

# The nature of nutrition: a unifying framework

Stephen J. Simpson<sup>A,C</sup> and David Raubenheimer<sup>B,C</sup>

<sup>A</sup>School of Biological Sciences & The Charles Perkins Centre, The University of Sydney, Sydney, NSW 2006, Australia.

<sup>B</sup>Institute of Natural Sciences, Massey University, Albany, Private Bag 102 904, North Shore Mail Centre, Auckland, New Zealand.

<sup>C</sup>Corresponding authors. Email: stephen.simpson@sydney.edu.au; d.raubenheimer@massey.ac.nz

**Abstract.** We present a graphical approach, which we believe can help to integrate nutrition into the broader biological sciences, and introduce generality into the applied nutritional sciences. This ‘Geometric Framework’ takes account of the fact that animals need multiple nutrients in changing amounts and balance, and that nutrients come packaged in foods that are often hard to find, dangerous to subdue and costly to process. We then show how the Geometric Framework has been used to understand the links between nutrition and relevant aspects of the biology of individual animals. These aspects include the physiological mechanisms that direct the nutritional interactions of the animal with its environment, and the fitness consequences of these interactions. Having considered the implications of diet for individuals, we show that these effects can translate into the collective behaviour of groups and societies, and in turn ramify throughout food webs to influence the structure of ecosystems.

**Additional keywords:** ageing, cannibalism, conservation, Geometric Framework.

Received 31 August 2011, accepted 18 April 2012, published online 9 May 2012

## Introduction

Nutrition touches, links and shapes all aspects of the biological world (Simpson and Raubenheimer 2012). It builds the components of organisms, and fuels the dynamic interactions between these components; it determines whether or not wild animals thrive, how their populations grow, decline and evolve, and how assemblages of interacting species (ecological communities) and ecosystems are structured. Nutrition also drives the affairs of humans, from individuals to global geopolitics. Climate change, population growth, urbanisation, environmental degradation and species extinctions all are in one way or another linked to the need for nutrients. In short, nutrients are the interconnecting threads in the web of life.

And yet the science of nutrition remains fragmented. Because of its direct importance to human health and food animal production, nutrition has traditionally been considered the domain of the medical and agricultural sciences. Research in these areas has produced a tremendously detailed account of the nutritional biology of a few species. By contrast, with some exceptions, nutrition in the biological sciences has tended to adopt simpler, more general approaches that are applicable across the diversity of animals. Foraging might, for example, be considered a process of acquiring energy or minimising time exposed to predators, rather than a complex balancing act of obtaining enough – but not too much – of the many nutrients that are needed

for sustaining health and reproduction. The advantage of this simplified approach is that it has supported the development of powerful general frameworks for biological processes, unhindered by the staggering nutritional complexity that has been uncovered in the more applied nutritional sciences. Such unidimensional approaches to nutrition have generated valuable insights into foraging behaviour and provided a heuristic framework for thinking about ecological processes (White 1983; Stephens and Krebs 1986; Stephens *et al.* 2007). They have, however, contributed little to the understanding of which nutrients or combinations of nutrients influence foraging, and how animal requirements for these influence ecological processes. For this, models are needed that are *nutritionally explicit*, in the sense that they enable a study to address these questions directly (Raubenheimer *et al.* 2009).

Achieving nutritional homeostasis involves a complex interplay between multiple and changing nutrient needs and variable foods. Long-term evolutionary processes have ensured that animals are equipped with the mechanisms to deal with these complexities, but considerable challenges remain for nutritional biologists to understand these processes. Many aspects of the relationship between animals and their nutritional environments underscore this challenge. First, foods are complex mixtures of multiple components, each of which has its own functional implications for the animal. Some are necessary for maintaining

good health, while others are hazardous and best avoided (e.g. the antipredator chemical defences produced by some plants and animals: Sotka *et al.* 2009). To further complicate issues, some toxins can be beneficial if ingested in low quantities, and even essential nutrients can be toxic if overingested (Raubenheimer and Simpson 2009). Second, a given food component (at a stipulated dose) can have multiple influences on an animal. Third, most aspects of animal function are influenced by multiple food components. Finally, and in some respects most challenging of all, food components interact in intricate ways in their effects on animals.

Disentangling this web of interconnections can be as complex and daunting as it is important for understanding the biology of animals, and for managing the complex relationships between our own species and the world that we inhabit. To succeed, an approach is needed that systematically deals with each of the challenges mentioned above. Namely, it should provide a framework within which multiple food components and animal attributes can be distinguished, and the relationships among components and attributes disentangled and then linked to individual performance, ecological outcomes and evolutionary consequences. Importantly, it should not seek to recapitulate the full complexity of nutrition, nor to oversimplify it, but rather to provide a level of complexity that best suits the research question at hand.

### The geometry of nutrition

Such an integrative framework must be able to represent the animal, the environment, and the nutritional basis for the interaction between animal and environment (Raubenheimer *et al.* 2009). A second requirement is that an integrative framework should take account of the fact that the nutritional interactions between animals and their environment take place on a stage that is constructed of many food components. Third, if the framework is to be grounded within the powerful paradigm of evolutionary biology, it is imperative that the consequences for the animal of its behavioural and physiological responses to the nutritional environment can be represented. These consequences – which include such factors as reproductive outputs, development rates, and the risk of premature death (e.g. the effectiveness of the immune system) – are relevant not only to individual wellbeing but also to population sizes. A framework that takes into account consequences therefore extends its reach both to evolution and also to nutritional aspects of population ecology. A further step, to community ecology, would be achievable if the framework fulfilled a fourth requirement, of being able to incorporate the nutritional basis of interactions among multiple species in food webs.

The Geometric Framework (GF) was designed with these core requirements in mind (Raubenheimer and Simpson 1993, 1999; Simpson and Raubenheimer 1993, 1995, 2012; Simpson *et al.* 2004; Raubenheimer *et al.* 2009). It satisfies the multiple-food-components requirement using a simple device known as a *nutrient space*. A nutrient space is a geometric space built of two or more axes, where each axis represents a food component that is suspected to play a role in influencing the animal's responses to its environment. In most cases these food components will be nutrients but, as discussed below, this is not invariably the case.

The nutrient space provides the common context in which to describe the pertinent aspects of the animal, its environment, the interactions between animal and environment, and the consequences of these interactions. In the sections that follow we describe how this is achieved.

### *Nutrient needs: the intake target*

An aspect of the animal that is fundamental to its interactions with the environment is its nutrient requirements – the amounts and balance of nutrients that it needs to eat over a given period to gain maximal benefit (usually evaluated in terms of evolutionary fitness). Nutrient requirements are fundamental in nutritional models because they provide a reference point for predicting how an animal should respond to its environment and for understanding the relationships between foods and the performance consequences of eating those foods. For example, if we know the nutrient needs of an animal and the nutritional composition of the foods available to it, we can predict which foods it would eat and which it should avoid, and if it did otherwise we would be justified in wondering why. Likewise, if the animal persisted in eating foods that do not satisfy its nutrient needs, we would have good cause to wonder about the long-term consequences of this – would it become obese, have impaired reproduction, and/or become susceptible to infectious diseases? In GF models, the optimal nutrient requirement of an animal is represented as a point (or, over time, a trajectory) in a nutrient space, called an *intake target*.

### *Foods*

Except for special cases such as salt licks, nutrients in the environment come packaged together as mixtures – foods. The diet of the animal may comprise a single food or, more usually, a combination of different amounts of several foods. Foods are modelled in a nutrient space in two ways: by the amounts of nutrients they contain and, more generally, by the balance of nutrients they contain. For many purposes, some of which will be discussed shortly, it is useful to disregard the precise amount of nutrients in a specific food item, considering rather the balance of the nutrients in the food. This general property of a food type is pictured in a geometric model as a line that passes through the origin of the graph and through a point representing any quantity of the food type. The slope of such a line indicates the ratio of the nutrient in the food. Such lines representing the balance of nutrients in foods are called *nutritional rails*, for reasons that will become clear below.

### *Reaching the intake target*

If foods are the most fundamental aspect of an animal's nutritional environment, then feeding is the primary nexus of interaction with this aspect of the environment. For the animal, the important thing about feeding is that it provides a means to change – and regulate – its nutritional state. Feeding is therefore represented in GF models by the change in the nutritional state of the animal that results from eating. This change is plotted as a trajectory through nutrient space. Such feeding trajectories have the same angle as the nutritional rail for the food being eaten, because as the animal eats it gains the nutrients in the same proportion as they are present in the food being eaten. By feeding, animals are therefore

channelled, like a train, along tracks in nutrient space set by the nutritional rails of the foods they select, with the distance of movement along these rails being determined by the amount of the food eaten.

The challenge for animals is to select foods that direct them to their intake target, and ensure that they eat enough to arrive there. The simplest way to do this is to eat a *nutritionally balanced* food, which leads directly to the target – i.e. contains the same balance of the nutrients as is needed – and is plentiful enough to enable the animal to get there. Food items that are *nutritionally imbalanced* do not lead to the intake target. The animal could nonetheless use such a food to navigate indirectly to the intake target, if it combined its intake with a second imbalanced food whose rail falls on the opposite side of the intake target. Combinations of nutritionally imbalanced foods that jointly enable animals to reach their intake target in this way are known as *nutritionally complementary* foods. The fact that many animals are able to reach their intake target by mixing nutritionally complementary foods provides a useful means for researchers to estimate the position of the intake target, based on the testable prediction that animals will have evolved regulatory mechanisms to ensure ingestion of a balanced diet (e.g. Chambers *et al.* 1995; Raubenheimer and Jones 2006).

#### *What to eat when the intake target can't be reached: rules of compromise*

In some cases, the animal might have access neither to nutritionally balanced nor nutritionally complementary foods, but only to non-complementary nutritionally imbalanced foods. In this predicament the animal cannot reach its intake target. The animal's response in this circumstance is known as a *rule of compromise* because it reflects the compromise selected by the animal between overeating some nutrients and undereating others. To derive a general description of the rule of compromise we have to measure the responses of the animal to a range of nutritionally imbalanced foods. The intake points for such an experiment jointly form an *intake array*, the shape of which provides a comprehensive description of the rule of compromise. The shape of the intake array may differ according to the animal in question and the nutrients being modelled.

#### *Processing ingested nutrients*

Our use of the phrase 'nutrient requirements' up to this point refers to the amounts and balance of nutrients that an animal needs to eat, which we have represented in geometric models as an intake target. There is, however, an even more fundamental context in which the term 'nutrient requirements' can be used, and that is in relation to the amounts and balance of nutrients that the animal needs to make available to its tissues to satisfy its various demands for energy metabolism, tissue growth, storage and so forth. How does this requirement, which we call the *nutrient target*, differ from the intake target? It differs because animals are usually unable to allocate 100% of the nutrient they eat to useful purposes, but lose some in the faeces, urine and, in some animals, through permeable membranes on the body surface (e.g. the gills of fish). To satisfy its tissue-level requirements (the nutrient target) an animal therefore needs to compensate by eating enough to cover both the nutrient target and the constrained losses. These two

components, required amount + constrained losses, jointly comprise the intake target.

Just as the intake target can be partitioned into the nutrient target + constrained losses, so too can the nutrient target be partitioned into various subtargets. When an animal reaches its nutrient target, the total pool of available nutrients needs to be divided among several functions, including growth, metabolism and reproduction (Raubenheimer and Simpson 1992, 1994, 1995). How the animal allocates these nutrients is critical to fitness and, as a result, natural selection has fashioned animal physiology to achieve a favourable strategy for investing its nutritional 'income' across its various requirements. Experiments show that, within limits, animals are able to regulate nutrient utilisation postingestively to defend a growth target, even when eating foods that prevent them from reaching the intake target, e.g. by adjusting the digestion and absorption of nutrients from the gut (e.g. Clissold *et al.* 2010) and by voiding excess ingested nutrients postabsorptively (e.g. Zanutto *et al.* 1993, 1997).

The above discussion concerns optimal nutrient allocation to growth, but we could equally model metabolic targets, reproductive targets and so forth. The overall point, however, is to demonstrate that the GF can be used to construct multidimensional models of nutrient budgets, which include functionally optimal nutrient allocations (targets) as well as the allocations that are actually achieved (Raubenheimer and Simpson 1995). If we had additional information about the benefits of achieving these optima (or the costs of failing to do so), this would put us in a good position to understand how diverse nutritional strategies have evolved.

#### *The consequences of nutritional imbalance*

If we assume that the nutritional responses of animals, including regulation to an intake target, rules of compromise and postingestive regulation, have been fashioned by natural selection, it follows that an animal that achieves its intake target will enjoy maximal Darwinian fitness. The challenge is how to include such fitness consequences in geometric models. They differ from the components we have so far integrated into the models – foods, nutrient requirements, nutrient intake and postingestive utilisation – in that fitness components are not measured in terms of nutrients, but in terms of other units such as numbers of offspring, probability of premature death, longevity and so forth. Fitness consequences cannot, therefore, be depicted in the nutrient space in the usual way.

Instead, we have borrowed from evolutionary biologists the metaphor of the 'fitness landscape', in which the consequences of nutrient intake are plotted in a map-like surface superimposed on the nutrient space. This enables us to envisage the intake target state as the summit of a Darwinian fitness mountain mapped onto nutrient intake space. The slope, steepness and curvature of the decline in elevation with distance from the intake target need not be the same for excesses and deficits of the same nutrient, or be the same for different nutrients, or be independent between nutrients. Fitness costs can be described to take account of these various possibilities using a mathematical function, the Taylor series expansion (Simpson *et al.* 2004).

Experimental examples in which the components of fitness have been mapped as landscapes onto detailed nutrient intake

arrays have begun to accumulate in recent years, notably in invertebrates (e.g. Simpson *et al.* 2004; Lee *et al.* 2008a; Maklakov *et al.* 2008; Jensen *et al.* 2012). These experiments involve maintaining individuals on one of a large number of diet compositions, and are thus laborious to conduct. Nevertheless, as we shall see below, the insights gained can be substantial.

### Mechanisms of nutritional regulation

It is clear that animals possess not one appetite system but several, and are able to regulate independently their intakes and utilisation of different nutrients to maintain intake and growth targets. We will consider the mechanisms involved in outline, highlighting the major principles and key components of regulatory systems.

Maintaining an intake target involves animals achieving three things: (1) assessing the nutritional composition of available foods, (2) assessing their own nutritional state, and (3) comparing these two to produce appropriate feeding responses. The simplest means to assess the nutritional composition of foods is to detect different nutrients by tasting them. Not surprisingly, therefore, all organisms, from bacteria to mammals, possess specialised receptors for the detection of key nutrients such as amino acids, sugars and salts. Together, these receptors provide the central nervous system of an animal with information about the nutritional composition of food before, during and after ingestion (Dethier 1976; Finger 1997; Yarmolinsky *et al.* 2009). Other chemoreceptors respond to deterrent and toxic compounds in the food (Schoonhoven *et al.* 2005; Dong *et al.* 2009). The presence or absence of nutrients that are not themselves tasted is 'inferred' by learning to associate features of a food with the consequences of ingesting it. Such associations may be positive (Sclafani 2000; Touzani and Sclafani 2005; Burke and Waddell 2011; Fujita and Tanimura 2011) or aversive (Bernays 1993; Tomé 2004; Dukas 2008), and may be made individually or in a social context (Galef and Laland 2005).

Nutritional state is assessed through systemic nutrient-sensing mechanisms and hormonal feedbacks from body reserves (Zhang *et al.* 1994; Crespi and Denver 2006; Morton and Schwartz 2011). Integration of information about food composition and nutritional state occurs both at the periphery, by nutrient-specific modulation of taste receptors (Simpson and Raubenheimer 2000; Newland and Yates 2008; Carleton *et al.* 2010), and more centrally as signals from systemic and peripheral sources converge onto the neural circuitry that controls feeding behaviour (Schwartz *et al.* 2000; Cota *et al.* 2007). Learning also plays a role, and in some cases animals are able to associate their current nutritional state with food cues previously associated with particular nutrients (e.g. Simpson and White 1990; Raubenheimer and Tucker 1997; Booth and Thibault 2000).

Postingestive regulatory responses assist in rebalancing an imbalanced nutrient intake. The gastrointestinal tract sits at the interface between the food and the internal milieu and is now appreciated to be a key site of regulation, being plastic in both structure and function in response to nutritional state (Raubenheimer and Bassil 2007; Clissold *et al.* 2010; Sørensen *et al.* 2010). Having been digested and absorbed across the gut, further rebalancing of nutrient supply can be achieved by differentially voiding excess nutrients and conserving nutrients

that are in limited supply (Zanotto *et al.* 1997; Stock 1999; Silva 2006; Cypess *et al.* 2009).

### Less food, less sex, live longer?

Nutritional geometry has been used to relate nutrition to several different fitness-related consequences. Among the most interesting and significant are the relationships between nutrition, longevity and reproductive output.

Since a seminal publication on rats by McCay *et al.* (1935), the view that dietary restriction without severe malnutrition prolongs life has become *the* big idea in research on the biology of ageing (Weindruch and Walford 1988; Masoro 2005; Colman *et al.* 2009; Everitt *et al.* 2010). The list of species that live longer when modestly food deprived is now extensive: yeasts, nematode worms, fruit flies, rodents, monkeys and many more. But what does dietary restriction actually mean? The most widely accepted view is that eating fewer calories (called 'caloric restriction' or 'calorie restriction') is the reason for living longer, whatever the source of those calories might be, whether protein, carbohydrate or fat (Weindruch and Walford 1988; Masoro 2005; Everitt *et al.* 2010). Some had suggested, however, that calories may not hold the key to a long life; rather, specific nutrients such as proteins and certain amino acids may be responsible (e.g. Ross 1961; Zimmerman *et al.* 2003; Mair *et al.* 2005).

As discussed above, by restricting study animals throughout their lives to one of a sufficiently large number of dietary treatments it becomes possible to map response surfaces onto nutrient intake arrays for aspects of performance, including lifespan, rate of ageing, reproductive success and other variables. From these plots the main and interactive effects of different nutrients, as distinct from energy intake alone, can be separated and attributed (Simpson and Raubenheimer 2007; Archer *et al.* 2009).

Lee *et al.* (2008a) set out to try such an experiment on one of the best known model systems for ageing research, the fruit fly (*Drosophila melanogaster*). More than 1000 mated female fruit flies were kept individually on one of 28 diets varying in the ratio and concentration of yeast to sugar and intake and egg production were measured over the lifespan of each fly. Intakes of protein and carbohydrate, which are the major energy-yielding nutrients for *Drosophila*, were calculated from the volumes of diet ingested. The consequences for lifespan and egg production were examined by plotting response surfaces on top of the array of protein and carbohydrate intakes. Flies lived longest on a diet containing a 1 to 16 ratio of protein to carbohydrate and lived progressively less long as the ratio of protein to carbohydrate either decreased to zero or increased beyond 1 : 16. If lifespan corresponded with the number of calories eaten, as predicted by the calorie-restriction hypothesis, the lifespan contours would run parallel with lines of equal calorie intake (isocaloric lines). However, the contours of the longevity landscape ran almost at right angles to these lines. The data therefore prove that caloric restriction cannot explain the variation in lifespan; instead, the balance of carbohydrate to protein ingested correlated most strongly with longevity.

A parallel experiment was conducted by Fanson *et al.* (2009) on the Queensland fruit fly (*Bactrocera tryoni*). The results were very similar to those for *Drosophila*: dietary protein to carbohydrate ratio and not energy intake was strongly associated

with lifespan. A subsequent experiment using a chemically defined mixture of amino acids instead of yeast confirmed that protein rather than some other correlated component in yeast was responsible for the life-shortening effect of a high-yeast, low-sugar diet (Fanson and Taylor 2011). Others reached similar conclusions regarding the importance of the protein to carbohydrate ratio in the diet for longevity in flies (e.g. Carey *et al.* 2008; Skorupa *et al.* 2008; Ja *et al.* 2009; Le Rohellec and Le Bourg 2009; Vigne and Frelin 2010), crickets (Maklakov *et al.* 2008), ants (Dussutour and Simpson 2009; Cook *et al.* 2010) and honey bees (Pirk *et al.* 2010), and the same may well be true for mammals (Orentreich *et al.* 1993; Zimmerman *et al.* 2003; Miller *et al.* 2005; Simpson and Raubenheimer 2007, 2009).

### *Less sex, live longer?*

Living long enough to reproduce is the *raison d'être* of all organisms, so it is no surprise that when resources are scarce many organisms shut down reproduction and wait it out until conditions improve and the prospects for reproduction are brighter (Zera and Harshman 2001). Since 'waiting it out' may lead to organisms living longer than if they had reproduced earlier in life – sometimes tens or hundreds of times longer in the case of creatures that enter a state of suspended animation (Withers and Cooper 2010) – the idea arose that reproduction and ageing trade-off against one another by competing for resources. Hence, limited resources are allocated with highest priority to maintaining and repairing the organism's body (its 'soma'), thereby improving the chances that it will live long enough to experience better conditions and reproduce (Kirkwood 2005). The obverse of this trade-off hypothesis is that when there are sufficient resources to support reproduction, somatic maintenance is only allocated sufficient resources to survive the reproductive period – so, not only does inhibition of reproduction cause delayed ageing, reproduction effectively shortens lifespan. A variant of this idea is that reproduction doesn't just compete with somatic maintenance for resources it produces its own damaging side-effects that shorten lifespan (Tatar 2007; O'Brien *et al.* 2008). Under all of these various manifestations of the trade-off hypothesis, maximal lifespan and maximal reproductive output are mutually exclusive.

We will come back to the issue of mutual exclusiveness, but we will first consider what is meant by 'resources'. When considering the trade-offs between reproduction and ageing, 'resources' and 'energy' are usually used interchangeably. However, as we have discussed above, energy alone cannot explain ageing and lifespan: so how do lifespan and reproduction relate to one another in a more complex representation of the nutritional landscape? Lee *et al.* (2008a) found that mated female *Drosophila* lived longest on a diet containing a 1 : 16 ratio of protein to carbohydrate (P : C). The same flies, however, laid most eggs across their lifespan when confined to a diet comprising a more protein-rich diet, 1 : 4 P : C. An increase in P : C beyond 1 : 4 resulted in a drop in lifetime egg production. These results suggest two important conclusions. First, calories cannot explain the relationships between diet, longevity and reproduction; instead, these relationships can only be understood by considering nutrient balance. Second, because the performance landscapes for lifespan and lifetime egg production had different shapes and their peaks lay in different locations, flies cannot maximise both lifespan and lifetime egg production on a single diet.

An interesting question is which peak do the flies choose to climb if given a choice? To address this, Lee *et al.* (2008a) offered flies one of nine complementary food choices in the form of separate yeast and sugar solutions differing in concentration. The flies converged upon a diet comprising 1 : 4 P : C, thereby maximising lifetime egg production and paying the price of a diminished lifespan. The experiment of Maklakov *et al.* (2008) on field crickets added a new dimension by considering reproductive effort in both females and males. A major investment in reproduction by male crickets is calling to attract females. Not only is singing energetically expensive, but in addition to luring females it may attract the unwelcome attention of predators. For females, the number of eggs laid was counted, and for males the amount of time spent singing throughout their lives was recorded on 24 diets varying in protein and carbohydrate content. Both sexes of cricket lived longest on low P : C diets, and like flies, female crickets laid most eggs on a more protein-rich diet (P : C 1 : 1). Males, however, sang most on low P : C diets. For males, the response surfaces for lifespan and singing were similar, but female crickets, like female flies, were faced with a quandary: they could not live longest and lay most eggs on the same diet.

South *et al.* (2011) used nutritional geometry to show that nutrient balance plays another role in defining the battle of the sexes, this time in cockroaches. These authors manipulated the intake of proteins and carbohydrates by male cockroaches, *Nauphoeta cinerea*, and related this to their production of sex pheromones, their success in being chosen as mates by females, and their status among competing males. Carbohydrate (but not protein) intake affected sex pheromone production and male attractiveness, but did not affect male dominance status. When males were given a choice between complementary foods, they selected a higher carbohydrate intake, thereby smelling more attractive to females.

Returning to the matter of the mutual exclusiveness of reproduction and somatic maintenance, the peaks for maximal reproduction and longevity occupied different regions in nutritional space for female insects (although they came close to aligning for male field crickets), such that females could not achieve maximal fecundity and lifespan on the same diet. The trade-off theory proposes that longevity and reproductive output cannot occur simultaneously either because they compete for the same resources, or there are direct costs to reproduction that shorten lifespan. Is this true? Grandison *et al.* (2009) have shown that, for flies at least, the answer is 'no'. When fed a low P : C diet, flies laid fewer eggs and lived longer than if fed a higher P : C diet. When a mixture of essential amino acids was added to the restricted diet, flies increased their egg production but lived less long – as expected from the trade-off hypothesis. However, when the restricted diet was augmented with only methionine, flies both lived long *and* had high egg production. Maximal longevity and fecundity can co-exist under the appropriate nutritional conditions; which means that the trade-off hypothesis as usually formulated is not correct, nor is the variant hypothesis that there are direct costs of reproduction that shorten lifespan (see also Flatt 2011; Tatar 2011).

### **Beyond macronutrients**

To this point in we have explored geometric models that include the macronutrients as focal dimensions. This is for a reason: as the

data show, macronutrients can explain a good deal of the variation in the behavioural, physiological and performance responses of animals. Macronutrients are, however, clearly not the only functionally important nutritional components of foods: the constituent molecules in macronutrients (amino acids and fatty acids, for example) and micronutrients such as vitamin and minerals also play a critical role in an animal's nutritional strategies and physiology, as do other components of foods that would not normally be considered nutrients. Examples of the latter include refractory fibre (e.g. lignin from plants and chitin from some animal foods), and a range of secondary metabolites that play various roles from toxic defences to essential antioxidants and even antibiotic components that act as medicines (Huffman 2001, 2003; Villalba and Provenza 2007).

Conventionally, those chemical compounds in food that enhance health and fitness and elicit appetitive responses (e.g. amino acids, fatty acids, sugars) are categorised as 'nutrients', while those that are deleterious and trigger aversive and defensive responses (e.g. alkaloids, polyphenolics, terpenoids) are classified as 'toxins'. In fact, 'toxin' versus 'nutrient' is a loose distinction, and for many purposes a more helpful dichotomy is between the adjectival versions 'toxic' versus 'nutritious' or, more generally, 'deleterious' versus 'beneficial' (Berenbaum 1995; Raubenheimer and Simpson 2009). We will use two well established phenomena to illustrate this point: hormesis and Bertrand's rule.

#### *Hormesis and Bertrand's rule*

Hormesis is a concept developed in toxicology, in which the effects on biological systems (cells, tissues, organs, organisms, populations) of exposure to a substance are reversed with increasing concentration (Calabrese and Baldwin 2003). Hormetic dose–response relationships can take two forms. The most common of these is the inverted U-shape, in which low doses of a substance stimulate and high-doses inhibit beneficial biological responses (e.g. growth, fecundity or longevity). A second form is the 'J-shaped' curve, where low doses reduce and high doses exacerbate a deleterious response (e.g. tumour formation, mortality, or growth suppression).

Over recent decades, it has become apparent that hormetic responses are common, having been observed for a wide range of chemicals, organisms, and biological responses (Calabrese *et al.* 1999; Calabrese 2005). We have argued (Raubenheimer and Simpson 2009), however, that this unification will remain incomplete until it encompasses parallel developments in the nutritional sciences.

Although some researchers in the area of hormesis have recognised similarities between toxin and nutrient dose–response curves (Luckey and Stone 1960; Hayes 2007), there have been separate developments in the nutritional sciences that are directly relevant. These developments stem back to the French scientist Gabriel Bertrand, who in 1912 established a rule concerning the dose–response curve for mineral nutrients, which is essentially that too little or too much of a mineral impairs biological function.

Bertrand's rule is believed to apply to all essential micronutrients, with the detailed shape of the curve depending on the nutrient in question and the biological context (Mertz 1981). It was generally assumed that Bertrand's rule does not

apply to macronutrients, despite evidence to the contrary – not least of which is the starvation (costs of energy shortage) to obesity (costs of energy excesses) spectrum in modern humans. Indeed, the concepts of targets and rules of compromise in the GF are predicated on there being costs to excesses as well as deficits of macro- and micronutrients. That there are indeed measurable costs to both excesses and deficits of macronutrients is now experimentally verified in several instances (Raubenheimer *et al.* 2005, 2009; and see previous section). We suspect that Bertrand's rule is at least as prevalent in nutrition as is hormesis in toxicology (see also Boersma and Elser 2006).

#### *'Toxin' or 'toxic'*

As we have seen, there is good evidence that animals balance their intake of foods so as to avoid ingesting both excesses and deficits of specific nutrients, and in many cases a good deal is known about the mechanisms involved. The recent literature shows that animals likewise regulate their intake of toxins so as to avoid ingesting a toxic overdose – leading to the term 'nutritional toxicology' being coined (Torregrossa and Dearing 2009). An important question is whether animals also select foods specifically in the context of ingesting toxins at levels that are beneficial. The interesting phenomenon of 'self-medication' suggests that they do.

#### *Self-medication and ecological immunology*

Self-medication, or zoopharmacognosy (Rodriguez and Wrangham 1993), is the phenomenon in which animals use plant secondary metabolites or other non-nutritional substances to prevent or treat disease (Huffman 2003; Villalba and Provenza 2007). Although much remains to be done in this field (Lozano 1998; Hutchings *et al.* 2003), research shows that poisoned or parasitised animals can specifically select foods containing non-nutrient compounds that help them to recover. Might they also select diets with a nutritional composition that neutralises toxins or reduces parasite load and associated disease? In addressing this question for parasitic infection, Hutchings *et al.* (2003) discussed several experiments that have shown a link between diet and immune responses. Lee *et al.* (2006a) found that caterpillars exposed to a highly virulent nucleopolyhedrovirus resisted infection better as the proportion of protein in the diet increased (Lee *et al.* 2006a, 2008b). A similar response was reported by Peck *et al.* (1992), who found that mice survived better on diets containing a higher ratio of protein to carbohydrate after being inoculated with *Salmonella typhimurium*. In the study by Lee *et al.* (2006a), uninfected caterpillars performed best on a diet with lower protein concentration than the infected individuals. When allowed to self-select their diet, caterpillars that survived infection increased their relative intake of protein, compared with controls and with caterpillars that died of infection. This experiment demonstrates that these caterpillars are able to combat viral infection by modulating the macronutrient composition of their diet.

Povey *et al.* (2009) extended the study of Lee *et al.* (2006a) to another pathogen, the bacterium *Bacillus subtilis*, with similar results, and also found that components of the immune system of the caterpillars interacted with one another in relation to diet. Elevation of some immune components was associated with a

decline in others. The traditional view of such immune trade-offs is that different components compete for a limiting pool of resources, typically energy. However, as we saw above for lifespan and reproduction, another possibility is that, rather than competing for a single limiting resource, there is something akin to internal 'niche partitioning' among different elements within the immune system, with each component having a different nutritional optimum within a multidimensional nutrient space.

This idea was explored by Cotter *et al.* (2011) in experiments on caterpillars in which individuals were restricted to one of 20 diets varying in the quantity and ratio of protein and carbohydrate. Results indicated that immune traits do in fact exist within an organismal 'ecology', in which each trait has its own specific nutritional requirements. This was shown by plotting response surfaces for different immune traits onto nutrient intake arrays. No one diet simultaneously optimises all immune components, but the opportunity exists for the animal to adjust its food-selection behaviour to attain a nutritional state that supports the balance of immune responses that best resists infection by a particular pathogen.

The other side to ecological immunology, from a nutritional perspective, is that parasites and pathogens rely on the host for provision of resources. Because these organisms may not share the same nutritional requirements as their hosts, there are possibilities for resource competition and manipulation between the different parties (Smith and Holt 1996; Ponton *et al.* 2011a, 2011b). The complexity of the nutritional interactions between hosts and pathogens is made even greater because animals play host to entire communities of commensal and symbiotic microorganisms, which receive their nutrition from the host and in turn contribute essential nutrients (Turnbaugh *et al.* 2009; De Filippo *et al.* 2010; Kau *et al.* 2011). These interactions are ripe for geometric analysis (Ponton *et al.* 2011b).

#### *Toxins and nutrients interact*

We have argued that for many purposes nutrient and non-nutrient components cannot be simply distinguished, because both can be nutritious, medicinal or toxic. On the other hand, there clearly are cases in which nutrients are nutritious and toxins are categorically deleterious – as is true for many defensive compounds (Sotka *et al.* 2009). Even when toxins can be distinguished categorically in this way, it is often the case that their effects can only be understood in the context of the background nutritional milieu. The modes of interaction of nutrients and toxins are diverse, involving intake, digestion and absorption, as well as postabsorptive effects (Slansky 1992; Sotka *et al.* 2009).

Application of the GF to a study of nutrient–toxin interactions shows how complex and important these interactions can be (Simpson and Raubenheimer 2001). Juveniles of *Locusta migratoria* were confined to one of five diets varying in protein to carbohydrate ratio (P : C) and containing either 0, 3.3, 6.7 or 10% tannic acid (TA). The striking result was that locusts were impervious to TA, even at the highest level of 10%, providing that the diet was near balanced in its P : C relative to the intake target. As the diet became either protein- or carbohydrate-biased, locusts were progressively more susceptible to TA; however, the reason why nutritional imbalance affected susceptibility to TA differed according to the direction of nutritional imbalance. As the diet

became more carbohydrate-biased, insects suffered because of the increasingly powerful antifeedant effects of TA. TA mixed in a low P : C food caused locusts to eat so little that they starved to death. By contrast, food intake was not affected on high P : C diets, but instead TA had its toxic effects postingestively. Behmer *et al.* (2002) next explored the interactive effects of tannic acid and macronutrients in a more complex environment, in which two nutritionally complementary foods (one high in carbohydrate, the other high in protein) containing TA were provided along with a third, TA-free food. Providing that the TA-free food was carbohydrate-biased, locusts were able to maintain their macronutrient intake target by mixing this food with the TA-containing high-protein food. However, they abandoned intake target regulation if the TA-free food was protein-biased, because of their aversion to TA-containing high-carbohydrate foods.

#### **Moving targets**

Intake target, rules of compromise and postingestive responses are fundamental, inter-related traits, against which the adequacy of diets and nutritional environments can be calibrated and the consequences of nutritional imbalance assessed. For clarity, we have treated intake and growth targets as static points integrated across a particular period in the life of an animal. In reality they are trajectories that move in time. In the short term, the requirements of the animal change as environmental circumstances impose differing demands for nutrients and energy. At a somewhat longer timescale, targets move as the animal passes through the various stages of its life, from early growth and development to maturity, reproduction and senescence. On an even longer time-scale, nutritional traits are subject to natural selection and move as species evolve to exploit new or changing nutritional environments and to adopt differing life-history strategies. Presaging such evolutionary change in gene frequencies within populations are epigenetic effects, whereby the nutritional experiences of parents influence the behaviour and metabolism of their offspring without requiring changes in gene frequencies.

#### *Moving targets in the short term*

An animal's nutritional requirements will depend on the physiological demands placed upon it and the intake target should move accordingly. Another major influence over nutritional requirements is environmental temperature. Nutrient intake varies as a function of body temperature, as do metabolic rate and the efficiency with which ingested nutrients are utilised (e.g. Angilletta 2009; Miller *et al.* 2009; Coggan *et al.* 2011). In warm-blooded animals the target mixture of nutrients can vary as a function of the energy demands for thermoregulation, as seen in the increased consumption of carbohydrate but not protein in weanling rats kept at 8°C rather than 23°C (Musten *et al.* 1974; Simpson and Raubenheimer 1997). As we have already discussed, yet another change in environmental circumstances that will shift the intake target is the need for animals to meet the nutritional demands associated with fighting pathogens and mitigating the effects of toxins.

Changes in demand for nutrients imposed by activity, temperature, pathogens, toxins or other short-term influences will act via the suite of regulatory mechanisms briefly summarised

above, but over what periods are these changes detected and tracked? In theory an animal with appropriate complementary foods available could adjust its feeding behaviour and food choices mouthful by mouthful to track its moving intake target with very high resolution; but each time the animal shifts between foods it risks attracting unwarranted attention from natural enemies and wastes both time and energy (Houston and Sumida 1985). At the other extreme, an animal might switch just once, maintaining fidelity to one food until it reaches the point where it must swap foods if it is to attain its target integrated over some longer period. However, by following this strategy the animal will suffer the cost of increased time in a state of nutritional imbalance (far from the target trajectory). The optimal switching interval for a real animal under natural conditions should reflect the relative costs of staying on an imbalanced food versus those of moving to find a complementary food (Chambers *et al.* 1997; Simpson *et al.* 2010).

#### *Moving targets in developmental time*

As animals grow, develop, reproduce and grow old, their nutritional requirements change, both in the amount and blend of nutrients needed. For example, young rat pups require protein, carbohydrate and fat to grow, but once they reach puberty their intake of protein declines and carbohydrate and fat dominate the diet (Leibowitz *et al.* 1991). Male and female rats have different intake trajectories, with males consuming more protein as they grow, presumably to sustain their greater size and muscle mass. When females reach maturity, their macronutrient intake varies as a function of reproduction. During pregnancy they ingest more protein than before mating, but not more carbohydrate or fat. When suckling the resulting pups, mothers increase intake of protein and fat but not carbohydrate. After the pups are weaned, the mother reduces her protein intake to pre-mating levels but sustains high fat intake to restore body reserves that have been depleted during reproduction (Simpson and Raubenheimer 1997).

There are two basic mechanisms for tracking developmental shifts in the intake target. Barton Browne (1995) termed these 'demand-mediated' and 'non-demand-mediated' mechanisms. The former are those mechanisms involving dynamic responses to changing nutritional needs. Hence, the animal experiences a developmental change in patterns of metabolism and growth (induced, for example, by environmental or hormonal signals) and the resulting shift in demand for nutrients causes an appropriate change in feeding behaviour. Alternatively, the developmental switch may itself cause the shift in diet that achieves a new intake target without involving nutrient feedbacks, as seen, for example, in female blow flies and mosquitoes, which upon mating start to develop eggs and become responsive to odours from carrion and live animal hosts, respectively (Barton Browne 1995). Attraction to these resources results in ingestion of high-protein food (rotting flesh or fresh blood) that supports egg development.

Recent studies on the fruit fly, *Drosophila melanogaster*, have begun to provide molecular insights into the relationship between demand-mediated and non-demand-mediated mechanisms associated with female reproduction. Ribeiro and Dickson (2010) found that the rapidly induced preference for

protein-rich yeast in female flies after mating results not from depletion of protein reserves as eggs are developed, but as a direct result of a sex peptide that is introduced with the male's seminal fluid during mating and stimulates special sensory neurons in the female's reproductive tract. An additional demand-driven mechanism then modulates how much yeast is eaten, involving TOR/S6 kinase and serotonin signalling pathways in the central nervous system (Ribeiro and Dickson 2010; Vargas *et al.* 2010).

#### *From parents to offspring – epigenetics*

Some changes in the nutritional environment are sufficiently persistent that intake targets, rules of compromise and postingestive responses need to shift across generations. The main mechanism allowing the phenotype to track changing environments over generations is, of course, natural selection acting via changing gene frequencies within populations. However, it has become increasingly appreciated that effects of the nutritional environment experienced by one generation can be passed to subsequent generations without requiring changes in gene frequencies. For example, dietary-induced obesity in mothers and even grandmothers has a direct influence over the metabolism and risk of obesity in offspring (Barker 1998; McMillen and Robinson 2005; Gluckman and Hanson 2006a). Obesity in fathers can, it seems, also affect metabolic function in offspring (Ng *et al.* 2010). That nutritional consequences in one generation can reverberate and become amplified across subsequent generations has profound health and socioeconomic implications (Gluckman *et al.* 2009).

The search for controlling mechanisms of epigenetic effects is well underway. A major focus of research has been on inherited patterns of gene activation, involving processes such as gene methylation, histone modification and involvement of micro-RNAs, but other pathways are also possible, including changes in milk production by lactating mothers and cultural influences. Another focus of interest is the extent to which epigenetic influences represent adaptive developmental responses, or are simply unavoidable changes that do not improve or might even reduce the fitness of the offspring. Responses that are specifically adaptive have been termed 'predictive adaptive responses', to denote the fact that the developmental trajectory is cued to take a course that is adaptive (beneficial) with respect to the predicted state of the future environment. Such responses can, however, lead to problems when there is a mismatch between the predicted future environment and that actually encountered (Gluckman and Hanson 2004, 2006b).

#### *Evolving targets*

As organisms evolve to exploit different nutritional niches or to track changing nutritional environments, their intake targets will evolve to match the diet. As a result, the optimal diet composition will differ substantially among organisms. The optimal diet for an aphid that feeds on sugary plant sap low in nitrogen is not the same as that for a locust that eats whole plant tissues, or a carnivore that eats predominantly meat. Comparative data on intake targets from many species can be used to explore the association between an animal's diet, its life-history and evolutionary past. One example concerns an analysis in which the optimal protein to carbohydrate ratios (P : C) in the larval diets of 117 insect species



were estimated from a combination of geometric experiments and a published compendium of diet recipes used for rearing insects in the laboratory (Simpson and Raubenheimer 1993). These ratios were superimposed upon a phylogenetic tree of the insect groups, to control for shared ancestry when associating diet composition with features of life-history. The strongest pattern to emerge from the comparative analysis was the association between a reduced P:C and the acquisition of endosymbiotic microorganisms. Species with endosymbionts have a capacity to thrive on nitrogen-poor diets, such as phloem sap (aphids), dead wood (borer beetles) and detritus (cockroaches). Indeed, such insects have evolved a mutual association with symbionts precisely because they provide the host with a novel metabolic capability – the upgrading of dietary nitrogen (Douglas 2010). Possessing symbionts allows the host to exploit low-protein resources that are inadequate for other species. The corollary of acquiring this new metabolic capacity is that diets with higher level of protein, although optimal for other species, have become nutritionally imbalanced for species with endosymbionts (Abisgold *et al.* 1994).

#### *Evolving rules of compromise: nutrient specialists and generalists*

Not only is the intake target dynamic over physiological, developmental and evolutionary timescales, so too are rules of compromise. An instructive example comes from a consistent difference found in rules of compromise between host plant specialist and generalist insect herbivores (Raubenheimer and Simpson 1999, 2003; Simpson *et al.* 2002; Lee *et al.* 2002, 2003, 2006b; Raubenheimer and Jones 2006). When rules of compromise for protein and carbohydrate were derived, generalists were consistently more willing to overeat surplus nutrient in nutritionally imbalanced diets than were specialists. A plausible explanation for this difference relates to the degree of nutritional heterogeneity experienced in the environment of specialists and generalists (Raubenheimer and Simpson 1999). A compatible prediction is that generalists should be better able to deal with excesses postingestively. This was confirmed in studies of locusts and caterpillars (Simpson *et al.* 2002; Lee *et al.* 2002, 2003, 2004, 2006b; Raubenheimer and Simpson 2003). In each instance, nutrient-utilisation budgets indicated that generalists were better able than specialists to use excess ingested protein on high-protein, low-carbohydrate diets for energy metabolism, thereby simultaneously reducing the cost of surplus ingested protein and offsetting the carbohydrate deficit (see Lee *et al.* 2003).

#### *Evolving postingestive responses*

An example of how postingestive responses can change over generations in response to a shift in the nutritional environment comes from the work of Warbrick-Smith *et al.* (2006), who reared multiple lines of the diamondback moth, *Plutella xylostella*, for eight generations either on a carbohydrate-rich or a protein-rich diet. The carbohydrate-rich diet comprised either chemically defined artificial food or a high-starch mutant of the plant *Arabidopsis*. Over the eight generations, caterpillars progressively developed the ability to eat excess carbohydrate without laying it down as body fat, a result that provided strong evidence that storing excess fat has fitness costs. Other replicate

lines were reared for eight generations in a protein-rich, carbohydrate-scarce environment, comprising either artificial diet or a low-starch *Arabidopsis* mutant. In contrast to insects reared under a high-carbohydrate environment, these caterpillars developed an increased propensity to store ingested carbohydrate as fat. Female moths also developed a preference for laying their eggs on the low-starch plant, whereas those selected on the high-starch *Arabidopsis* mutant showed no preference.

By becoming less prone to laying down body fat in a high-carbohydrate world, caterpillars could minimise the costs of obesity – but at the risk of starving for want of energy reserves if food became scarce. By contrast, in a low-carbohydrate world, retaining and storing carbohydrates as fat was favoured; but should such a phenotype be placed into a high-carbohydrate world it would be at risk of obesity. It was not known whether these changes in physiology were the result of genetic selection or accumulating epigenetic effects (see above).

#### **From individuals to populations and societies**

So far, we have viewed the world of nutrition from the perspective of individual animals. But individuals interact with one another – in family groups, aggregations, swarms and societies – and many of these interactions involve nutrition. Next we show how an individual's nutritional state can influence the way in which an entire group behaves.

#### *Cannibal crickets and locusts*

Late spring and early summer in western North America regularly sees millions of large, flightless Mormon crickets, *Anabrus simplex*, forming marching bands that extend for up to 10 km in length. Even though Mormon crickets are omnivores with highly catholic food tastes, they do not strip the habitat bare as they go, hence if limitation of some nutritional resource explains mass migration then it must be more specific than edible food *per se*.

Experiments indicated that Mormon crickets in the marching bands were selectively feeding for protein and salt. When crickets were taken from the band and housed with access to plentiful amounts of protein-rich and carbohydrate-rich foods in a simple GF experiment, they initially ate predominantly protein-rich food, but over the next day they mixed a diet that was much more typical of other crickets and grasshoppers, containing protein and carbohydrate in near equal proportions. In other words, the crickets in the marching bands were not obligatorily protein-seeking, but rather they were selectively deprived of protein as a result of local environmental conditions. However, a highly abundant source of lightly salted protein in the midst of a cricket horde is other Mormon crickets. So, is the reason that cricket bands migrate because they go on a forced march for protein and salt, with each insect chasing the moving meal in front and escaping the cannibals behind? This macabre hypothesis makes two predictions: (1) if crickets become less mobile they should stand a higher chance of getting cannibalised, and (2) satiating crickets with protein and salt should reduce cannibalism and marching. Both of these predictions were met in experiments conducted in the field (Simpson *et al.* 2006).

The conclusions from these experiments and others by Sword *et al.* (2005) are that mass migration at the group level emerges as the indirect effect of individual nutritional responses, mediated

via cannibalistic interactions between individuals. As a result, there is a two-fold incentive to move, to escape the cannibal behind and to catch the meal in front, and a Mormon cricket band travels 2 km each day rather than only tens of metres when the crickets are in low-density populations (Lorch *et al.* 2005). The consequence of migration is a greatly increased chance of encountering fresh nutritional resources but, meanwhile, the crickets are 'travelling with their lunch' (Hansen *et al.* 2011). Bazazi *et al.* (2008, 2011) subsequently showed that protein deprivation is also associated with enhanced marching in groups of cannibalistic desert locust nymphs.

#### *Communal nutrition in ants*

Social insect colonies are far more collegial than Mormon crickets and locusts: interactions among individuals somehow coordinate the activities of the entire colony so that it acts as a nutrient-acquiring, -distributing and -digesting 'superorganism' (Hölldobler and Wilson 2009) that is able to meet the disparate needs of all its members. Indeed, it may well be that nutrition has provided a major feature in the evolution of advanced sociality among animals, including the ants, bees and wasps (Hunt and Nalepa 1994; Kamakura 2011).

The complication for social animals such as ants is that a minority of individuals collects the food for the colony, and these foragers have very different nutritional requirements to other members of their colony (Cassill and Tschinkel 1999; Behmer 2009). In particular, the egg-laying queen and the legless larval ants have a much higher need for protein than do workers, but they do not contribute to food collection. A forager ant who responded only to her own needs would collect carbohydrate-rich nectars and exudates in small amounts, since she is already fully grown. If all foragers behaved in this manner, the brood, queen, nurse ants and other non-foraging workers would soon starve to death. How then does the entire colony communicate its needs to the forager ants?

To answer this question, Dussutour and Simpson (2008) set about posing nutritional conundrums for an Australian species, the green-headed ant, *Rhytidoponera metallica*. Results indicated that ant colonies compensate over time by increasing their collection of dilute over concentrated sugar solutions. Initially, ants consume most and recruited in largest numbers to a concentrated sugar solution, as would be expected were ants maximising their rate of sugar acquisition. However, over successive days, the numbers of ants recruited and the volumes collected by each ant reversed, such that the greatest amount was consumed by colonies with the dilute solution and least by those with the concentrated solution. Ants regulated sugar collection more precisely when the number of larvae was increased than when the number of adult workers was doubled, indicating that larvae were the source of nutritional feedback from the colony to the workers.

Dussutour and Simpson (2009) next challenged colonies of green-headed ants with or without larvae to maintain intake of both protein and sugar by giving them choices between two foods varying in the ratio and concentration of protein and carbohydrate. Colonies with larvae adjusted the amounts of each food collected to maintain the ratio and amounts of protein and sugar collected remarkably constant. Colonies without larvae

differed in three respects: (1) they regulated to a lower protein to carbohydrate ratio, (2) they collected less total quantity of nutrients, and (3) they were unable to respond as effectively to dilution of nutrients in the food. Hence, the presence of fast-growing, energy- and protein-demanding larvae changed both the quantity and blend of nutrients required and determined the effectiveness of nutritional regulation.

#### **How does nutrition structure ecosystems?**

As we have seen, the success of individual organisms can be related to their intake of multiple nutrients by mapping measures of performance (lifespan, reproductive output, immunity to disease, and so on) as landscapes onto nutrient intake arrays. And the nutritional responses of individuals can, in turn, fashion the behaviour of groups and societies. We will now take this one step further to consider the consequences of individual nutrition for populations and the assemblages of species that comprise ecological communities. We will argue that trophic dynamics are in large part an emergent property of regulation of nutrient balance, and that such regulation takes place at *all* trophic levels.

#### *From individual fitness to population growth rates*

If the nutritional responses of individual organisms represent those of the species, it ought to be possible to use performance landscapes measured from a representative sample of individuals to model the success of an entire population of that species. Ecologists measure the success of a population in terms of its population growth rate (*pgr*). By definition, a population will increase in an environment when *pgr* is greater than zero. Population responses have been described in similar geometric terms to those we have used for individual organisms, most famously by Tilman (1982), who classified population growth rate responses as surfaces in a two-resource space.

While it is undoubtedly the case that population growth rates can be defined within the same multidimensional nutritional spaces used to describe the responses of individual organisms, populations are not the simple sum of their individual parts: members within a population interact with one another and with the rest of their environment. The direct and indirect interactions occurring among individuals, both within and between species, can have profound effects on a population's growth rate and may result in very different responses to diet composition than would be predicted from the study of individual organisms (e.g. Kytö *et al.* 1996). We cannot understand the nature and consequences of such interactions unless we know the differing nutritional needs, priorities and regulatory capacities of the different interacting organisms.

#### *Interactions among organisms and the environment*

The most direct nutritional interaction of all is when one organism attempts to eat another, but the interaction might be ecologically significant even if the prey is not killed. It might, for example, change its subsequent behaviour to avoid the risk of further attack, or the interaction might result in the induction of defensive chemicals and immune responses to prevent or counteract future threats (Karban and Baldwin 1997). Being removed from the environment as a living organism and returned as faeces – or being

changed in behaviour, chemical composition and defensive armoury – may resonate throughout the ecosystem in unexpected ways (Simpson *et al.* 2010).

This is illustrated by the study of Hawlena and Schmitz (2010) on the North American grasshopper *Melanoplus femurrubrum*. This herbivore selects a diet of grasses and herbs and is preyed upon by hunting spiders, such as *Pisuarina mira*. Hawlena and Schmitz housed grasshoppers in the presence of spiders, which were kept in clear plastic cylinders where they were visible but unable to attack the grasshoppers. Compared with controls that were not exposed to spiders, the grasshoppers selected a diet containing 40% more carbohydrate, whereas protein intake was not affected by the presence of spiders. This shift in the intake target towards a higher carbohydrate to protein ratio was due to grasshoppers exposed to spiders having an increased metabolic rate: the ‘fear effect’. There was also a change in the chemical composition of the grasshopper’s faeces, which were lower in carbon to nitrogen (C:N) ratio when spiders were around, reflecting the higher metabolic need for carbohydrate. Finally, the body composition of spider-exposed grasshoppers differed, being higher in C:N than controls. Hawlena and Schmitz calculated that the change in the grasshoppers’ intake target when at risk of spider predation would shift the structure of the natural host plant community as a result of selective foraging by fearful grasshoppers. The changes in body and faecal chemistry and plant community structure would, in turn, affect the chemical composition of the pool of detritus in the ecosystem.

Trophic interactions may also act over longer timeframes. For example, any characteristic that influences an organism’s susceptibility to being consumed is likely to be under strong genetic selection, which, over evolutionary time, will allow such genetic influences to affect other trophic levels (Shuster *et al.* 2006). Traits that are heritable can result in patchy nutritional environments at multiple scales. For instance, foliar concentrations of plant secondary metabolites that are toxic to mammals are genetically determined in *Eucalyptus* species. Because of the spatial distribution of different genotypes of trees, herbivorous marsupials such as possums and gliders must move over distances greater than 40 m to encounter significant variation in plant secondary chemistry within a eucalypt forest (Andrew *et al.* 2007).

#### *Do predators regulate nutrient intake?*

Understanding nutritional interactions and their effects requires that we know the differing nutritional requirements, priorities and regulatory abilities of the various interacting organisms. Herbivores and omnivores, including species of insects, birds and mammals, have been shown using GF designs to regulate their intake of macronutrients and some micronutrients and to make postingestive adjustments to help attain the optimal balance of nutrients to meet their various requirements. In contrast, the prevailing view has been that predators do not need to practice nutrient balancing and are unlikely to possess mechanisms to do so. Recent work has shown that this view is incorrect, with evidence for separate regulation of protein and lipid intake coming from invertebrate predators (Mayntz *et al.* 2005; Raubenheimer *et al.* 2007; Jensen *et al.* 2011, 2012), mink (Mayntz *et al.* 2009), predatory fish (Sánchez-Vázquez *et al.*

1999; Rubio *et al.* 2003), and domestic cats (Hewson-Hughes *et al.* 2011).

#### *The nutritional geometry of food webs*

Most theories for explaining food web dynamics have been based on fluxes of mass and energy between consumed and consumers (but see DeAngelis *et al.* 1989; Sterner *et al.* 1996; Sterner and Elser 2002). We have shown several times already in this review that unidimensional models are insufficient to capture the essential features of nutritional systems, and we suspect the same is true for trophic ecology. We would argue that an important aspect of the animal that needs to be represented in models of food webs is the concept of nutritional targets – intake, nutrient and growth targets. These are fundamental in ecological interactions because targets provide a good deal of predictability about the nutritional and foraging decisions of animals. We believe that incorporating the target concept into models of ecological communities can, similarly, help to predict and understand the fluxes of nutrients in ecosystems (Raubenheimer *et al.* 2009).

As trophic levels are ascended, regulatory feeding and growth responses will progressively narrow the range of food compositions and, more importantly, shift the mean composition towards a higher proportion of protein (Raubenheimer *et al.* 2009). This predicted pattern is supported by data on invertebrates showing that the percentage of body nitrogen increases as trophic levels are ascended, whereas the ratio of carbon to nitrogen (C:N) in food versus the body of consumers (C:N food/C:N consumer) narrows progressively (Denno and Fagan 2003). Denno and Fagan (Denno and Fagan 2003; Fagan and Denno 2004) concluded from these trends in body elemental composition that carnivorous arthropods are nitrogen (protein) limited. This, they suggested, explains the tendency for many predators to demonstrate *trophic omnivory* – to feed not only on herbivores, but also on other carnivores (which, as we have seen, tend to have higher nitrogen concentration). However, taking into account the respiratory needs of carnivores we would argue that the reverse is true: consumers become progressively non-protein energy-limited as trophic levels are ascended. This will be especially, but not exclusively, the case when body fat stores need to be increased to meet the demands of periods of starvation, hibernation, reproduction or migration (Raubenheimer *et al.* 2007). It is this energy shortage that motivates feeding back down the food chain (Raubenheimer *et al.* 2009) and perhaps explains why populations of predatory marine birds and sea lions declined in the Gulf of Alaska after the loss of high-fat species of fish (Rosen and Trites 2000; Romano *et al.* 2006). Recently, Wilder and Eubanks (2010) have reiterated and elaborated upon the logic of this argument.

#### *The nutritional niche*

We have seen that it may not be possible to predict the growth rate of a population or species from the nutritional responses of individuals, because of direct and indirect interactions with other organisms within food webs. To use the terminology of niche theory, the fundamental nutritional niche of a population may not be realised (Kearney 2006).

The GF shares fundamental features with Hutchinson's (1957) niche concept, focusing on the resources component of a more inclusive Hutchinsonian environmental space. To broaden the power of GF models to predict the performance, population dynamics and distribution of species, it will be necessary to incorporate abiotic influences as extra dimensions and to transpose this niche space onto real environments in space and time (Kearney *et al.* 2010). Effective models in nutritional ecology must, therefore, not only be 'nutritionally explicit' (able to express a situation in terms of nutrition), but also 'organismally explicit' and 'ecologically explicit' (able to incorporate organism-level and ecological factors in the model) (Raubenheimer *et al.* 2009). We now consider some ways in which the GF can be made more ecologically explicit, by incorporating abiotic and biotic aspects of the ecological environment, so enhancing the capacity of this approach to integrate nutrition into ecosystem-level analyses.

#### *Incorporating abiotic factors*

In addition to food resources, Hutchinson's niche model includes dimensions for abiotic factors such as temperature, light, humidity and pH. A research field known as 'Biophysical Ecology' has developed to represent the 'climatic' niches of organisms in 'climate space' (Porter and Gates 1969). These models define the combinations of environmental variables that are suitable for survival and reproduction for a species, and have been applied at the scale of landscapes to predict the distribution, temperature, energy and water relations of organisms (Kearney and Porter 2009).

Abiotic factors can shape nutritional interactions in two major ways. The first is through influences on the physiology and behaviour of organisms. For example, changes in body temperature alter metabolic rate, rates of water loss, amounts and ratios of nutrients required, and tolerance to toxins and diseases. Hence, a decrease in ambient temperature results in rats increasing carbohydrate or fat consumption but not changing their protein intake (Aubert *et al.* 1995), and changes in temperature affect tolerance of white-throated woodrats to plant secondary metabolites (Dearing *et al.* 2008). Miller *et al.* (2009) showed that locusts developed faster but converted protein and carbohydrate less efficiently to growth if kept at 38°C rather than at 32°C. If given a choice of temperatures under conditions of food abundance, locusts selected 38°C, therefore prioritising development rate over utilisation efficiency. In a subsequent experiment, Coggan *et al.* (2011) allowed recently fed locusts to select a temperature within a thermal gradient, and after 50 min (the average intermeal interval) they either fed the locusts again or left them without food. Locusts initially selected a temperature of 38°C and returned to that temperature after having been fed again. However, if they were not fed at 50 min, they soon moved to a cooler region of the gradient, 32°C; they were now prioritising efficient extraction of nutrients in the previous meal over rapid growth in response to the absence of food.

The second way in which abiotic factors influence nutritional interactions is through the distribution of environmental conditions in space and time. Environmental gradients, concentrations and ratios of chemical elements in soil or water, microclimatic conditions and other habitat features will shape

patterns of food availability over multiple spatial and time scales and will interact with an animal's size, mobility and regulatory behaviour (Pincebourde and Casas 2006; Kearney and Porter 2009).

#### *Dynamic Energy Budget theory as a computational engine*

Kearney *et al.* (2010) proposed that Dynamic Energy Budget (DEB) theory (Sousa *et al.* 2008; Kooijman 2009) could be used as a 'computational engine' for integrating the GF and Biophysical Ecology, allowing population responses to be modelled dynamically within environmental space. A special type of DEB model is needed to integrate DEB theory with GF – one that includes a separate reserve for each nutritional component (Kearney *et al.* 2010). An example of how a two-currency DEB model could be used as a computational engine for implementing GF designs to predict population responses is shown in Kearney *et al.* (2010), who applied a DEB model developed by Kuyper *et al.* (2004) to calculate egg production in a copepod as a function of ingested carbohydrate and protein. DEB could generate at the level of populations (in the absence of other biological interactions) the types of responses seen in GF studies on individuals.

#### *Agent-based modelling of nutritional interactions: from individuals to ecosystems*

An appropriately defined DEB model offers a means of implementing GF models and calculating the fundamental niche of an organism, but the problem of dealing with interactions among organisms – of calculating the realised niche – remains. Since interactions among individual organisms are the most fundamental units from which ecosystems are built, we have argued that agent-based models may provide a means of scaling from individuals to ecosystems (Simpson *et al.* 2010). Agent-based models (ABMs) have been used to explore patterns of resource exploitation in foraging animals (Oom *et al.* 2004; Grimm and Railsback 2005; Nonaka and Holme 2007), but they have yet to take account of multiple nutrient dimensions; in other words, they are not yet nutritionally explicit (Raubenheimer *et al.* 2009; Simpson *et al.* 2010). However, if ABMs were to incorporate aspects of GF models, they would offer the promise of dynamic, spatially explicit models of nutritional interactions, which could take account of an individual organism's simultaneous membership of a group, population, community and ecosystem. From such 'heterarchical' models (Raubenheimer *et al.* 2009) may well emerge step-shifts in organisational complexity, such as have been observed already at the level of groups and populations.

Much of the information required for such a model already exists and other data could readily be collected (Simpson *et al.* 2010). The aim in future will be to use such a platform to explore the evolution of nutritional phenotypes, to model populations, and to help explain the structure and dynamics of species assemblages within changing environments. If successful, the outcome would be a synthesis of nutritional ecology that is dynamic as well as nutritionally, organismally, ecologically and spatially explicit.

### Wildlife conservation

We will end with a consideration of the role that GF can play in wildlife conservation. A challenge arises in conservation ecology when the natural habitat of wild animals is diminished or altered, and decisions need to be made about providing a nutritional environment that will sustain the species. In some cases this will involve setting priorities for conserving remnants of the natural habitat, or identifying suitable habitats for establishing translocated populations. In other, usually more extreme, cases the natural diet is not available in sufficient quantities, and therefore needs to be supplemented or replaced by alternative foods. We now provide an example where the GF has been applied in the former scenario. For an example of the latter, see Raubenheimer and Simpson (2006).

At one level, it might be argued that there is no need to perform nutrient-level analyses of the foraging habitats of wildlife, and even less so the nutritional regulatory responses of the target species. In this line of thinking animals eat foods, are pretty good at knowing what they want and how much to eat, and as long as management decisions are geared towards conserving sufficient quantities of the relevant foods, then from the nutritional point of view the job of the conservationist is complete. This logic might well work in situations where the evolved link between the distribution of foods in the environment and the animal's nutritional regulatory system remains intact. However, where this is not the case – for example, where the habitat in which the regulatory systems evolved is diminished or altered – then there might be a mismatch between the regulatory systems and the altered environment in which they need to operate. To understand, avoid or manage this mismatch, we need to think in terms of foods as well as their nutrient and other components.

A recent example where nutritional geometry was used to understand the habitat requirements of a priority conservation species was the study of Peruvian spider monkeys, *Ateles chamek*, in their natural forested habitat in Bolivia (Felton *et al.* 2009a, 2009b). Felton first spent five months habituating the monkeys. When they were sufficiently comfortable with her presence, she followed individual monkeys throughout their feeding day (from dawn to dusk), recording everything they ate during the 12-h observation period and estimating the amount of each food that was eaten. Representative samples of each food item were collected, and later taken back to the laboratory and their nutritional compositions analysed. From these data, 38 daily intakes of protein, fat and carbohydrate were estimated and analysed using the GF.

Several interesting results emerged. First, although primarily frugivorous, the monkeys ate a range of foods that differed widely in their macronutrient composition. When available, they specifically targeted ripe figs, *Ficus* spp., but when these were not available in sufficient quantities they mixed their intake from foods with high and low protein : non-protein energy balances, thereby moving in a zig-zag pattern through nutrient space. Interestingly, the macronutrient balance of the daily diet when eating figs or composing the diet from diverse foods did not differ. This suggests that figs are a nutritionally balanced food for the monkeys, and when they are not available the monkeys compose a balanced diet through mixing nutritionally complementary foods.

Another pattern of feeding was observed during the late wet season, when ripe figs were scarce but other fruits, particularly

high in sugars and fats, were abundant. In this period, the monkeys ate very similar amounts of protein as when eating figs or diverse complementary foods (i.e. this most likely being the target level of protein), but in so doing ate considerably more carbohydrates and fat than observed when eating figs or mixing complementary foods. The resulting intake array was thus a vertical line, with little variation on the protein axis compared with the axis for non-protein. This is very similar to the rule of compromise that has been observed in experimental studies of humans (Simpson and Raubenheimer 2005; Gosby *et al.* 2011), which appear to be an important contributory factor to the epidemic of obesity that has swept many countries in recent decades.

The study of Felton *et al.* (2009a, 2009b) has several implications for understanding the habitat requirements of Peruvian spider monkeys. First, the results provide an estimate of the position of the macronutrient intake target of monkeys, and suggest that figs (*Ficus* spp.) are nutritionally balanced with respect to the macronutrient requirements of these monkeys. This underscores the importance of regulating the commercial harvesting of *Ficus* timber in the habitat of spider monkeys. Second, the study suggests that the monkeys are not reliant on balanced foods, but can compose a balanced diet through mixing nutritionally complementary foods. This shows that the monkeys are to some extent flexible in their food needs, and also provides an indication of which combinations of foods are important components of their nutritional environment. Third, when exposed to foods with a low protein : non-protein energy balance spider monkeys will defend their protein intake but overeat non-protein energy. This helps us to predict how these monkeys would respond to distortions in the macronutrient composition of their foraging environment. Specifically, it suggests that prolonged exposure to foods with a low protein : non-protein energy balance is unlikely to result in protein deficit, but rather a surplus of non-protein energy and, conversely, these monkeys are unlikely to overeat protein to meet their requirements for non-protein energy.

It remains to be determined, however, whether and how being constrained to suffer macronutrient imbalance would impact on the monkeys. It is easy to imagine a situation in which energy shortage, as might be expected to result from high-protein diets, can be a limiting factor to wild primates. But might a sustained energy surplus, as predicted on low-protein diets, also impact adversely on spider monkeys? Indirect evidence raises a cautionary note. Like their human cousins, non-human primates are prone to obesity under certain conditions, which include exposure to readily-available foods high in non-protein energy (Hansen 2001). While this is most common in captive primates (e.g. in zoos), significant levels of obesity have also been observed in free-ranging populations of baboons (Altmann *et al.* 1993) and macaque monkeys on the island of Cayo Santiago off the coast of Puerto Rico (Schwartz *et al.* 1993). In both cases this was associated not with natural foods, but with access to garbage dumps (baboons) and the management practice of supplementing the natural diet with primate chow (macaques). At the very least, this warrants careful consideration of the food species that should be conserved in the habitat of spider monkeys, and provides information regarding the required macronutrient composition of any supplementary or captive diets for this species.

## Concluding remarks

We began with the assertions that nutrients are the connecting threads in the web of life, and that new approaches are needed for understanding the nutritional networks that comprise and link organisms. We then set out to demonstrate how geometric analysis (the Geometric Framework) can contribute by selectively reducing the pall of nutritional complexity to those interactions that help to illuminate the problem at hand. In so doing, the Geometric Framework can help to integrate nutrition research within a framework that encompasses the different levels of biological organisation from genes to ecosystems, spans taxa from slime moulds to humans, and offers the promise of practical solutions to pressing problems in agriculture, human health and conservation ecology. The examples we have presented represent a few superficial scratches to the surface of a very large world of unresolved issues in nutritional biology.

## Acknowledgements

We are grateful to Princeton University Press for their permission to base this Perspective on the text of our book, *The Nature of Nutrition: A unifying framework from animal adaptation to human obesity* (Simpson and Raubenheimer 2012).

## References

- Abisgold, J. D., Simpson, S. J., and Douglas, A. E. (1994). Nutrient regulation in the pea aphid *Acyrtosiphon pisum*: application of a novel geometric framework to sugar and amino acid consumption. *Physiological Entomology* **19**, 95–102. doi:10.1111/j.1365-3032.1994.tb01081.x
- Altmann, J., Schoeller, D., Altmann, S. A., Muruthi, P., and Sapolsky, R. M. (1993). Body size and fatness of free-living baboons reflect food availability and activity levels. *American Journal of Primatology* **30**, 149–161. doi:10.1002/ajp.1350300207
- Andrew, R., Wallis, I. R., Harwood, C. E., Henson, M., and Foley, W. J. (2007). Heritable variation in the foliar secondary metabolite sideroxylonal in *Eucalyptus* confers cross-resistance to herbivores. *Oecologia* **153**, 891–901. doi:10.1007/s00442-007-0784-1
- Angilletta, M. J. (2009). 'Thermal Adaptation: a Theoretical and Empirical Synthesis.' (Oxford University Press: Oxford.)
- Archer, C. R., Royle, N., South, S., Selman, C., and Hunt, J. (2009). Nutritional geometry provides food for thought. *The Journals of Gerontology. Series A, Biological Sciences and Medical Sciences* **64A**, 956–959. doi:10.1093/gerona/glp069
- Aubert, A., Goodall, G., and Dantzer, R. (1995). Compared effects of cold ambient temperature and cytokines on macronutrient intake in rats. *Physiology & Behavior* **57**, 869–873. doi:10.1016/0031-9384(94)00391-H
- Barker, D. J. P. (1998). 'Mothers, Babies and Health in Later Life.' 2nd edn. (Churchill Livingstone: London.)
- Barton Browne, L. (1995). Ontogenetic changes in feeding behavior. In 'Regulatory Mechanisms in Insect Feeding'. (Eds R. F. Chapman and G. de Boer.) pp. 307–342. (Chapman and Hall: New York.)
- Bazazi, S., Buhl, J., Hale, J. J., Anstey, M. L., Sword, G. A., Simpson, S. J., and Couzin, I. D. (2008). Collective motion and cannibalism in locust migratory bands. *Current Biology* **18**, 735–739. doi:10.1016/j.cub.2008.04.035
- Bazazi, S., Romanczuk, P., Thomas, S., Schimansky-Geier, L., Hale, J. J., Miller, G. A., Sword, G. A., Simpson, S. J., and Couzin, I. D. (2011). Nutritional state and collective motion: from individuals to mass migration. *Proceedings. Biological Sciences* **278**, 356–363. doi:10.1098/rspb.2010.1447
- Behmer, S. T. (2009). Insect herbivore nutrient regulation. *Annual Review of Entomology* **54**, 165–187. doi:10.1146/annurev.ento.54.110807.090537
- Behmer, S. T., Simpson, S. J., and Raubenheimer, D. (2002). Herbivore foraging strategies in chemically heterogeneous environments: allelochemical–nutrient interactions. *Ecology* **83**, 2489–2501. doi:10.1890/0012-9658(2002)083[2489:HFICHE]2.0.CO;2
- Berenbaum, M. R. (1995). Turnabout is fair play: secondary roles for primary compounds. *Journal of Chemical Ecology* **21**, 925–940. doi:10.1007/BF02033799
- Bernays, E. A. (1993). Food aversion learning. In 'Insect Learning'. (Eds A. C. Lewis and D. Papaj.) pp. 1–17. (Chapman and Hall: New York.)
- Boersma, M., and Elser, J. J. (2006). Too much of a good thing: on stoichiometrically balanced diets and maximal growth. *Ecology* **87**, 1325–1330. doi:10.1890/0012-9658(2006)87[1325:TMOAGT]2.0.CO;2
- Booth, D. A., and Thibault, L. (2000). Macronutrient-specific hungers and satieties and their neural bases, learnt from pre- and postingestinal effects of eating particular foodstuffs. In 'Neural and Metabolic Control of Macronutrient Intake'. (Eds H.-R. Berthoud and R. J. Seeley.) pp. 61–91. (CRC Press: Boca Raton, FL.)
- Burke, C. J., and Waddell, S. (2011). Remembering nutrient quality of sugar in *Drosophila*. *Current Biology* **21**, 746–750. doi:10.1016/j.cub.2011.03.032
- Calabrese, E. J. (2005). Paradigm lost, paradigm found: the re-emergence of hormesis as a fundamental dose response model in the toxicological sciences. *Environmental Pollution* **138**, 379–412. doi:10.1016/j.envpol.2004.10.001
- Calabrese, E. J., and Baldwin, L. A. (2003). Toxicology rethinks its central belief – hormesis demands a reappraisal of the way risks are assessed. *Nature* **421**, 691–692. doi:10.1038/421691a
- Calabrese, E. J., Baldwin, L. A., and Holland, C. D. (1999). Hormesis: a highly generalizable and reproducible phenomenon with important implications for risk assessment. *Risk Analysis* **19**, 261–281. doi:10.1111/j.1539-6924.1999.tb00404.x
- Carey, J. R., Harshman, L. G., Liedo, P., Müller, H.-G., Wang, J.-L., and Zhang, Z. (2008). Longevity–fertility trade-offs in the tephritid fruit fly, *Anastrepha ludens*, across dietary-restriction gradients. *Aging Cell* **7**, 470–477. doi:10.1111/j.1474-9726.2008.00389.x
- Carleton, A., Accolla, R., and Simon, S. A. (2010). Coding in the mammalian gustatory system. *Trends in Neurosciences* **33**, 326–334. doi:10.1016/j.tins.2010.04.002
- Cassill, D. L., and Tschinkel, W. R. (1999). Regulation of diet in the fire ant, *Solenopsis invicta*. *Journal of Insect Behavior* **12**, 307–328. doi:10.1023/A:1020835304713
- Chambers, P. G., Simpson, S. J., and Raubenheimer, D. (1995). Behavioural mechanisms of nutrient balancing in *Locusta migratoria*. *Animal Behaviour* **50**, 1513–1523. doi:10.1016/0003-3472(95)80007-7
- Chambers, P. G., Simpson, S. J., and Raubenheimer, D. (1997). The functional significance of switching interval in food mixing by *Locusta migratoria*. *Journal of Insect Physiology* **44**, 77–85. doi:10.1016/S0022-1910(97)00085-1
- Clissold, F. J., Tedder, B. J., Conigrave, A. D., and Simpson, S. J. (2010). The gastrointestinal tract as a nutrient balancing organ. *Proceedings. Biological Sciences* **277**, 1751–1759. doi:10.1098/rspb.2009.2045
- Coggan, N., Clissold, F. J., and Simpson, S. J. (2011). Locusts use dynamic thermoregulatory behaviour to optimise nutritional outcomes. *Proceedings. Biological Sciences* **278**, 2745–2752. doi:10.1098/rspb.2010.2675
- Colman, R. J., Anderson, R. M., Johnson, S. C., Kastman, E. K., Kosmatka, K. J., Beasley, T. M., Allison, D. B., Cruzen, C., Simmons, H. A., Kennnitz, J. W., and Weindruch, R. (2009). Caloric restriction delays disease onset and mortality in rhesus monkeys. *Science* **325**, 201–204. doi:10.1126/science.1173635

- Cook, S. C., Eubanks, M. D., Gold, R. E., and Behmer, S. T. (2010). Colony-level macronutrient regulation in ants: mechanisms, hoarding and associated costs. *Animal Behaviour* **79**, 429–437. doi:10.1016/j.anbehav.2009.11.022
- Cota, D., Proulx, K., and Seeley, R. J. (2007). The role of CNS fuel sensing in energy and glucose regulation. *Gastroenterology* **132**, 2158–2168. doi:10.1053/j.gastro.2007.03.049
- Cotter, S. C., Simpson, S. J., Raubenheimer, D., and Wilson, K. (2011). Macronutrient balance mediates trade-offs between immune function and life history traits. *Functional Ecology* **25**, 186–198. doi:10.1111/j.1365-2435.2010.01766.x
- Crespi, E. J., and Denver, R. J. (2006). Leptin (*ob* gene) of the South African clawed frog *Xenopus laevis*. *Proceedings of the National Academy of Sciences of the United States of America* **103**, 10092–10097. doi:10.1073/pnas.0507519103
- Cypess, A. M., Lehman, S., Williams, G., Tal, I., Rodman, D., Goldfine, A. B., Kuo, F. C., Palmer, E. L., Tseng, Y.-H., Doria, A., Kolodny, G. M., and Kahn, C. R. (2009). Identification and importance of brown adipose tissue in adult humans. *The New England Journal of Medicine* **360**, 1509–1517. doi:10.1056/NEJMoa0810780
- De Filippo, C., Cavalieri, D., Di Paola, M., Ramazzotti, M., Poullet, J. B., Massart, S., Collini, S., Pieraccini, G., and Lionetti, P. (2010). Impact of diet in shaping gut microbiota revealed by a comparative study in children from Europe and rural Africa. *Proceedings of the National Academy of Sciences of the United States of America* **107**, 14691–14696. doi:10.1073/pnas.1005963107
- DeAngelis, D. L., Mulholland, P. J., Palumbo, A. V., Steinmen, A. D., Huston, M. A., and Elwood, J. W. (1989). Nutrient dynamics and food-web stability. *Annual Review of Ecology and Systematics* **20**, 71–95. doi:10.1146/annurev.es.20.110189.000443
- Dearing, M. D., Forbey, J. S., McLister, J. D., and Santos, L. (2008). Ambient temperature influences diet selection and physiology of an herbivorous mammal, *Neotoma albigula*. *Physiological and Biochemical Zoology* **81**, 891–897. doi:10.1086/588490
- Denno, R. F., and Fagan, W. F. (2003). Might nitrogen limitation promote omnivory among carnivorous arthropods? *Ecology* **84**, 2522–2531. doi:10.1890/02-0370
- Dethier, V. G. (1976). 'The Hungry Fly.' (Princeton University Press: Princeton, NJ)
- Dong, D., Jones, G., and Zhang, S. (2009). Dynamic evolution of bitter taste receptor genes in vertebrates. *BMC Evolutionary Biology* **9**, 1–9. doi:10.1186/1471-2148-9-12
- Douglas, A. E. (2010). 'The Symbiotic Habit.' (Princeton University Press: Princeton, NJ)
- Dukas, R. (2008). Evolutionary biology of insect learning. *Annual Review of Entomology* **53**, 145–160. doi:10.1146/annurev.ento.53.103106.093343
- Dussutour, A., and Simpson, S. J. (2008). Carbohydrate regulation in relation to colony growth in ants. *The Journal of Experimental Biology* **211**, 2224–2232. doi:10.1242/jeb.017509
- Dussutour, A., and Simpson, S. J. (2009). Communal nutrition in ants. *Current Biology* **19**, 740–744. doi:10.1016/j.cub.2009.03.015
- Everitt, A. V., Rattan, S., Le Couteur, D., and de Cabo, R. (Eds) (2010). 'Calorie Restriction, Aging, and Longevity.' (Springer: Dordrecht.)
- Fagan, W. F., and Denno, R. F. (2004). Stoichiometry of actual vs. potential predator–prey interactions: insights into nitrogen limitation for arthropod predators. *Ecology Letters* **7**, 876–883. doi:10.1111/j.1461-0248.2004.00641.x
- Fanson, B. G., and Taylor, P. W. (2011). Protein: carbohydrate ratios explain lifespan patterns found in Queensland fruit fly on diets varying in yeast : sugar ratios. *Age*. doi:10.1007/s11357-011-9308-3
- Fanson, B. G., Weldon, C. W., Pérez-Staples, D., Simpson, S. J., and Taylor, P. W. (2009). Nutrients, not caloric restriction, extend lifespan in Queensland fruit flies (*Bactrocera tryoni*). *Aging Cell* **8**, 514–523. doi:10.1111/j.1474-9726.2009.00497.x
- Felton, A. M., Felton, A., Raubenheimer, D., Simpson, S. J., Foley, W. J., Wood, J. T., Wallis, I. R., and Lindenmayer, D. B. (2009a). Protein content of diets dictates the daily energy intake of a free-ranging primate. *Behavioral Ecology* **20**, 685–690. doi:10.1093/beheco/arp021
- Felton, A. M., Felton, A., Wood, J. T., Foley, W. J., Raubenheimer, D., Wallis, I. R., and Lindenmayer, D. B. (2009b). Nutritional ecology of *Ateles chamek* in lowland Bolivia: how macronutrient balancing influences food choices. *International Journal of Primatology* **30**, 675–696. doi:10.1007/s10764-009-9367-9
- Finger, T. E. (1997). Evolution of taste and solitary chemoreceptor cell systems. *Brain, Behavior and Evolution* **50**, 234–243. doi:10.1159/000113337
- Flatt, T. (2011). Survival costs of reproduction in *Drosophila*. *Experimental Gerontology* **46**, 369–375. doi:10.1016/j.exger.2010.10.008
- Fujita, M., and Tanimura, T. (2011). *Drosophila* evaluates and learns the nutritional value of sugars. *Current Biology* **21**, 751–755. doi:10.1016/j.cub.2011.03.058
- Galef, B. G. Jr. and Laland, K. N. (2005). Social learning in animals: empirical studies and theoretical models. *Bioscience* **55**, 489–499. doi:10.1641/0006-3568(2005)055[0489:SLIAES]2.0.CO;2
- Gluckman, P. D., and Hanson, M. A. (2004). The developmental origins of the metabolic syndrome. *Trends in Endocrinology and Metabolism* **15**, 183–187. doi:10.1016/j.tem.2004.03.002
- Gluckman, P. D., and Hanson, M. A. (Eds) (2006a). 'Developmental Origins of Health and Disease.' (Cambridge University Press: Cambridge.)
- Gluckman, P. D., and Hanson, M. A. (2006b). 'Mismatch: why Our World No Longer Fits Our Bodies.' (Oxford University Press: Oxford.)
- Gluckman, P. D., Hanson, M. A., Bateson, P., Beedle, A. S., Law, C. M., Bhutta, Z. A., Anokhin, K. V., Bougnères, P., Chandak, G. R., Dasgupta, P., Smith, G. D., Ellison, P. T., Forrester, T., Gilbert, S. F., Jablonka, E., Kaplan, H., Prentice, A. M., Simpson, S. J., Uauy, R., and West-Eberhard, M. J. (2009). Towards a new developmental synthesis: adaptive developmental plasticity and human disease. *Lancet* **373**, 1654–1657. doi:10.1016/S0140-6736(09)60234-8
- Gosby, A. K., Conigrave, A. D., Lau, N. S., Iglesias, M. A., Hall, R. M., Jebb, S. A., Brand-Miller, J., Caterson, I. D., Raubenheimer, D., and Simpson, S. J. (2011). Testing protein leverage in lean humans: a randomised controlled experimental study. *PLoS ONE* **6**, e25929. doi:10.1371/journal.pone.0025929
- Grandin, R. C., Piper, M. D. W., and Partridge, L. (2009). Amino-acid imbalance explains extension of lifespan by dietary restriction in *Drosophila*. *Nature* **462**, 1061–1064. doi:10.1038/nature08619
- Grimm, V., and Railsback, S. F. (2005). 'Individual-based Modeling and Ecology.' (Princeton University Press: Princeton, NJ.)
- Hansen, B. C. (2001). Causes of obesity and consequences of obesity prevention in non-human primates and other animals. In 'International Textbook of Obesity'. (Ed. P. Björntorp.) pp. 181–201. (John Wiley and Sons: Chichester, UK.)
- Hansen, M. J., Buhl, J., Bazazi, S., Simpson, S. J., and Sword, G. A. (2011). Cannibalism in the lifeboat – collective movement in Australian plague locusts. *Behavioral Ecology and Sociobiology* **65**, 1715–1720. doi:10.1007/s00265-011-1179-1
- Hawlena, D., and Schmitz, O. J. (2010). Herbivore physiological response to predation risk and implications for ecosystem nutrient dynamics. *Proceedings of the National Academy of Sciences of the United States of America* **107**, 15503–15507. doi:10.1073/pnas.1009300107
- Hayes, D. P. (2007). Nutritional hormesis. *European Journal of Clinical Nutrition* **61**, 147–159. doi:10.1038/sj.ejcn.1602507

- Hewson-Hughes, A. K., Hewson-Hughes, V. L., Miller, A. T., Hall, S. R., Simpson, S. J., and Raubenheimer, D. (2011). Geometric analysis of macronutrient selection in the adult domestic cat, *Felis catus*. *The Journal of Experimental Biology* **214**, 1039–1051. doi:10.1242/jeb.049429
- Hölldobler, B., and Wilson, E. O. (2009). 'The Superorganism: The Beauty, Elegance, and Strangeness of Insect Societies.' (W.W. Norton & Co.: New York.)
- Houston, A., and Sumida, B. (1985). A positive feedback model for switching between two activities. *Animal Behaviour* **33**, 315–325. doi:10.1016/S0003-3472(85)80145-7
- Huffman, M. A. (2001). Self-medicative behavior in the African great apes: an evolutionary perspective into the origins of human traditional medicine. *Bioscience* **51**, 651–661. doi:10.1641/0006-3568(2001)051[0651:SMBITA]2.0.CO;2
- Huffman, M. A. (2003). Animal self-medication and ethno-medicine: exploration and exploitation of the medicinal properties of plants. *The Proceedings of the Nutrition Society* **62**, 371–381. doi:10.1079/PNS2003257
- Hunt, J. H., and Nalepa, C. A. (Eds) (1994). 'Nourishment and Evolution in Insect Societies.' (Westview Press: Boulder, CO.)
- Hutchings, M. R., Athanasiadou, S., Kyriazakis, I., and Gordon, I. J. (2003). Can animals use foraging behaviour to combat parasites? *The Proceedings of the Nutrition Society* **62**, 361–370. doi:10.1079/PNS2003243
- Hutchinson, G. E. (1957). Concluding remarks. *Cold Spring Harbor Symposia on Quantitative Biology* **22**, 415–427. doi:10.1101/SQB.1957.022.01.039
- Ja, W. W., Carvalho, G. B., Zid, B. M., Mak, E. M., Brummel, T., and Benzer, S. (2009). Water- and nutrient-dependent effects of dietary restriction on *Drosophila* lifespan. *Proceedings of the National Academy of Sciences of the United States of America* **106**, 18 633–18 637. doi:10.1073/pnas.0908016106
- Jensen, K., Mayntz, D., Tøft, S., Raubenheimer, D., and Simpson, S. J. (2011). Nutrient regulation in a predator, the wolf spider *Pardosa prativaga*. *Animal Behaviour* **81**, 993–999. doi:10.1016/j.anbehav.2011.01.035
- Jensen, K., Mayntz, D., Tøft, S., Clissold, F. J., Hunt, J., Raubenheimer, D., and Simpson, S. J. (2012). Optimal foraging for specific nutrients in predatory beetles. *Proceedings of the Royal Society B: Biological Sciences*. doi:10.1098/rspb.2011.2410
- Kamakura, M. (2011). Royalactin induces queen differentiation in honeybees. *Nature* **473**, 478–483. doi:10.1038/nature10093
- Karban, R., and Baldwin, I. T. (1997). 'Induced Responses to Herbivory.' (University of Chicago Press: Chicago, IL.)
- Kau, A. L., Ahern, P. P., Griffin, N. W., Goodman, A. L., and Gordon, J. I. (2011). Human nutrition, the gut microbiome and the immune system. *Nature* **474**, 327–336. doi:10.1038/nature10213
- Kearney, M. (2006). Habitat, environment and niche: what are we modelling? *Oikos* **115**, 186–191. doi:10.1111/j.2006.0030-1299.14908.x
- Kearney, M., and Porter, W. P. (2009). Mechanistic niche modelling: combining physiological and spatial data to predict species' ranges. *Ecology Letters* **12**, 334–350. doi:10.1111/j.1461-0248.2008.01277.x
- Kearney, M., Simpson, S. J., Raubenheimer, D., and Helmuth, B. (2010). Modelling the ecological niche from functional traits. *Philosophical Transactions of the Royal Society B: Biological Sciences* **365**, 3469–3483. doi:10.1098/rstb.2010.0034
- Kirkwood, T. B. L. (2005). Understanding the odd science of aging. *Cell* **120**, 437–447. doi:10.1016/j.cell.2005.01.027
- Kooijman, S. A. L. M. (2009). 'Dynamic Energy Budget Theory for Metabolic Organisation.' (Cambridge University Press: Cambridge.)
- Kuijper, L. D. J., Anderson, T. R., and Kooijman, S. A. L. M. (2004). C and N gross growth efficiencies of copepod egg production studied using a dynamic energy budget model. *Journal of Plankton Research* **26**, 213–226. doi:10.1093/plankt/fbh020
- Kytö, M., Niemelä, P., and Larsson, S. (1996). Insects on trees: population and individual response to fertilization. *Oikos* **75**, 148–159. doi:10.2307/3546238
- Le Rohellec, M., and Le Bourg, É. (2009). Contrasted effects of suppressing live yeast from food on longevity, aging and resistance to several stresses in *Drosophila melanogaster*. *Experimental Gerontology* **44**, 695–707. doi:10.1016/j.exger.2009.08.001
- Lee, K. P., Behmer, S. T., Simpson, S. J., and Raubenheimer, D. (2002). A geometric analysis of nutrient regulation in the generalist caterpillar *Spodoptera littoralis* (Boisduval). *Journal of Insect Physiology* **48**, 655–665. doi:10.1016/S0022-1910(02)00088-4
- Lee, K. P., Raubenheimer, D., and Simpson, S. J. (2003). A correlation between nutrient balancing and insect host-plant range: evidence from the specialist caterpillar *Spodoptera exempta*. *Journal of Insect Physiology* **49**, 1161–1171. doi:10.1016/j.jinsphys.2003.08.013
- Lee, K. P., Simpson, S. J., and Raubenheimer, D. (2004). A comparison of nutrient regulation between solitary and gregarious phases of the specialist caterpillar, *Spodoptera exempta* (Walker). *Journal of Insect Physiology* **50**, 1171–1180. doi:10.1016/j.jinsphys.2004.10.009
- Lee, K. P., Cory, J. S., Wilson, K., Raubenheimer, D., and Simpson, S. J. (2006a). Flexible diet choice offsets protein costs of pathogen resistance in a caterpillar. *Proceedings. Biological Sciences* **273**, 823–829. doi:10.1098/rspb.2005.3385
- Lee, K. P., Behmer, S. T., and Simpson, S. J. (2006b). Nutrient regulation in relation to diet breadth: a comparison of *Heliothis* sister species and a hybrid. *The Journal of Experimental Biology* **209**, 2076–2084. doi:10.1242/jeb.02253
- Lee, K. P., Simpson, S. J., Clissold, F. J., Brooks, R., Ballard, J. W. O., Taylor, P. W., Soran, N., and Raubenheimer, D. (2008a). Lifespan and reproduction in *Drosophila*: new insights from nutritional geometry. *Proceedings of the National Academy of Sciences of the United States of America* **105**, 2498–2503. doi:10.1073/pnas.0710787105
- Lee, K. P., Simpson, S. J., and Wilson, K. (2008b). Dietary protein-quality influences melanization and immune function in an insect. *Functional Ecology* **22**, 1052–1061. doi:10.1111/j.1365-2435.2008.01459.x
- Leibowitz, S. F., Lucas, D. J., Leibowitz, K. L., and Jhanwar, Y. S. (1991). Developmental patterns of macronutrient intake in female and male rats from weaning to maturity. *Physiology & Behavior* **50**, 1167–1174. doi:10.1016/0031-9384(91)90578-C
- Lorch, P. D., Sword, G. A., Gwynne, D. T., and Anderson, G. L. (2005). Radiotelemetry reveals differences in individual movement patterns between outbreak and non-outbreak Mormon cricket populations. *Ecological Entomology* **30**, 548–555. doi:10.1111/j.0307-6946.2005.00725.x
- Lozano, G. A. (1998). Parasitic stress and self-medication. *Advances in the Study of Behavior* **27**, 291–317. doi:10.1016/S0065-3454(08)60367-8
- Luckey, T. D., and Stone, P. C. (1960). Hormology in nutrition. *Science* **132**, 1891–1893. doi:10.1126/science.132.3443.1891
- Mair, W., Piper, M. D. W., and Partridge, L. (2005). Calories do not explain extension of life span by dietary restriction in *Drosophila*. *PLoS Biology* **3**, e223. doi:10.1371/journal.pbio.0030223
- Maklakov, A. A., Simpson, S. J., Zajitschek, F., Hall, M. D., Dessmann, J., Clissold, F., Raubenheimer, D., Bonduriansky, R., and Brooks, R. C. (2008). Sex-specific fitness effects of nutrient intake on reproduction and lifespan. *Current Biology* **18**, 1062–1066. doi:10.1016/j.cub.2008.06.059
- Masoro, E. J. (2005). Overview of caloric restriction and ageing. *Mechanisms of Ageing and Development* **126**, 913–922. doi:10.1016/j.mad.2005.03.012
- Mayntz, D., Raubenheimer, D., Salomon, M., Tøft, S., and Simpson, S. J. (2005). Nutrient-specific foraging in invertebrate predators. *Science* **307**, 111–113. doi:10.1126/science.1105493



- Mayntz, D., Nielsen, V. H., Sørensen, A., Tøft, S., Raubenheimer, D., Hejlesen, C., and Simpson, S. J. (2009). Balancing of protein and lipid intake by a mammalian carnivore, the mink, *Mustela vison*. *Animal Behaviour* **77**, 349–355. doi:10.1016/j.anbehav.2008.09.036
- McCay, C. M., Crowell, M. F., and Maynard, L. A. (1935). The effect of retarded growth upon the length of life and upon ultimate size. *Journal of Nutrition* **10**, 63–79.
- McMillen, C., and Robinson, J. S. (2005). Developmental origins of the metabolic syndrome: prediction, plasticity, and programming. *Physiological Reviews* **85**, 571–633. doi:10.1152/physrev.00053.2003
- Mertz, W. (1981). The essential trace elements. *Science* **213**, 1332–1338. doi:10.1126/science.7022654
- Miller, R. A., Buehner, G., Chang, Y., Harper, J. M., Sigler, R., and Smith-Wheelock, M. (2005). Methionine-deficient diet extends mouse lifespan, slows immune and lens aging, alters glucose, T4, IGF-I and insulin levels, and increases hepatocyte MIF levels and stress resistance. *Aging Cell* **4**, 119–125. doi:10.1111/j.1474-9726.2005.00152.x
- Miller, G. A., Clissold, F. J., Mayntz, D., and Simpson, S. J. (2009). Speed over efficiency: locusts select body temperatures that favour growth rate over efficient nutrient utilization. *Proceedings. Biological Sciences* **276**, 3581–3589. doi:10.1098/rspb.2009.1030
- Morton, G. J., and Schwartz, M. W. (2011). Leptin and the central nervous system control of glucose metabolism. *Physiological Reviews* **91**, 389–411. doi:10.1152/physrev.00007.2010
- Musten, B., Peace, D., and Anderson, G. H. (1974). Food intake regulation in the weanling rat: self-selection of protein and energy. *The Journal of Nutrition* **104**, 563–572.
- Newland, P. L., and Yates, P. (2008). Nitric oxide modulates salt and sugar responses via different signaling pathways. *Chemical Senses* **33**, 347–356. doi:10.1093/chemse/bjm094
- Ng, S.-F., Lin, R. C. Y., Laybutt, D. R., Barres, R., Owens, J. A., and Morris, M. J. (2010). Chronic high-fat diet in fathers programs  $\beta$ -cell dysfunction in female rat offspring. *Nature* **467**, 963–966. doi:10.1038/nature09491
- Nonaka, E., and Holme, P. (2007). Agent-based model approach to optimal foraging in heterogeneous landscapes: effects of patch clumpiness. *Ecography* **30**, 777–788. doi:10.1111/j.2007.0906-7590.05148.x
- O'Brien, D. M., Min, K.-J., Larsen, T., and Tatar, M. (2008). Use of stable isotopes to examine how dietary restriction extends *Drosophila* lifespan. *Current Biology* **18**, R155–R156. doi:10.1016/j.cub.2008.01.021
- Oom, S. P., Beecham, J. A., Legg, C. J., and Hester, A. J. (2004). Foraging in a complex environment: from foraging strategies to emergent spatial properties. *Ecological Complexity* **1**, 299–327. doi:10.1016/j.ecocom.2004.06.003
- Orentreich, N., Matias, J. R., DeFelice, A., and Zimmerman, J. A. (1993). Low methionine ingestion by rats extends life span. *The Journal of Nutrition* **123**, 269–274.
- Peck, M. D., Babcock, G. F., and Alexander, J. W. (1992). The role of protein and calorie restriction in outcome from *Salmonella* infection in mice. *Journal of Parenteral and Enteral Nutrition* **16**, 561–565. doi:10.1177/0148607192016006561
- Pincebourde, S., and Casas, J. (2006). Multitrophic biophysical budgets: thermal ecology of an intimate herbivore insect plant interaction. *Ecological Monographs* **76**, 175–194. doi:10.1890/0012-9615(2006)076[0175:MBBTEO]2.0.CO;2
- Pirk, C. W. W., Boodhoo, C., Human, H., and Nicolson, S. W. (2010). The importance of protein type and protein to carbohydrate ratio for survival and ovarian activation of caged honeybees (*Apis mellifera scutellata*). *Apidologie* **41**, 62–72. doi:10.1051/apido/2009055
- Ponton, F., Lalubin, F., Fromont, C., Wilson, K., Behm, C., and Simpson, S. J. (2011a). Hosts use altered macronutrient intake to circumvent parasite-induced reduction in fecundity. *International Journal for Parasitology* **41**, 43–50. doi:10.1016/j.ijpara.2010.06.007
- Ponton, F., Wilson, K., Cotter, S. C., Raubenheimer, D., and Simpson, S. J. (2011b). Nutritional immunology: a multi-dimensional approach. *PLoS Pathogens* **7**, e1002223. doi:10.1371/journal.ppat.1002223
- Porter, W. P., and Gates, D. M. (1969). Thermodynamic equilibria of animals with environment. *Ecological Monographs* **39**, 227–244. doi:10.2307/1948545
- Povey, S., Cotter, S. C., Simpson, S. J., Lee, K. P., and Wilson, K. (2009). Can the protein costs of bacterial resistance be offset by altered feeding behaviour? *Journal of Animal Ecology* **78**, 437–446. doi:10.1111/j.1365-2656.2008.01499.x
- Raubenheimer, D., and Bassil, K. (2007). Separate effects of macronutrient concentration and balance on plastic gut responses in locusts. *Journal of Comparative Physiology. B, Biochemical, Systemic, and Environmental Physiology* **177**, 849–855. doi:10.1007/s00360-007-0180-5
- Raubenheimer, D., and Jones, S. A. (2006). Nutritional imbalance in an extreme generalist omnivore: tolerance and recovery through complementary food selection. *Animal Behaviour* **71**, 1253–1262. doi:10.1016/j.anbehav.2005.07.024
- Raubenheimer, D., and Simpson, S. J. (1992). Analysis of covariance: an alternative to nutritional indices. *Entomologia Experimentalis et Applicata* **62**, 221–231. doi:10.1111/j.1570-7458.1992.tb00662.x
- Raubenheimer, D., and Simpson, S. J. (1993). The geometry of compensatory feeding in the locust. *Animal Behaviour* **45**, 953–964. doi:10.1006/anbe.1993.1114
- Raubenheimer, D., and Simpson, S. J. (1994). The analysis of nutrient budgets. *Functional Ecology* **8**, 783–791. doi:10.2307/2390238
- Raubenheimer, D., and Simpson, S. J. (1995). Constructing nutrient budgets. *Entomologia Experimentalis et Applicata* **77**, 99–104. doi:10.1111/j.1570-7458.1995.tb01990.x
- Raubenheimer, D., and Simpson, S. J. (1999). Integrating nutrition: a geometrical approach. *Entomologia Experimentalis et Applicata* **91**, 67–82. doi:10.1046/j.1570-7458.1999.00467.x
- Raubenheimer, D., and Simpson, S. J. (2003). Nutrient balancing in grasshoppers: behavioural and physiological correlates of dietary breadth. *The Journal of Experimental Biology* **206**, 1669–1681. doi:10.1242/jeb.00336
- Raubenheimer, D., and Simpson, S. J. (2006). The challenge of supplementary feeding: can geometric analysis help save the kakapo? *Notornis* **53**, 100–111.
- Raubenheimer, D., and Simpson, S. J. (2009). Nutritional pharmacology: doses, nutrients, toxins, and medicines. *Integrative and Comparative Biology* **49**, 329–337. doi:10.1093/icb/icip050
- Raubenheimer, D., and Tucker, D. (1997). Associative learning by locusts: pairing of visual cues with consumption of protein and carbohydrate. *Animal Behaviour* **54**, 1449–1459. doi:10.1006/anbe.1997.0542
- Raubenheimer, D., Lee, K. P., and Simpson, S. J. (2005). Does Bertrand's rule apply to macronutrients? *Proceedings. Biological Sciences* **272**, 2429–2434. doi:10.1098/rspb.2005.3271
- Raubenheimer, D., Mayntz, D., Simpson, S. J., and Tøft, S. (2007). Nutrient specific compensation following overwintering diapause in a generalist predatory invertebrate: implications for intraguild predations. *Ecology* **88**, 2598–2608. doi:10.1890/07-0012.1
- Raubenheimer, D., Simpson, S. J., and Mayntz, D. (2009). Nutrition, ecology and nutritional ecology: toward an integrated framework. *Functional Ecology* **23**, 4–16. doi:10.1111/j.1365-2435.2009.01522.x
- Ribeiro, C., and Dickson, B. J. (2010). Sex peptide receptor and neuronal TOR/S6K signaling modulate nutrient balancing in *Drosophila*. *Current Biology* **20**, 1000–1005. doi:10.1016/j.cub.2010.03.061
- Rodríguez, E., and Wrangham, R. (1993). Zoopharmacognosy: the use of medicinal plants by animals. In 'Recent Advances in Phytochemistry. Phytochemical Potential of Tropical Plants. Vol. 27'. (Eds K. R. Downum, J. T. Romeo, and H. A. Stafford.) pp. 89–105. (Plenum Press: New York.)

- Romano, M. D., Piatt, J. F., and Roby, D. D. (2006). Testing the junk-food hypothesis on marine birds: effects of prey type on growth and development. *Waterbirds* **29**, 407–414. doi:10.1675/1524-4695(2006)29[407:TTJHOM]2.0.CO;2
- Rosen, D. A. S., and Trites, A. W. (2000). Pollock and the decline of Steller sea lions: testing the junk-food hypothesis. *Canadian Journal of Zoology* **78**, 1243–1250. doi:10.1139/z00-060
- Ross, M. H. (1961). Length of life and nutrition in the rat. *The Journal of Nutrition* **75**, 197–210.
- Rubio, V. C., Sánchez-Vázquez, F. J., and Madrid, J. A. (2003). Macronutrient selection through postingestive signals in sea bass fed on gelatine capsules. *Physiology & Behavior* **78**, 795–803. doi:10.1016/S0031-9384(03)00082-9
- Sánchez-Vázquez, F. J., Yamamoto, T., Akiyama, T., Madrid, J. A., and Tabata, M. (1999). Macronutrient self-selection through demand-feeders in rainbow trout. *Physiology & Behavior* **66**, 45–51. doi:10.1016/S0031-9384(98)00313-8
- Schoonhoven, L. M., van Loon, J. J. A., and Dicke, M. (2005). 'Insect-Plant Biology.' (Oxford University Press: Oxford.)
- Schwartz, S. M., Kemnitz, J. W., and Howard, C. F. (1993). Obesity in free-ranging rhesus macaques. *International Journal of Obesity* **17**, 1–9.
- Schwartz, M. W., Woods, S. C., Porte, D. Jr. Seeley, R. J., and Baskin, D. G. (2000). Central nervous control of food intake. *Nature* **404**, 661–671.
- Sclafani, A. (2000). Macronutrient-conditioned flavor preferences. In 'Neural and Metabolic Control of Macronutrient Intake'. (Eds H.-R. Berthoud and R. J. Seeley.) pp. 93–107. (CRC Press: Boca Raton, FL.)
- Shuster, S. M., Lonsdorf, E. V., Wimp, G. M., Bailey, J. K., and Whitham, T. G. (2006). Community heritability measures the evolutionary consequences of indirect genetic effects on community structure. *Evolution* **60**, 991–1003.
- Silva, J. E. (2006). Thermogenic mechanisms and their hormonal regulation. *Physiological Reviews* **86**, 435–464. doi:10.1152/physrev.00009.2005
- Simpson, S. J., and Raubenheimer, D. (1993). A multi-level analysis of feeding behaviour: the geometry of nutritional decisions. *Philosophical Transactions of the Royal Society B. Biological Sciences* **342**, 381–402. doi:10.1098/rstb.1993.0166
- Simpson, S. J., and Raubenheimer, D. (1995). The geometric analysis of feeding and nutrition: a user's guide. *Journal of Insect Physiology* **41**, 545–553. doi:10.1016/0022-1910(95)00006-G
- Simpson, S. J., and Raubenheimer, D. (1997). The geometric analysis of feeding and nutrition in the rat. *Appetite* **28**, 201–213. doi:10.1006/appe.1996.0077
- Simpson, S. J., and Raubenheimer, D. (2000). The hungry locust. *Advances in the Study of Behavior* **29**, 1–44. doi:10.1016/S0065-3454(08)60102-3
- Simpson, S. J., and Raubenheimer, D. (2001). The geometric analysis of nutrient-allelochemical interactions: a case study using locusts. *Ecology* **82**, 422–439.
- Simpson, S. J., and Raubenheimer, D. (2005). Obesity: the protein leverage hypothesis. *Obesity Reviews* **6**, 133–142. doi:10.1111/j.1467-789X.2005.00178.x
- Simpson, S. J., and Raubenheimer, D. (2007). Caloric restriction and aging revisited: the need for a geometric analysis of the nutritional bases of aging. *The Journals of Gerontology. Series A, Biological Sciences and Medical Sciences* **62**, 707–713. doi:10.1093/gerona/62.7.707
- Simpson, S. J., and Raubenheimer, D. (2009). Macronutrient balance and lifespan. *Aging* **1**, 875–880.
- Simpson, S. J., and Raubenheimer, D. (2012). 'The Nature of Nutrition: a Unifying Framework from Animal Adaptation to Human Obesity.' (Princeton University Press: Princeton, NJ.)
- Simpson, S. J., and White, P. R. (1990). Associative learning and locust feeding: evidence for a 'learned hunger' for protein. *Animal Behaviour* **40**, 506–513. doi:10.1016/S0003-3472(05)80531-7
- Simpson, S. J., Raubenheimer, D., Behmer, S. T., Whitworth, A., and Wright, G. A. (2002). A comparison of nutritional regulation in solitary and gregarious phase nymphs of the desert locust, *Schistocerca gregaria*. *The Journal of Experimental Biology* **205**, 121–129.
- Simpson, S. J., Sibly, R. M., Lee, K. P., Behmer, S. T., and Raubenheimer, D. (2004). Optimal foraging when regulating intake of multiple nutrients. *Animal Behaviour* **68**, 1299–1311. doi:10.1016/j.anbehav.2004.03.003
- Simpson, S. J., Sword, G. A., Lorch, P. D., and Couzin, I. D. (2006). Cannibal crickets on a forced march for protein and salt. *Proceedings of the National Academy of Sciences of the United States of America* **103**, 4152–4156. doi:10.1073/pnas.0508915103
- Simpson, S. J., Raubenheimer, D., Charleston, M. A., and Clissold, F. J. ARC-NZ Vegetation Function Network Herbivory Working Group. (2010). Modelling nutritional interactions: from individuals to communities. *Trends in Ecology & Evolution* **25**, 53–60. doi:10.1016/j.tree.2009.06.012
- Skorupa, D. A., Dervisevendic, A., Zwiener, J., and Pletcher, S. D. (2008). Dietary composition specifies consumption, obesity, and lifespan in *Drosophila melanogaster*. *Aging Cell* **7**, 478–490. doi:10.1111/j.1474-9726.2008.00400.x
- Slansky, F. (1992). Allelochemical-nutrient interactions in herbivore nutritional ecology. In 'Herbivores: Their Interaction with Secondary Plant Metabolites'. (Eds G. A. Rosenthal and M. R. Berenbaum.) pp. 135–174. (Academic Press: New York.)
- Smith, V. H., and Holt, R. D. (1996). Resource competition and within-host disease dynamics. *Trends in Ecology & Evolution* **11**, 386–389. doi:10.1016/0169-5347(96)20067-9
- Sørensen, A., Mayntz, D., Simpson, S. J., and Raubenheimer, D. (2010). Dietary ratio of protein to carbohydrate induces plastic responses in the gastrointestinal tract of mice. *Journal of Comparative Physiology. B, Biochemical, Systemic, and Environmental Physiology* **180**, 259–266. doi:10.1007/s00360-009-0402-0
- Sotka, E. E., Forbey, J., Horn, M., Poore, A., Raubenheimer, D., and Whalen, K. (2009). The emerging role of pharmacology in understanding marine and freshwater consumer-prey interactions. *Integrative and Comparative Biology* **49**, 291–313. doi:10.1093/icb/icip049
- Sousa, T., Domingos, T., and Kooijman, S. A. L. M. (2008). From empirical patterns to theory: a formal metabolic theory of life. *Philosophical Transactions of the Royal Society B. Biological Sciences* **363**, 2453–2464. doi:10.1098/rstb.2007.2230
- South, S. H., House, C. M., Moore, A. J., Simpson, S. J., and Hunt, J. (2011). Male cockroaches prefer a high carbohydrate diet that makes them more attractive to females: implications for the study of condition-dependence. *Evolution* **65**, 1594–1606. doi:10.1111/j.1558-5646.2011.01233.x
- Stephens, D. W., and Krebs, J. R. (1986). 'Foraging Theory.' (Princeton University Press: Princeton, NJ.)
- Stephens, D. W., Brown, J. S., and Ydenberg, R. C. (2007). 'Foraging: Behavior and Ecology.' (The University of Chicago Press: Chicago, IL.)
- Sterner, R. W., and Elser, J. J. (2002). 'Ecological Stoichiometry: The Biology of Elements from Molecules to the Biosphere.' (Princeton University Press: Princeton, NJ.)
- Sterner, R. W., Elser, J. J., Chrzanowski, T. H., Schampel, J. H., and George, N. B. (1996). Biogeochemistry and trophic ecology: a new food web diagram. In 'Food Webs: Integration of Patterns and Dynamics'. (Eds G. A. Polis and K. O. Winemiller.) pp. 72–80. (Chapman and Hall: New York.)
- Stock, M. J. (1999). Gluttony and thermogenesis revisited. *International Journal of Obesity* **23**, 1105–1117. doi:10.1038/sj.ijo.0801108
- Sword, G. A., Lorch, P. D., and Gwynne, D. T. (2005). Migratory bands give crickets protection. *Nature* **433**, 703. doi:10.1038/433703a
- Tatar, M. (2007). Diet restriction in *Drosophila melanogaster*. In 'Mechanisms of Dietary Restriction in Aging and Disease'. (Eds C. V. Mobbs, K. Yen, and P. R. Hof.) pp. 115–136. (Karger: Basel.)

- Tatar, M. (2011). The plate half-full: status of research on the mechanisms of dietary restriction in *Drosophila melanogaster*. *Experimental Gerontology* **46**, 363–368. doi:10.1016/j.exger.2010.12.002
- Tilman, D. (1982). 'Resource Competition and Community Structure.' (Princeton University Press: Princeton, NJ.)
- Tomé, D. (2004). Protein, amino acids and the control of food intake. *The British Journal of Nutrition* **92**, S27–S30. doi:10.1079/BJN20041138
- Torregrossa, A.-M., and Dearing, D. (2009). Nutritional toxicology of mammals: regulated intake of plant secondary compounds. *Functional Ecology* **23**, 48–56. doi:10.1111/j.1365-2435.2008.01523.x
- Touzani, K., and Sclafani, A. (2005). Critical role of amygdala in flavor but not taste preference learning in rats. *The European Journal of Neuroscience* **22**, 1767–1774. doi:10.1111/j.1460-9568.2005.04360.x
- Turnbaugh, P. J., Ridaura, V. K., Faith, J. J., Rey, F. E., Knight, R., and Gordon, G. I. (2009). The effect of diet on the human gut microbiome: a metagenomic analysis in humanized gnotobiotic mice. *Science Translational Medicine* **1**, 6–14.
- Vargas, M. A., Luo, N., Yamaguchi, A., and Kapahi, P. (2010). A role for S6 kinase and serotonin in postmating dietary switch and balance of nutrients in *D. melanogaster*. *Current Biology* **20**, 1006–1011. doi:10.1016/j.cub.2010.04.009
- Vigne, P., and Frelin, C. (2010). Hypoxia modifies the feeding preferences of *Drosophila*. Consequences for diet dependent hypoxic survival. *BMC Physiology* **10**, 1–9. doi:10.1186/1472-6793-10-8
- Villalba, J. J., and Provenza, F. D. (2007). Self-medication and homeostatic behaviour in herbivores: learning about the benefits of nature's pharmacy. *Animal* **1**, 1360–1370. doi:10.1017/S1751731107000134
- Warbrick-Smith, J., Behmer, S. T., Lee, K. P., Raubenheimer, D., and Simpson, S. J. (2006). Evolving resistance to obesity in an insect. *Proceedings of the National Academy of Sciences of the United States of America* **103**, 14 045–14 049. doi:10.1073/pnas.0605225103
- Weindruch, R., and Walford, R. L. (1988). 'The Retardation of Aging and Disease by Dietary Restriction.' (Charles C. Thomas: Springfield, IL.)
- White, T. C. R. (1983). 'The Inadequate Environment: Nitrogen and the Abundance of Animals.' (Springer: Berlin.)
- Wilder, S. M., and Eubanks, M. D. (2010). Might nitrogen limitation promote omnivory among carnivorous arthropods? *Ecology* **91**, 3114–3117. doi:10.1890/09-2080.1
- Withers, P. C., and Cooper, C. E. (2010). Metabolic depression: a historical perspective. In 'Aestivation: molecular and Physiological Aspects. Progress in Molecular and Subcellular Biology. Vol. 49'. (Eds C. A. Navas and J. E. Carvalho.) pp. 1–23. (Springer: Berlin.)
- Yarmolinsky, D. A., Zuker, C. S., and Ryba, N. J. (2009). Common sense about taste: from mammals to insects. *Cell* **139**, 234–244. doi:10.1016/j.cell.2009.10.001
- Zanotto, F. P., Simpson, S. J., and Raubenheimer, D. (1993). The regulation of growth by locusts through post-ingestive compensation for variation in the levels of dietary protein and carbohydrate. *Physiological Entomology* **18**, 425–434. doi:10.1111/j.1365-3032.1993.tb00617.x
- Zanotto, F. P., Gouveia, S. M., Simpson, S. J., Raubenheimer, D., and Calder, P. C. (1997). Nutritional homeostasis in locusts: is there a mechanism for increased energy expenditure during carbohydrate overfeeding? *The Journal of Experimental Biology* **200**, 2437–2448.
- Zera, A. J., and Harshman, L. G. (2001). The physiology of life history trade-offs in animals. *Annual Review of Ecology and Systematics* **32**, 95–126. doi:10.1146/annurev.ecolsys.32.081501.114006
- Zhang, Y., Proenca, R., Maffei, M., Barone, M., Leopold, L., and Friedman, J. M. (1994). Positional cloning of the mouse obese gene and its human homologue. *Nature* **372**, 425–432. doi:10.1038/372425a0
- Zimmerman, J. A., Malloy, V., Krajcik, R., and Orentreich, N. (2003). Nutritional control of aging. *Experimental Gerontology* **38**, 47–52. doi:10.1016/S0531-5565(02)00149-3

Handling Editor: Paul Cooper