed efficiency. This makes ingestion and defecation proportional to assimilation, when the process of digestion is 'instantaneous'.

The assimilation energy is added to the reserves. Energy is used from the reserves at a rate called the catabolic rate, which depends on the state of the animal. A fixed fraction κ of the catabolic power is spent on growth plus (somatic) maintenance, the rest is spent on development plus reproduction. All types of work, such as feeding behaviour, are included in somatic maintenance. Endotherms spend energy to maintain a constant body temperature. This power also plays the role of a maintenance power. In the embryo and juvenile, energy for development is used to maintain the achieved state of maturity and to increase the state of maturity, i.e. differentiation and installation of regulation systems, etc. I assume that this usage of energy does not affect the elemental composition of the animal. The process of maturation is completed at a given size, l_p, when the power that increases the state of maturity is used for reproduction. Feeding is started upon exceeding size l_b at the switch from the embryo to the juvenile state. The various powers are derived in Kooijman (1993) and tested against experimental data. Table 1 gives a summary and assigns symbols to the various powers. For the present purpose it suffices to appreciate that the powers are functions of the states of the animal; other choices will do as well.

The relationships between powers and mass fluxes can be classified into three groups.

Table 1: The powers as specified by the DEB model for an isomorph of scaled size l and scaled reserve density ϵ. Their relationships are given in the diagram, where the rounded boxes indicate sources or sinks. All powers contribute a bit to dissipating heat, but this is not indicated in order to simplify the diagram. The catabolic power is $\dot{C} = \dot{M} + \dot{G}_s + \dot{G}_m + \dot{R}$, with $\dot{M} \equiv \dot{M}_s + \dot{M}_m + \dot{M}_h$. Ectotherms do not heat, therefore $l_h = 0$, where l_h denotes the scaled 'heating length'. Parameters: V_m maximum structural biovolume, g investment ratio, $\dot{m}$ maintenance rate coefficient, $[E_m]$ maximum reserve density, κ partitioning parameter for catabolic power, f scaled functional response: a dimensionless function of food density.



power / $[E_m]V_m\dot{m}g$	embryo $0 < l \leq l_h$	juvenile $l_h < l \leq l_p$	adult $l_p < l < 1$
assimilation, $\dot{A}$	0	$l^2 f$	$l^2 f$
catabolic, $\dot{C}$	$\epsilon l^2 \frac{l+g}{\epsilon+g}$	$\epsilon l^2 \frac{l+g}{\epsilon+g}$	$\epsilon l^2 \frac{l+g}{\epsilon+g}$
somatic maintenance, $\dot{M}_s$	κl^3	κl^3	κl^3
maturity maintenance, $\dot{M}_m$	$(1-\kappa) l^3$	$(1-\kappa) l^3$	$(1-\kappa) l_p^3$
endothermic heating, $\dot{M}_h$	0	$\kappa l^2 l_h$	$\kappa l^2 l_h$
somatic growth, $\dot{G}_s$	$\kappa l^2 g \frac{\epsilon - l}{\epsilon+g}$	$\kappa l^2 \left(g \frac{\epsilon - l}{\epsilon+g} - l_h \right)$	$\kappa l^2 \left(g \frac{\epsilon - l}{\epsilon+g} - l_h \right)$
maturity growth, $\dot{G}_m$	$(1-\kappa) l^2 g \frac{\epsilon - l}{\epsilon+g}$	$(1-\kappa) l^2 g \frac{\epsilon - l}{\epsilon+g}$	0
reproduction, $\dot{R}$	0	0	$(1-\kappa)(l^2 g \frac{\epsilon - l}{\epsilon+g} + l^3 - l_p^3)$

First we have assimilation power, $\dot{A}$, which is tightly coupled to food intake. Then we have anabolic power (somatic growth $\dot{G}_s$), which is tightly coupled to the synthesis of structural biomass. The third group of powers $\dot{D}$ comprises the dissipating powers, i.e. powers that combine a reduction of reserves without any increase in structural biomass. Reproduction power $\dot{R}$ has a special status because reserves of the adult female are converted into reserves of the embryo which have the same composition. The efficiency of this conversion is denoted by q, which means that $(1-q)\dot{R}$ is dissipating and $q\dot{R}$ returns to the reserve component. The dissipating powers of the DEB model thus amount to $\dot{D} = \dot{M} + \dot{G}_m + (1-q)\dot{R}$. Part of each group of powers end up as dissipating heat that leaves the thermodynamic system of animal plus relevant compounds. This process will be studied in more detail in the section on indirect calorimetry.

Table 2: Various nitrogen wastes that animals use (Withers, 1992).

nitrogen waste	formula	solubility (mM)	insects	crustaceans	fish	birds	mammals
ammonia	NH_3	52.1		○	○		
amm. bicarbonate	NH_4HCO_3	1.5		○			
urea	$CO(NH_2)_2$	39.8					○
allantoin	$C_4H_6O_3N_4$	0.015					○
allantoic acid	$C_4H_8O_4N_4$	slight					○
uric acid	$C_5H_4O_3N_4$	0.0015				○	
sodium urate	$C_5H_2O_3N_4Na_2$	0.016	○			○	
potassium urate	$C_5H_2O_3N_4K_2$	slight	○			○	
guanine	$C_4H_5ON_5$	0.0013	○			○	
xanthine	$C_5H_4O_2N_4$	0.068	○			○	
hypoxanthine	$C_5H_4ON_4$	0.021	○			○	
arginine	$C_6H_{14}O_2N_4$	3.4	○			○	

MASS TRANSFORMATIONS

Apart from the 'organic' components (food, faeces, reserves, structural biomass), 'mineral' components are to be distinguished: carbon dioxide, water, oxygen and nitrogen waste. The latter may contain organic compounds, but is nonetheless treated as 'mineral'. The composition of nitrogen waste of embryos is frequently different from that of juveniles and adults, because is has to be stored in the egg. To prevent toxic effects, an insoluble type has to be selected. Frequently used nitrogen wastes are given in table 2.

For simplicity's sake, water in the nitrogen waste (urine) is included in its chemical 'composition', as is done for methane in faeces (which is relevant for mammals). Faeces includes bile and enzymes that are excreted in the gut, since these excretions are tightly coupled to the feeding process. In contrast to the 'static' energy budget tradition, the urine production (the nitrogen waste) is not tightly coupled to the feeding process, because maintenance processes contribute via protein turnover.

The derivations will be given here for a constant chemical composition of food and faeces, starting from the macro-chemical reaction equation that describes all transformations in terms of C-moles. Since energetics become more complex in the sequence embryo, juvenile, adult, we start with the embryo and then introduce feeding and reproduction.

EMBRYONIC MASS TRANSFORMATION

The macro-chemical reaction equation summarizes all chemical transformations as

$$0 = {}_mY_{EE}\,\underset{\text{reserves, }E}{CH_{n_{HE}}O_{n_{OE}}N_{n_{NE}}} + {}_mY_{VE}\,\underset{\text{struc.biovolume, }V}{CH_{n_{HV}}O_{n_{OV}}N_{n_{NV}}} + {}_mY_{AE}\,\underset{\text{water, }A}{H_2O} + {}_mY_{OE}\,\underset{\text{oxygen, }O}{O_2} + {}_mY_{CE}\,\underset{\text{C-dioxide, }C}{CO_2} + {}_mY_{NE}\,\underset{\text{N-waste, }N}{C_{n_{CN}}H_{n_{HN}}O_{n_{ON}}N_{n_{NN}}} \tag{1}$$

The symbols for the relevant compounds are here introduced, as well as the chemical indices $n_{*_1*_2}$, where $*_1$ refers to the element and $*_2$ to the compound. The reaction progresses slowly and irreversibly in time; each compound appears or disappears at its own rate, set by the state of the animal. The stoichiometric coefficients ${}_mY_{*_1*_2}$ are called yield coefficients in the microbiological literature, where subscript m refers to the basis of C-moles. They are defined as ${}_mY_{*_1*_2} \equiv \dot{k}_{*_1}/\dot{k}_{*_2}$, where the $\dot{k}$'s are the appearance ($\dot{k} > 0$) or disappearance ($\dot{k} < 0$) rates. We have used our freedom to multiply the macro-chemical reaction equation by an arbitrary factor to define ${}_mY_{EE} \equiv -1$. The second index of the ${}_mY_{*E}$'s refers to this choice, which gives $\dot{k}_E$ a special role among the $\dot{k}$'s.

The mass balance must hold for each time increment. It can most easely be written as

$$\begin{pmatrix} 1 & 0 & 0 & n_{CN} \\ 0 & 2 & 0 & n_{HN} \\ 2 & 1 & 2 & n_{ON} \\ 0 & 0 & 0 & n_{NN} \end{pmatrix} \begin{pmatrix} {}_mY_{CE} \\ {}_mY_{AE} \\ {}_mY_{OE} \\ {}_mY_{NE} \end{pmatrix} = -\begin{pmatrix} 1 & 1 \\ n_{HV} & n_{HE} \\ n_{OV} & n_{OE} \\ n_{NV} & n_{NE} \end{pmatrix} \begin{pmatrix} {}_mY_{VE} \\ {}_mY_{EE} \end{pmatrix} \tag{2}$$

or

$$\begin{pmatrix} {}_mY_{CE} \\ {}_mY_{AE} \\ {}_mY_{OE} \\ {}_mY_{NE} \end{pmatrix} = -\begin{pmatrix} 1 & 0 & 0 & -\frac{n_{CN}}{n_{NN}} \\ 0 & 2^{-1} & 0 & -\frac{n_{HN}}{2n_{NN}} \\ -1 & -1^{-1} & 2^{-1} & \frac{n}{2n_{NN}} \\ 0 & 0 & 0 & \frac{1}{n_{NN}} \end{pmatrix} \begin{pmatrix} 1 & 1 \\ n_{HV} & n_{HE} \\ n_{OV} & n_{OE} \\ n_{NV} & n_{NE} \end{pmatrix} \begin{pmatrix} {}_mY_{VE} \\ {}_mY_{EE} \end{pmatrix} \tag{3}$$

summarized as

$$\mathbf{Y}_M = -\mathbf{u}\mathbf{n}\mathbf{Y}_D \tag{4}$$

with $n \equiv 4n_{CN} + n_{HN} - 2n_{ON}$. In the microbiological literature the yield on substrate is usually defined to be -1, but this choice is obviously problematic in the case of the embryo, since it does not feed. The index D in $\mathbf{Y}_D$ refers to the DEB model, which specifies the 'organic' yield coefficients, while the 'mineral' yield coefficients $\mathbf{Y}_M$ follow from the conservation law of mass. This will be obvious from the analysis of mass fluxes.

The 'mineral' fluxes follow from the 'organic' fluxes, since

$$\dot{\mathbf{k}}_M \equiv \begin{pmatrix} \dot{k}_C & \dot{k}_A & \dot{k}_O & \dot{k}_N \end{pmatrix}^T = \dot{k}_E \mathbf{Y}_M = -\mathbf{u}\mathbf{n}\dot{k}_E \mathbf{Y}_D = -\mathbf{u}\mathbf{n}\dot{\mathbf{k}}_D \tag{5}$$

The 'organic' fluxes follow from the specification of the energy budget model

$$\dot{\mathbf{k}}_D \equiv \begin{pmatrix} \dot{k}_V \\ \dot{k}_E \end{pmatrix} = \dot{\mu}_E^{-1} \begin{pmatrix} 0 & \frac{[\dot{J}_{Am}]}{[\dot{J}_{Vm}]}\frac{1}{\kappa q} \\ -1 & -1 \end{pmatrix} \begin{pmatrix} \dot{D} \\ \dot{G}_s \end{pmatrix} \equiv \dot{\mu}_E^{-1} \mathbf{J} \begin{pmatrix} \dot{D} \\ \dot{G}_s \end{pmatrix} \tag{6}$$

where $\tilde{\mu}_E \equiv [E_m]/[d_{mv}]$ is the chemical potential of the reserves ($[E_m]$ is the maximum reserve density in Gibbs free energy per structural biovolume, $[d_{mv}]$ converts volume to C-moles). The expression for $\dot{k}_E$ follows from the notion that $\dot{C} = \dot{D} + \dot{G}_s$, while $\tilde{\mu}_E^{-1}$ converts the energy flux into a molar flux. The expression for $\dot{k}_V$ follows from the definition of the energy investment ratio $g \equiv \frac{[G]}{\kappa[E_m]}$, where the parameter $[G]$ stands for the amount of free energy that is required to synthesize a unit of structural biovolume. The conversion from growth power to structural biomass flux is therefore given by $\tilde{\mu}_E^{-1}\frac{[d_{mv}]}{[d_{mv}]}\frac{1}{\kappa g} = \frac{[d_{mv}]}{[G]}$, where $[d_{mv}]$ converts structural biovolume into C-moles.

Biovolume yield is given by

$$ {}_mY_{VE} = \dot{k}_V/\dot{k}_E = -\frac{[d_{mv}]}{[d_{mc}]}\frac{1}{c}\frac{e-l}{g+l} \tag{7}$$

Note that mass fluxes can be written as weighted sums of powers, and that the yield coefficients are ratios of mass fluxes. This means that yield coefficients cannot be written as weighted sums of powers. In the microbiological tradition, where normalization on substrate uptake is standard, the experimentist rather than the organism controls the uptake rate (via chemostat settings). The flux in the denominator of the yield is in that case a constant rather than a weighted sum of powers. Consequently, yield coefficients can be written are weighted sums of powers (see e.g. Kooijman, 1993) which gives a focus on yield coefficients, rather than mass fluxes.

The Respiration Quotient (RQ) is of practical interest because it contains information about the relative contributions of protein, carbohydrates and lipids. Its explicit expression is

$$ -{}_mY_{CO} = -{}_mY_{CL}/{}_mY_{OL} = -\dot{k}_C/\dot{k}_O \tag{8}$$

$$ = \frac{1 - n_{NE}\frac{n_{CN}}{n_{NN}} - (1 - n_{NV}\frac{n_{CN}}{n_{NN}})\,{}_mY_{VE}}{1 + \frac{n_{HE}}{4} - \frac{n_{OE}}{2} - \frac{n}{4}\frac{n_{NE}}{n_{NN}} - (1 + \frac{n_{HV}}{4} - \frac{n_{OV}}{2} - \frac{n}{4}\frac{n_{NV}}{n_{NN}})\,{}_mY_{VE}} \tag{9}$$

The contribution of energetics to the RQ is thus via the module for growth. The respiration quotient is in practice usually taken to be a constant for a particular species. Within the DEB model, the RQ is independent of the states of the animal (size l and reserve density e) if the following condition on the composition of E, V and N holds

$$ \frac{1 + \frac{n_{HE}}{4} - \frac{n_{OE}}{2} - \frac{n}{4}\frac{n_{NE}}{n_{NN}}}{1 + \frac{n_{HV}}{4} - \frac{n_{OV}}{2} - \frac{n}{4}\frac{n_{NV}}{n_{NN}}} = \frac{1 - n_{NE}\frac{n_{CN}}{n_{NN}}}{1 - n_{NV}\frac{n_{CN}}{n_{NN}}} \tag{10}$$

in which case

$$ -{}_mY_{CO} = \frac{1 - n_{NE}\frac{n_{CN}}{n_{NN}}}{1 + \frac{n_{HE}}{4} - \frac{n_{OE}}{2} - \frac{n}{4}\frac{n_{NE}}{n_{NN}}} = \frac{1 - n_{VN}\frac{n_{CN}}{n_{NN}}}{1 + \frac{n_{HV}}{4} - \frac{n_{OV}}{2} - \frac{n}{4}\frac{n_{NV}}{n_{NN}}} \tag{11}$$

The respiration rate (the oxygen consumption rate as well as the carbon dioxide production rate) is then proportional to the catabolic rate. This constraint involves the composition

of both structural biomass and reserves. It might be more interesting to exploit the more stringent conditions

$$\begin{pmatrix} n_{NE} & -2n_{NE} & -n_{HE}+2n_{OE} \\ n_{NV} & -2n_{NV} & -n_{HV}+2n_{OV} \end{pmatrix} \begin{pmatrix} n_{HN} \\ n_{ON} \\ n_{NN} \end{pmatrix} = \mathbf{0} \tag{12}$$

on the composition of the nitrogen waste, since this set uncouples, to some extent, the composition of structural biomass and reserves. These conditions are obtained from condition (10) by equating the numerators and denominators separately.

Several simple expressions can be obtained for changes over the whole incubation period that are useful for practical work. The initial weight (age $a = 0$) and the weight at birth (i.e. hatching, age $a = a_b$), excluding membranes and nitrogen waste, are:

$$\begin{pmatrix} W_w(0) & W_w(a_b) \end{pmatrix} = [d_{me}]V_m \begin{pmatrix} w_E & w_V \end{pmatrix} \begin{pmatrix} e_0 & e_b l_b^3 \\ 0 & l_b^3 \end{pmatrix} \tag{13}$$

with the molecular weights $\begin{pmatrix} w_E & w_V \end{pmatrix} = \begin{pmatrix} 12 & 1 & 16 & 14 \end{pmatrix} \mathbf{n}$. The scaled reserve densities e_0 and e_b are defined as $e_\cdot \equiv E_\cdot([E_m]V_m)^{-1}$, where $E_\cdot$ denotes the initial amount of reserves or the amount at hatching.

The relative weight at hatching is $W_w(a_b)/W_w(0) = (e_b + w_V/w_E)l_b^3/e_0$.

The total production of 'minerals' during incubation, $N_\cdot \equiv \int_0^{a_b} \dot{k}_E(a) Y_{\cdot E}(a)\, da$, amounts to

$$\begin{pmatrix} N_C & N_A & N_O & N_N \end{pmatrix}^T = \mathbf{un} \begin{pmatrix} \dot{\mu}_E^{-1}(E_0 - E_b) \\ -[d_{mv}]V_b \end{pmatrix} \tag{14}$$

where a negative production implies a consumption (in the case of N_O). Note that ${}_mY_{CO}N_O = N_C$.

Juvenile mass transformation

For the juvenile mass transformation we have to add

$$\underset{\text{food, } X}{+ {}_mY_{XE}\,\mathrm{CH}_{n_{HX}}\mathrm{O}_{n_{OX}}\mathrm{N}_{n_{NX}}} \underset{\text{faeces, } F}{+ {}_mY_{FE}\,\mathrm{CH}_{n_{HF}}\mathrm{O}_{n_{OF}}\mathrm{N}_{n_{NF}}} \tag{15}$$

to the macro-chemical reaction equation (1).

The mass balance equation now becomes

$$\mathbf{Y}_M = -\mathbf{u} \begin{pmatrix} 1 & 1 & 1 & 1 \\ n_{HX} & n_{HV} & n_{HE} & n_{HF} \\ n_{OX} & n_{OV} & n_{OE} & n_{OF} \\ n_{NX} & n_{NV} & n_{NE} & n_{NF} \end{pmatrix} \begin{pmatrix} {}_mY_{XE} \\ {}_mY_{VE} \\ -1 \\ {}_mY_{FE} \end{pmatrix} \equiv -\mathbf{u}\mathbf{n}\mathbf{Y}_D \tag{16}$$

where $\mathbf{n}$ and $\mathbf{Y}_D$ are supplemented with respect to the embryonic case to include the processes of feeding and defacation.

The 'mineral' fluxes are again given by $\dot{\mathbf{k}}_M = -\mathbf{u}\mathbf{n}\dot{\mathbf{k}}_D$. The 'organic' fluxes are

$$\dot{\mathbf{k}}_D \equiv \begin{pmatrix} \dot{k}_X \\ \dot{k}_V \\ \dot{k}_E \\ \dot{k}_F \end{pmatrix} = \tilde{\mu}_E^{-1} \begin{pmatrix} -\frac{d_{mx}}{[d_{me}]} \frac{\{\dot{I}_m\}}{\dot{v}} & 0 & 0 \\ 0 & 0 & \frac{[d_{me}]}{[d_{me}]} \frac{1}{\kappa g} \\ 1 & -1 & -1 \\ \frac{d_{mf}}{[d_{me}]} \frac{\{\dot{I}_m\}}{\dot{v}} & 0 & 0 \end{pmatrix} \begin{pmatrix} \dot{A} \\ \dot{D} \\ \dot{G}_s \end{pmatrix} \equiv \tilde{\mu}_E^{-1} \mathbf{J} \begin{pmatrix} \dot{A} \\ \dot{D} \\ \dot{G}_s \end{pmatrix} \tag{17}$$

The expression for $\dot{k}_E$ follows from $\dot{A} - \dot{D} - \dot{G}_s = \dot{A} - \dot{C}$, which simply tells that reserve energy is generated by assimilation and used by catabolism. The expression for $\dot{k}_X$ follows from the definition of the energy conductance $\dot{v} \equiv \frac{\{\dot{A}_m\}}{[E_m]}$, where $\{\dot{A}_m\}$ stands for the maximum surface area-specific assimilation energy. The factor $\tilde{\mu}_E^{-1} \frac{d_{mx}}{[d_{me}]} \frac{\{\dot{I}_m\}}{\dot{v}} = d_{mx} \frac{\{\dot{I}_m\}}{\{\dot{A}_m\}}$ converts assimilation energy to C-moles of food, where d_{mx} converts volume of food into C-moles of food. The actual conversion is from food to assimilation energy, of course. For the faeces flux $\dot{k}_F$ holds a similar relationship. As before, we have that the mass fluxes are weighted sums of powers. Note that the embryonic $\mathbf{J}$ is still contained in the juvenile $\mathbf{J}$, but that the latter is extended to include assimilation.

The yield coefficients become

$$_m\!Y_{XE} = \frac{\dot{k}_X}{\dot{k}_E} = \frac{d_{mx}}{[d_{me}]} \frac{\{\dot{I}_m\}}{\dot{v}} \frac{f}{f - \frac{g+l}{1+g/e}} \overset{e=f}{\rightarrow} \frac{d_{mx}}{[d_{me}]} \frac{\{\dot{I}_m\}}{\dot{v}} \frac{f+g}{f-l} \tag{18}$$

$$_m\!Y_{VE} = \frac{\dot{k}_V}{\dot{k}_E} = \frac{[d_{me}]}{[d_{me}]} \frac{e-l}{fe+fg-eg-el} \overset{e=f}{\rightarrow} \frac{[d_{me}]}{[d_{me}]} \frac{1}{f} \tag{19}$$

$$_m\!Y_{FE} = \frac{\dot{k}_F}{\dot{k}_E} = -_m\!Y_{XE} \frac{d_{mf}\{\dot{F}_m\}}{d_{mx}\{\dot{I}_m\}} \tag{20}$$

The specific dynamic action (SDA) is defined as the oxygen consumption that is associated with the feeding process. Apart from a small part that relates to the processing of proteins, the SDA is little understood (Withers, 1992). We can obtain it as the result of the conservation law for mass. The SDA per C-mole of food turns out to be independent of the states of the animal (reserves e and size l), but it still depends on a few DEB parameters as

$$\frac{\dot{k}_{OA}}{d_{mx}\dot{l}} = \mathbf{u}_{O\bullet}\mathbf{n} \begin{pmatrix} 1 \\ 0 \\ -\frac{[d_{me}]}{d_{mx}} \frac{\dot{v}}{\{\dot{I}_m\}} \\ -\frac{d_{mf}}{d_{mx}} \frac{\{\dot{F}_m\}}{\{\dot{I}_m\}} \end{pmatrix} \tag{21}$$

where $\mathbf{u}_{O\bullet}$ denotes the row of $\mathbf{u}$ that relates to O, which is the third row. The respiration quotient $-_m\!Y_{CO}$ is independent of l and e at starvation ($\dot{l} = 0$) if (10) applies.

Adult mass transformation

For the adult mass transformation in females, we have to add

$$+ _m\!Y_{RE}\ \mathrm{CH}_{n_{HE}}\mathrm{O}_{n_{OE}}\mathrm{N}_{n_{NE}}^{\text{eggs, }R} \tag{22}$$

to the macro-chemical reaction equation (1) and (15). The DEB model assumes that embryos initially consist of pure reserves, the structural component being negligibly small. Therefore no new components show up.

The 'mineral' fluxes are again given by $\dot{\mathbf{k}}_M = -\mathbf{u}\mathbf{m}\dot{\mathbf{k}}_D$. The fluxes of reserves and reproduction are

$$\dot{k}_E = \dot{\mu}_E^{-1}(\dot{A} - \dot{C}) \tag{23}$$

$$\dot{k}_R = \dot{\mu}_E^{-1} q \dot{R} \tag{24}$$

where q represents overhead costs of the reproduction process. Since adult reserves are transformed into embryonic reserves, this factor is probably close to 1. The reproductive output appears as a return flux to the reserve component because embryonic reserves have the same composition as adult reserves. The 'organic' fluxes are

$$\dot{\mathbf{k}}_D \equiv \begin{pmatrix} \dot{k}_N \\ \dot{k}_V \\ \dot{k}_E + \dot{k}_R \\ \dot{k}_F \end{pmatrix} = \dot{\mu}_E^{-1} \begin{pmatrix} -\frac{d_{mA}}{[d_{mc}]} \frac{\{\dot{k}_m\}}{v} & 0 & 0 \\ 0 & 0 & \frac{[d_{mv}]}{[d_{mc}]} \frac{1}{\kappa g} \\ 1 & -1 & -1 \\ \frac{d_{mA}}{[d_{mc}]} \frac{\{\dot{E}_m\}}{v} & 0 & 0 \end{pmatrix} \begin{pmatrix} \dot{A} \\ \dot{D} \\ \dot{G}_s \end{pmatrix} \equiv \dot{\mu}_E^{-1} \mathbf{J} \begin{pmatrix} \dot{A} \\ \dot{D} \\ \dot{G}_s \end{pmatrix} \tag{25}$$

Energy in urine is treated similar to energy in faeces in standard 'static' energy budget studies, by subtracting both from energy contained in food to arrive at metabolizable energy that is available to the animal. The DEB model leads to a different point of view, where dissipating power and anabolic power also contribute to the nitrogen waste. The flux of nitrogen waste can be written as

$$\dot{k}_N = -\frac{\mathbf{n}_{N\cdot}}{\dot{\mu}_E n_{NN}} \mathbf{J} \begin{pmatrix} \dot{A} \\ \dot{D} \\ \dot{G}_s \end{pmatrix} = \dot{k}_{NA} + \dot{k}_{ND} + \dot{k}_{NG} \tag{26}$$

where $\mathbf{n}_{N\cdot}$ denotes the row of $\mathbf{n}$ that relates to N, which is the fourth row. If $n_{NE} < \frac{[d_{mv}]}{[d_{mc}]} \frac{n_{NV}}{\kappa g}$, the flux of nitrogen waste that relates to anabolic power, $\dot{k}_{NG}$, is negative, meaning that nitrogen is built in rather than wasted in the transformation from reserves to structural biomass. The flux of nitrogen waste that relates to dissipating power amounts to $\dot{k}_{ND} = \frac{\dot{D}}{\dot{\mu}_E} \frac{n_{NE}}{n_{NN}}$, which can be a substantial part of the total flux of nitrogen waste. The crucial difference between the 'static' and the 'dynamic' points of view is in the existence of reserves and its role in energetics.

INDIRECT CALORIMETRY

Indirect calorimetry uses measurements of oxygen consumption, carbon dioxide and nitrogen production to estimate dissipating heat $\dot{k}_H$:

$$\dot{k}_H = \mathbf{h}' \dot{\mathbf{k}}_M \quad \text{with} \tag{27}$$

$$\mathbf{h}' = \begin{pmatrix} h_C & h_A & h_O & h_N \end{pmatrix} \tag{28}$$

Its basis is just empirical. Examples are: $h_C = 0.06$ kJ.mol^{-1}, $h_A = 0$, $h_O = -0.35$ kJ.mol^{-1} and $h_N = 0.55\frac{n_{CN}}{n_{NN}}$ kJ.mol^{-1} in aquatic animals (Brafield and Llewellyn, 1982) or $0.086\frac{n_{CN}}{n_{NN}}$ kJ.mol^{-1} in birds (Blaxter, 1989). For mammals, corrections for methane production have been proposed (Brouwer, 1958). These coefficients can be obtained by direct calorimetry, using multiple regression. In the present notation, we can write for the dissipation heat $\dot{k}_H$

$$\dot{k}_H = \mathbf{h}^t \dot{\mathbf{k}}_M = -\mathbf{h}^T \mathbf{un} \dot{\mathbf{k}}_D = -\tilde{\mu}_E^{-1} \mathbf{h}^T \mathbf{un} \mathbf{J} \begin{pmatrix} \dot{A} \\ \dot{D} \\ \dot{G}_s \end{pmatrix} \tag{29}$$

We can try to give a rationale for such relationships on the assumption that changes in entropy during the macro-chemical reaction are negligible. The chemical potentials of the various components can be collected in the vectors

$$\tilde{\boldsymbol{\mu}}_M^t \equiv \begin{pmatrix} \tilde{\mu}_C & \tilde{\mu}_A & \tilde{\mu}_O & \tilde{\mu}_N \end{pmatrix} \tag{30}$$

$$\tilde{\boldsymbol{\mu}}_D^t \equiv \begin{pmatrix} \tilde{\mu}_X & \tilde{\mu}_V & \tilde{\mu}_E & \tilde{\mu}_F \end{pmatrix} \tag{31}$$

The dissipating heat $\dot{k}_H$ now follows from the energy balance equation

$$0 = \dot{k}_H + \dot{k}_E \tilde{\boldsymbol{\mu}}_M^T \mathbf{Y}_M + \dot{k}_E \tilde{\boldsymbol{\mu}}_D^T \mathbf{Y}_D \tag{32}$$

$$= \dot{k}_H + (\tilde{\boldsymbol{\mu}}_D^T - \tilde{\boldsymbol{\mu}}_M^T \mathbf{un}) \dot{\mathbf{k}}_D \tag{33}$$

$$= \dot{k}_H + \tilde{\mu}_E^{-1} (\tilde{\boldsymbol{\mu}}_D^T - \tilde{\boldsymbol{\mu}}_M^T \mathbf{un}) \mathbf{J} \begin{pmatrix} \dot{A} \\ \dot{D} \\ \dot{G}_s \end{pmatrix} \tag{34}$$

Given the thermodynamic assumptions mentioned in the section 'powers', dissipating heat is again a weighted sum of the three powers, which justifies the method of indirect calorimetry. If the assumption of homeostasis holds strictly, nothing is gained by correcting for methane production in mammals, as long as comparisons are made within one type of food.

Now we can reverse the argument and wonder how measurements of heat dissipation can be used to obtain the chemical potentials. In the definition of chemical potentials, we still have the freedom to choose the frame of reference.

The combustion frame of reference is defined such that $\tilde{\boldsymbol{\mu}}_M \equiv \mathbf{0}$. When the superscript o refers to the standard thermodynamic frame of reference, the chemical potential of an organic compound $\mathrm{CH}_{n_H} \mathrm{O}_{n_O} \mathrm{N}_{n_N}$ in the combustion frame of reference is given in terms of that in the standard frame of reference by

$$\tilde{\mu} = \tilde{\mu}^\circ - \tilde{\boldsymbol{\mu}}_M^{\circ T} \begin{pmatrix} 1 - n_N \frac{n_{CN}}{n_{NN}} \\ \frac{n_H}{2} - \frac{n_N}{2} \frac{n_{HN}}{n_{NN}} \\ \frac{n_O}{2} - 1 - \frac{n_H}{4} - \frac{n_N}{4} \frac{4n_{CN} - n_{HN} + 2n_{ON}}{n_{NN}} \\ n_N \end{pmatrix} \tag{35}$$

Within this frame of reference, substitution of the dissipating heat results in

$$\tilde{\boldsymbol{\mu}}_D^t = \mathbf{h}^T \mathbf{un} \tag{36}$$

This leads us to the problem of how to obtain the relative abundances of the elements in the structural body mass and in the reserves. The problem can be solved by the use of measured total body compositions at various food densities. The DEB model describes how the amount of reserves relates to ingestion rates, which means that food-dependent changes in body composition can be used to disentangle reserves from structural biomass. This method is applied in Kooijman (1993).

Water balance

For aquatic animals, we have that the drinking rate equals the water flux, $\dot{k}_{A_d} = \dot{k}_A$, but terrestrial animals have to deal with evaporation of water. The DEB model implies a water outflux of $\dot{k}_A$, which must be compensated by evaporating, $\dot{k}_{A_e}$, and drinking, $\dot{k}_{A_d}$, such that $\dot{k}_{A_d} = \dot{k}_A + \dot{k}_{A_e}$. Embryos usually do not drink and are 'designed' such that evaporation takes care of water outflux, although small changes have been found. The water content of tissues in birds gradually decreases during growth, which led Ricklefs (Ricklefs and Webb, 1985) and Kowanarsky (1988) to model juvenile growth on the basis of the water content of the tissue. We here idealize the process by assuming strict homeostasis for both the structural biomass and the reserves, while focussing on juveniles and adults. Note that water emission via urine is incorporated in the composition of the nitrogen waste, which could be large enough to let the water outflux $\dot{k}_A$ be negative and turn it into a water influx.

Evaporation has two main routes, one via water loss linked to respiration and one via transpiration, so $\dot{k}_{A_e} = \dot{k}_{A_O} + \dot{k}_{A_t}$. Water loss via respiration is proportional to oxygen consumption via the amount of inhaled air, so $\dot{k}_{A_O} = \dot{k}_O d_{AO}$, while transpiration is proportional to surface area, so $\dot{k}_{A_t} = \{\dot{k}_{A_t}\} V_m^{2/3} l^2$, where $\{\dot{k}_{A_t}\}$ does not depend on the state of the animal. Both loss rates depend on water pressure in the air and temperature. The DEB model leads to a drinking rate of

$$\dot{k}_{A_d} = \begin{pmatrix} 0 & 1 & d_{AO} & 0 \end{pmatrix} \dot{\mathbf{k}}_M + \{\dot{k}_{A_t}\} V_m^{2/3} l^2 \tag{37}$$

This two-parameter model for the drinking process is, of course, an idealized picture which pushes the concept of homeostasis to the extreme. The water content of urine is actually rather variable, depending on environmental and behavioural factors. However, the model might be helpful as a first approximation to reveal the coupling that must exist between drinking and energetics.

Conclusion

Given a dynamic energy budget model that covers the full life cycle, all mass fluxes as well as the dissipation heat can be written as weighted sums of three powers

- assimilation power, which is directly coupled to food input
- dissipating power, which combines all non-assimilation powers that are not involved in the process of synthesis of structural components

- anabolic power, which is directly coupled to synthesis of structural components

Because of these three powers, dissipating heat can be written as a weighted sum of three 'mineral' fluxes, which underpins the method of indirect calorimetry. Moreover, indirect calorimetry can be used to obtain the chemical potentials of reserves and structural biomass.

These results show that nitrogen waste is not coupled to the feeding process only, which is the implicit assumption of standard static energy budget studies. They also explain the existence of the Specific Dynamic Action. Finally, the coupling between drinking and energetics has been elucidated.

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