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The problem of nitrogen disposal in the obese

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Abstract

Amino-N is preserved because of the scarcity and nutritional importance of protein. Excretion requires its conversion to ammonia, later incorporated into urea. Under conditions of excess dietary energy, the body cannot easily dispose of the excess amino-N against the evolutively adapted schemes that prevent its wastage; thus ammonia and glutamine formation (and urea excretion) are decreased. High lipid (and energy) availability limits the utilisation of glucose, and high glucose spares the production of ammonium from amino acids, limiting the synthesis of glutamine and its utilisation by the intestine and kidney. The amino acid composition of the diet affects the production of ammonium depending on its composition and the individual amino acid catabolic pathways. Surplus amino acids enhance protein synthesis and growth, and the synthesis of non-protein-N-containing compounds. But these outlets are not enough; consequently, less-conventional mechanisms are activated, such as increased synthesis of $NO^{\bullet}$ followed by higher nitrite (and nitrate) excretion and changes in the microbiota. There is also a significant production of N_2 gas, through unknown mechanisms. Health consequences of amino-N surplus are difficult to fathom because of the sparse data available, but it can be speculated that the effects may be negative, largely because the fundamental N homeostasis is stretched out of normalcy, forcing the N removal through pathways unprepared for that task. The unreliable results of hyperproteic diets, and part of the dysregulation found in the metabolic syndrome may be an unwanted consequence of this N disposal conflict.

Key words: Amino acids: Ammonia: Metabolic syndrome: Obesity: Urea: Nitrogen excretion

Introduction

When compared with the metabolism of carbohydrates and lipids, amino acid metabolism in man has been only sparsely studied in relation to overall energy metabolism. The roles of protein in starvation[1,2] and malnutrition[3,4], however, have received more attention; however, in any case, our actual knowledge of N metabolism in man is far more limited than the detailed information available on energy partition between carbohydrate (glucose) and lipids, including their regulation systems. Curiously, the study of proteins has been neglected despite being a key nutritional source of energy. Probably, the present situation of limited knowledge is a compounded consequence of the relatively large number of different molecular species, their easy interconversion, the multiple catabolic paths followed by their hydrocarbon skeletons, the methodological difficulties of tracing the fate of N, its close relationship with protein turnover, a multiplicity of functional amino acid pools coupled with an active interorgan metabolism, and last, but not least, an excessively sketchy knowledge of their catabolic paths and regulation in man (mammals).

Irrespective of the lack of information, the manipulation of protein content of diets has been actively developed for at least half a century, mainly by using hyperproteic very-low-energy diets for the treatment of obesity[5,6], with serious problems often arising from their application[7]. There has been a considerable utilisation of low-carbohydrate diets, in which the protein component is conspicuous[8,9], but most of the discussion of their effects has been centred on their ketogenic (i.e. lipid, absence of carbohydrate) nature[10], the amino acids essentially being considered potential gluconeogenic substrates[11], with little impact on protein synthesis[12]. More recently, the use of hyperproteic diets[13] is again on the rise, but we still lack the necessary basic knowledge to interpret the results obtained, largely because of few systematic analyses and the continued stress on their ketogenic nature[8,9].

However, under conditions of abundant food supply, the main question is not how amino acids fare under conditions of low energy availability, but the contrary: how the metabolic machinery can override the strong protective mechanisms preventing N wasting under conditions of excess energy (i.e. lipid, carbohydrate and protein) intake. There are few studies on how amino acids are

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used as substrates, especially on the fate of N under conditions of excess energy intake or obesity. In the present review, the main questions posed by the combination of excess energy and amino-N availability are analysed under the light of the scarce information available.

Amino acids as energy substrates: amino-nitrogen sparing

One of the most significant differences between what our ancestors ate (i.e. the diet for which our digestive system and metabolic energy partitioning are geared and optimised) and the present-day diet is, in addition to the overwhelming abundance of lipid, the constant presence of protein, with a relatively high proportion of high-quality protein. The tandem lipid–animal protein is substituting progressively complex carbohydrate–low-quality plant protein as the main dietary staple. The proportion of protein energy *v.* total energy intake is not too much different today from the ancestral diet[14], but the total amount of energy (after correcting for exercise) is higher, as is the proportion of essential amino acids[15], whilst the relationship between dietary amino acids *v.* glucose derived from dietary sources tends to change as a direct consequence of the substitution of starches by fats[16].

Our starvation resistance-prone mechanisms of adaptation preserve the use of amino acids as energy substrate when there is sufficient energy in the gut[17]. In addition, both amino acids and glucose are spared when (if) the availability of lipid is high[18,19]. In consequence, a diet rich in energy and lipids, with a sizeable proportion of easily digestible protein, rich in essential amino acids, will necessarily result in difficulties to process and oxidise the amino acid surplus, since we are metabolically conditioned to actively preserve them. However, under excess-energy diets (including protein), it is no longer necessary to retain so much amino-N and essential amino acids. Amino acids are used for energy when in relative excess[20,21], but the even higher availability of energy from other sources strongly limits our metabolic machinery to do so[22].

Amino acid catabolism tends to retain 2-amino-N, largely because most amino acid hydrocarbon skeletons are oxidised after transamination (typically to 2-oxoglutarate/glutamate). However, a few amino acids yield directly ammonia (Table 1). A quantitative analysis of several common food proteins shows that, as expected, the theoretical direct yield of 2-amino-N is much higher than that of direct ammonium production (Table 2). Thus, the parity needed to synthesise our main N excretion product, urea, requires (in a dietary equilibrium) the additional conversion of a varying proportion of dietary 2-amino-N to ammonium to reach the required (1:1) balance. The ratio amino-N:ammonium-N in most dietary proteins is close to 2 (Table 2), which leaves a wide margin for preservation of N under conditions of starvation, but in the end requires the mineralisation of about half of all amino-N to ammonia under normal feeding conditions.

When active (exercise), muscle uses most of the body energy available: its standard feed is glucose, but blood lipids (fatty acids) limit glucose uptake and favour fatty acid oxidation (insulin resistance)[23]. Some amino acids are oxidised in muscle (especially branched-chain[24]) in the postprandial state to save glucose, but excess energy hampers the process and its timing, since in fact there is no real scarcity of glucose. Amino-N conversion into ammonia is largely done in the liver and muscle through the purine nucleotide cycle[25], and its operation is both linked to active glycolysis[26] and increased AMP levels (i.e. low ATP availability, partly compensated by the action of adenylate kinase)[27]; thus, under excess energy availability, the cycle is largely idle. Glutamate dehydrogenases play a key role in ammonium metabolism in micro-organisms[28], and in the muscle of invertebrates[29]. However, Their role in ammoniagenesis in mammalian muscle is limited, because of the predominance of the purine nucleotide cycle[30] in this role, and the low presence of the enzyme compared with the liver[31], unaltered under starvation[32]. In the liver (and kidney), the activity of glutamate dehydrogenase is considerable[31], but its function is clearly that of resynthesising glutamate from 2-ketoglutaratre and excess ammonia, as determined by direct studies and the analysis of its kinetic constrictions[33–35].

Equilibrium between amino-nitrogen and ammonia for urea synthesis

Thus, in muscle there is no other major way to produce ammonia than the purine nucleotide cycle[30]. In consequence the muscle cannot use amino acids as an energy source in significant amounts[36] and to use their N to produce and release glutamine. This is an important question, since glutamine is the main form of blood transport of ammonia, towards the splanchnic bed[37], i.e. intestine and kidney; there, glutaminases free the ammonia again for its ultimate disposal as urea[38], or as urinary ammonium ion[39]. Lower muscle synthesis of glutamine results, then, in lower splanchnic synthesis of carbamoyl-P, insufficient to maintain an adequate flow of urea synthesis (Fig. 1). Alternative sources of ammonium, such as the microbiota[40,41], which is in part derived from glutamine[42,43], and the direct ammoniagenic amino acids cited above (serine, threonine, glycine), help maintain a steady albeit diminished rate of urea synthesis in the intestine–liver system, a rate insufficient to cope with the excess 2-amino-N pool generated by the diet and limited amino acid disposal.

High amino acids in conjunction with high energy availability can generate a paradoxical scarcity of ammonia, retaining a large and unshrinkable pool of amino-N because the mechanisms that protect its conversion to ammonia remain unaffected, and are both efficient and effective (and potentially crippling). In the metabolic syndrome (and in general, in energy-rich feeding), urea

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Table 1. Main amino acid (AA) catabolism pathways in man (adapted from Ferrer-Lorente *et al.*[130])*

AA	Flow	Pathway	-NH_2	Mean	NH_3	Mean
Gly	60	[Gly-cleavage system] → methylene-THF → methenyl-THF → formate	0	0·35	1	0·60
	35	[trans to] glyoxylate → glycolate	1		0	
	10	[via UC conjugated with Arg] to creatine → creatine-P → creatinine	0		0	
	5	Other: excretion intact or as peptides, conjugation of xenobiotics and hormones	0		0	
Ala	100	[trans to] pyr → AcCoA → [KC]	1	1·00	0	0·00
Ser	90	[Ser-dehydratase to] pyr → AcCoA → [KC]	0	0·05	1	0·95
	5	[trans to] pyr-OH → AcCoA → [KC]	1		0	
	5	[cleavage to] Gly + 1-C	0		1	
Cys	90	[trans + desulfuration to] pyr → AcCoA → [KC]	0	0·00	1	0·90
	10	Other: conversion to taurine	0		0	
Met	100	[conjugated with ATP] → S-adenosylMet → [de-methylation (→ 1-C) to] Hcys → [conjugated with Ser] → cystathionine → (pyr → AcCoA → [KC]) + oxobutyrate → propionylCoA → succ → OAA → pyr → AcCoA → [KC]	0	0·00	1	1·00
Thr	95	[Thr dehydratase to] oxobutyrate → propionylCoA → succ → OAA → pyr → AcCoA → [KC]	0	0·00	1	1·00
	5	[Thr aldolase to] Gly + acetaldehyde	0		1	
Asp	50	[trans to] OAA → pyr → AcCoA → [KC]	1	0·50	0	0·50
	50	[via PNC or UC to] fumarate → OAA → pyr → AcCoA → [KC]	0		1	
Asn	100	[asparaginase to] Asp	1	1·00	1	1·00
Glu	97	[trans to] oxoglutarate] → OAA → pyr → AcCoA → [KC]	1	1·00	0	0·00
	3	[decarboxylation to] γ-aminobutyrate → succ → OAA → pyr → AcCoA → [KC]	1		0	
Gln	100	[glutaminase to] Glu	1	1·00	1	1·00
Pro	50	[dehydrogen to] pyrroline-carboxylate → Glu	1	0·95	0	0·00
	45	[oxid to] pyrroline-carboxylate → Glu	1		0	
	5	Excretion. Intact or as peptides	0		0	
Hyp	45	[dehydrogen to] OH-pyrroline-carboxylate → erythro OH-Glu → OH-oxoglutarate → (glyoxylate → glycolate) + (pyr → AcCoA → [KC])	1	0·90	0	0·00
	45	[oxid to] pyrroline-carboxylate → erythro OH-Glu → OH-oxoglutarate → (glyoxylate → glycolate) + (pyr → AcCoA → [KC])	1		0	
	10	Excretion. Intact or as peptides	0		0	
Val	100	[trans to] oxoisovalerate → isobutyrylCoA → [BO] → succCoA → succ → OAA → pyr → AcCoA → [KC]	1	1·00	0	0·00
Leu	100	[trans to] oxoisocaproate → [BO] → (AcCoA → [KC]) + AcAc → AcCoA → [KC]	1	1·00	0	0·00
Ile	100	[trans to] oxomethylvalerate → methylbutyrylCoA → [BO] → (AcCoA → [KC]) + propionylCoA → succ → OAA → pyr → AcCoA → [KC]	1	1·00	0	0·00
Lys	90	[conjugated with oxoglutarate] → saccharopine → (Glu) + oxoadipate → AcAcCoA → AcCoA → [KC]	2	1·80	0	0·10
	10	[deamination to] pipecolate → oxoadipate → AcAcCoA → AcCoA → [KC]	1		1	
His	95	[deamination to] urocanate → formiminoGlu → [1-C] + Glu	1	0·95	2	1·90
	5	Other: excretion/conversion to histamine	0		0	
Arg	100	[via UC to] Orn → Glu-semi-aldehyde → pyrroline-carboxylate → Glu	1	1·00	0	0·00
Trp	70	[oxid to] kynurenine (+ formate) → (Ala → pyr → AcCoA → [KC]) + oxoadipate → crotonyl-CoA → [BO] → AcCoA → [KC]	2	1·70	0	0·00
	30	[trans to] indol-pyruvate → indol-acetate (excreted)	1		0	
Tyr	95	[trans to] OH-phenyl-pyruvate → homogentisate → (fumarate → OAA → pyr → AcCoA → [KC]) + AcAc → AcCoA → [KC]	1	0·95	0	0·00
	5	Other: [via dihydroxyphenylalanine. incorporation into] melanin, or catecholamines; excretion	0		0	
Phe	95	[oxid to] Tyr	1	1·00	0	0·00
	5	Other: largely [trans to] and excretion as phenylpyruvate)	1		0	

THF, tetrahydrofolate; [trans to], transamination to; UC, urea cycle; AcCoA, acetyl-coenzyme A; [KC], Krebs or tricarboxylic acid cycle; pyr, pyruvate; [1-C], one-carbon pathways (essentially via THF); succ, succinate; OAA, oxaloacetate; [dehydrogen to], dehydrogenation to; [oxid to], oxidation to; PNC, purine nucleotide cycle; [BO], β-oxidation pathway; AcAc, acetoacetate; AcAcCoA, acetoacetyl-coenzyme A.

* All values are estimations based on the scant data available from human subjects and other mammals; flow of amino acid catabolism via a given path depends largely on the size of amino-N pool, energy availability, metabolic needs and the relative abundance of the amino acid (essential amino acids).

Table 2. Amino acid (AA) content of a number of common food proteins, showing the -NH_2:NH_3 ratio that would be theoretically generated from the complete oxidation in the body of its constituent amino acids*

	AA content (mmol/g protein)	-NH_2 (mmol/g protein)	NH_3 (mmol/g protein)	-NH_2:NH_3 ratio	'Excess -NH_2' (% of -NH_2)
Cheese	888	775	296	2·61	62
Potatoes	912	774	312	2·48	60
Maize 1	928	766	309	2·48	60
Maize 2	935	771	316	2·44	59
Cows' milk	884	758	319	2·38	58
Pork	908	782	340	2·30	57
Spinach	923	748	337	2·22	55
Beef	933	770	349	2·21	55
Pilchard fish	919	770	348	2·21	55
Haddock fish	908	773	351	2·20	55
Lamb meat	928	764	351	2·18	54
Peas	896	750	344	2·18	54
Cassava	875	734	339	2·16	54
Lentils	900	745	347	2·14	53
Chickpeas	893	736	345	2·13	53
Soyabeans	899	744	352	2·11	53
Mollusks	920	743	356	2·09	52
Wheat	903	734	351	2·09	52
Chicken	909	748	360	2·08	52
Crustaceans	928	742	357	2·08	52
Tuna fish	903	774	375	2·06	52
Polished rice	915	723	357	2·03	51
Carrots	908	750	374	2·01	50
Beans	907	743	373	1·99	50
Hens' eggs	914	713	359	1·98	50
Cabbage	944	724	375	1·93	48
Sesame	898	698	362	1·93	48
Hazelnuts	915	680	377	1·81	45
Apples	928	743	423	1·75	43

* The data have been calculated from standard protein amino acid composition tables[131] and the yield in free ammonium and transaminable 2-amino groups resulting from the complete catabolism of these amino acids (Table 1). Since urea excretion requires equal proportions of amino and ammonium, any -NH_2:NH_3 ratio above 1·00 represents a relative excess of amino groups, which may be further converted to ammonium via the purine nucleotide cycle or (in certain tissues and physiological conditions) by glutamate dehydrogenase. The ammonium yield of the proteins listed may be underestimated, since most of the data gathered give a combined Glu + Gln (Glx) value in the overall analyses. From the analysis of whole rat protein[132] in which amide-N was analysed separately, we assumed, conservatively, and for the sake of these calculations only, that half the Glx values corresponded to Gln and half to Glu; this correction has been included in the calculations and is reflected in the data shown in the Table.

synthesis is decreased[44], but there is not – either – a massive accumulation of body-N[45]. Amino acids tend to be preserved in spite of excess energy availability[46], but in any case, the excess N is eventually lost, albeit not in the canonical way of urea formation[47]. A small but significant proportion of N is excreted as N_2 gas[48,49] by means of, so far, unknown pathways. In addition there is an increased (but relatively small) loss of dietary amino-N in the form of urinary nitrate and nitrite[50]. Obligatory N losses also include urinary losses of uric acid (from purine catabolism[51]), creatinine and small proportions of peptides and amino acids, as well as the ammonium ion, excreted by the kidney (especially in acidosis)[52]. Small amounts of ammonium may be also excreted by the lungs[53].

Muscle also accumulates fat, near mitochondrial clusters (C Cabot, K Pouillot, S Roy, MM Romero, R Vilà, MM Grasa, M Esteve, JA Fernández-López, M Alemany and X Remesar, unpublished results) and adapts itself to the utilisation of this main substrate (as well as to glucose, but to a lower extent)[54,55]. Exercise facilitates the massive utilisation of energy and streamlines the oxidation of fats[56], but also restores in part the production of ammonium via the purine nucleotide cycle[57], thus increasing the flow of glutamine to the splanchnic bed. However, a large proportion of glucose, lipids and amino acids can not be taken up by any of the above cited systems, leaving them unused and in high serum concentrations, waiting for their storage as fat in the last-recourse energy pool: white adipose tissue.

The nitric oxide pathway

$NO^{\bullet}$ is synthesised from arginine by $NO^{\bullet}$ synthases, yielding citrulline[58]. Excess N availability increases the synthesis of ornithine[59], including the intermediate step of acetylglutamate synthesis[60], which is also a key regulator of carbamoyl-P synthase 2, and thus also participates in the regulation of ammonium disposal[61]. In consequence, higher amino-N levels may increase those of arginine, irrespective of low carbamoyl-P (i.e. low ammonium) availability, shunting the $NO^{\bullet}$ cycle from arginine to citrulline and leaving out ornithine (and the synthesis of urea) (Fig. 2 (a) and (b)) [62]. In cells that do not have a fully operative urea cycle, the eventual regulation is even easier since it is largely dependent on arginine availability[63].

It may be expected, then, that under high energy and amino-N availability, the low ammonia concentrations[64]

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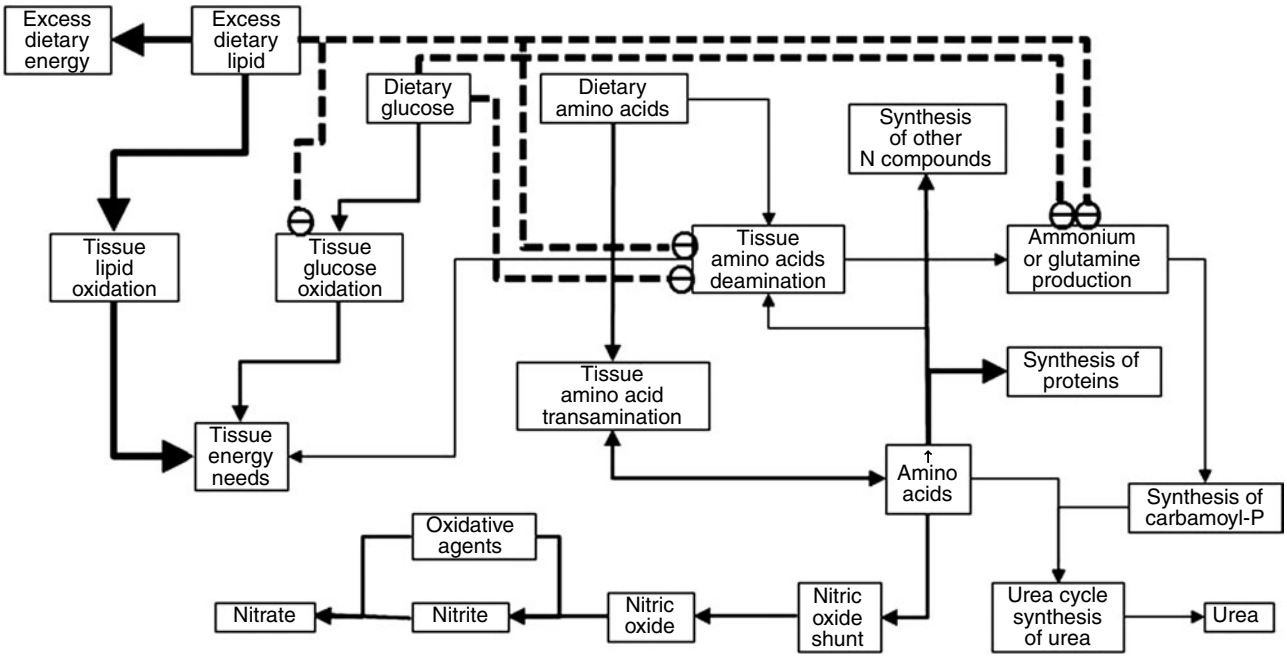

Fig. 1. Effect of excess dietary lipid on the main paths of N catabolism, driving to a decrease in the operation of the urea cycle because of lack of conversion of 2-amino-N to ammonium.

can not sustain an effective excretion of N through the urea cycle[65], indirectly favouring an increased activity of the NO• synthesis shunt. The excess NO• in blood vessels (derived from the activity of erythrocyte and endothelial NO• synthases)[66] may initially raise the blood flow, at least locally, increasing the availability of oxygen and substrates to the surrounding cells[67].

NO• is highly reactive and interacts with specific proteins, such as guanylate cyclase[68], increasing the production of cyclic GMP which activates protein kinase G (PKG)[69] which, in turn, relaxes the smooth muscle of small vessels and thus increases blood flow and lowers arterial tension[70]. This is the main recognised function of NO•[71], but NO• is also able to bind cysteine residues of other proteins, such as protein kinase A (PKA)[72], which may induce a phantom adrenergic stimulation (i.e. without the intervention of catecholamines or cAMP)[73].

Cytochrome *c* also efficiently oxidises NO• to nitrite[74]. Most of the NO•, however, rapidly reacts with oxyhaemoglobin, eventually oxidising NO• to nitrate[75]. Other highly reactive NO_x compounds, such as peroxynitrite[76], are formed by further oxidation with reactive oxygen species. Part of these nitrogen oxides react with proteins, fatty acids and other compounds yielding nitro-derivatives[77,78], often short-lived, but which can cause permanent structural changes[79].

Nitrite and other forms of nitrogen excretion

In the obese, the overall production and levels of NO• are increased[80], as is its loss in the air breathed[81], but there is a marked decrease in the excretion of urea[44]. A significant part of the difference in the N balance is made up of N_2 gas[45,47,82]. A possible source is the reaction of nitrite and free amino acids, which in an acidic medium yield N_2 gas and 2-hydroxyacids[83]; this reaction has been described to occur in the stomach lumen[84]. However, this reaction can hardly explain the large discrepancies found in N balances. There must be another – larger – source of N_2 gas integrated in the amino acid metabolism, which so far has not been discovered. We can hypothesise the existence of an 'emergence' pathway, shunting the action of NO• synthases towards the reaction of arginine with nitrite, yielding citrulline and N_2 gas under acidotic conditions. This way, nitrite, the main active product of NO• synthesis, would be rapidly removed and, at the same time, the excess 2-amino-N would be decreased at the expense of aspartate-derived arginine guanido-N; unfortunately, no enzyme has been found (so far) able to carry out this reaction, which nevertheless is known to proceed spontaneously under low pH conditions[83].

Glucocorticoids may elicit counteractive actions[85] to inhibit NO• synthesis, but catecholamines increase its production[86]. It is unclear whether NO• overproduction in the obese can be a consequence of leptin-related catecholamine vasoconstriction[87], a consequence of enhanced NO• synthesis through activation of endothelial or inducible NO• synthases[88], or a lower bioavailability of NO• favouring its increased synthesis[89,90].

Nitrite is considered a stabilised form of NO•[91], which can yield NO• under hypoxic conditions by reacting with Hb[92–94], thus helping increase blood flow to hypoxic areas[95]. Nitrite is also a source of NO• in the alimentary canal[96,97]; it is largely the product of reduction by the oral

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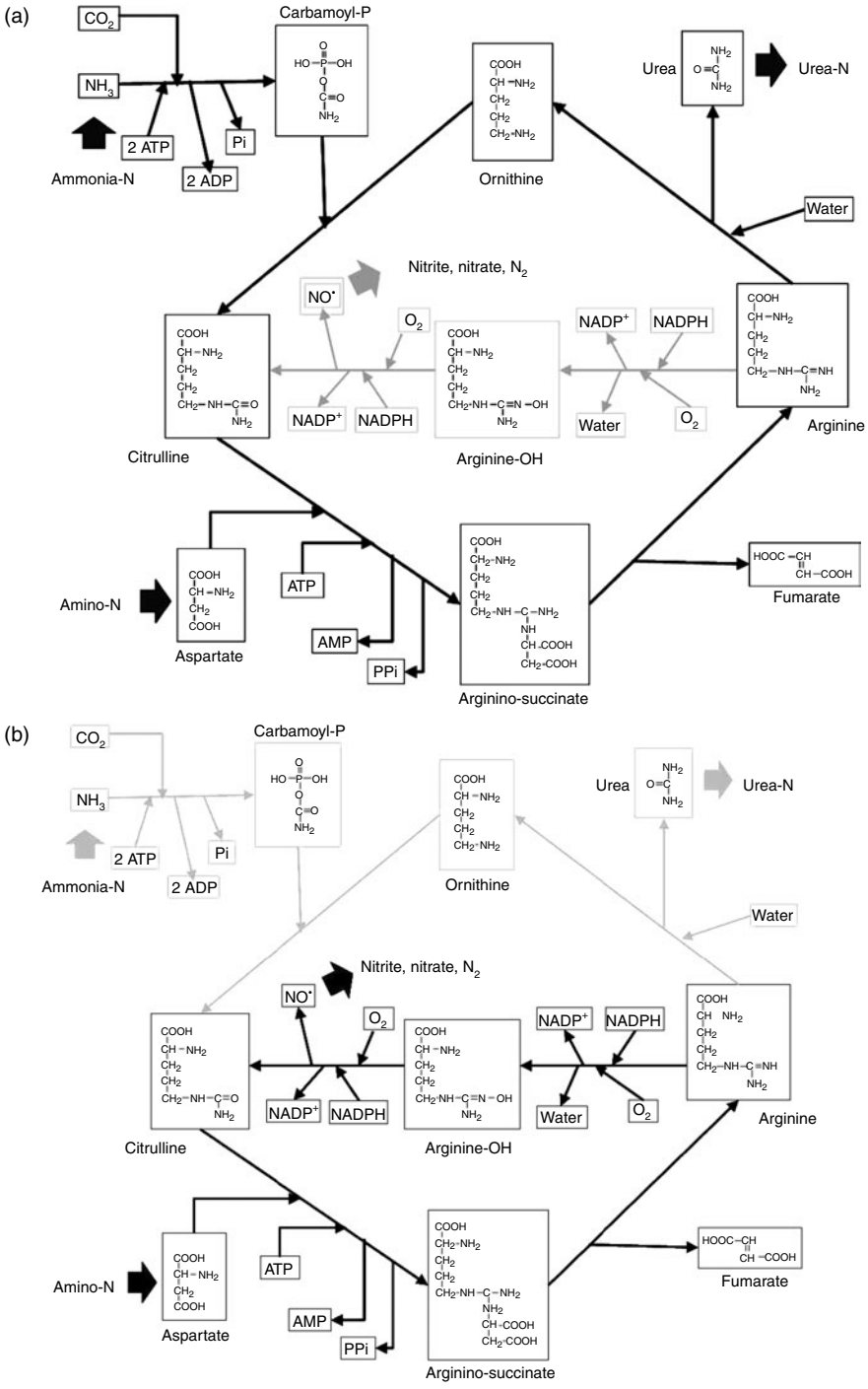

Fig. 2. Possible mechanism of activation of the NO· shunt under high energy availability–low ammonium production. (a) Urea cycle function under full operation, i.e. enough ammonium to produce carbamoyl-P and an adequate supply of 2-amino-N through aspartate. (b) Enhancement of the operation of the NO· shunt of the urea cycle under limited supply of ammonium (in the form of carbamoyl-P), but maintained supply of 2-amino-N through aspartate. Pi, inorganic phosphate; PPi, inorganic pyrophosphate.

biota of nitrate secreted by salivary glands[98]. Nitrite-derived NO· also kills bacteria in the stomach[99]; in this acidic medium, nitrite reacts with free amino acids yielding N-nitroso-proline from arginine[100], as well as N_2 as indicated above[84].

The 'obese' microbiota[101,102] is probably a consequence of this magnified effect of NO_x[103]; changes in the gut microbial ecosystem and composition also influence the relationships with the host through modulation of the immune response[104,105]. The relative abundance of protein debris in the intestine, a consequence of diets rich in lipid and protein, together with relatively scarce fibre and polysaccharides, also affects the composition of the microbiota, increasing the share of amino acid-related catabolism in the

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process of formation of stool[106,107]. The resulting higher pH, and the production of amines through amino acid decarboxylation[107], higher proportions of amines, ammonium, as well as amine- and sulfide-related catabolites may also help induce the development of intestinal cancer[108].

Health consequences of hampered nitrogen excretion in the obese

The main problem posed by this question is the almost absolute lack of information about the human patterns of N excretion in overnutrition, obesity and the metabolic syndrome. It has been found that a relative increase in dietary protein at the expense of carbohydrates facilitates the loss of weight[109,110], but we only know the short-term macroscopic changes; the dynamics of 2-amino-N under these conditions has not been studied.

We have mechanisms to adjust amino acid catabolism to their relative abundance with respect to glucose[17], but the large presence of lipids in the diet alters everything. Ketogenic diets favour the excretion of ammonium in the urine to counter the acidosis produced by ketone bodies[111,112], and increase liver gluconeogenesis from amino acids[113], but the problem of conversion of amino-N to ammonium remains. The possible negative effects of a few truly hyperproteic (i.e. not ketogenic) diets[114], and their limited effect on body fat point both to a generalised inefficiency of the so-called 'high-protein' diets[115] and support the relative danger of their uncontrolled application.

One of the most important aspects of amino acid metabolism is the synergistic complementarity of the roles of a number of peripheral organs, the liver and the rest of splanchnic bed organs[116,117]. Hyperproteic diets may induce changes in their roles in the absence of energy overload, i.e. under conditions of active amino acid catabolism[118]. However, it is highly improbable that the finely adjusted inter-organ relationships could be maintained under the pressure of high-energy diets, as the low urea output seems to indicate; as a consequence the whole body is affected by an excess of 2-amino-N.

There are few data on human subjects supporting an increase in the synthesis of $NO^{\bullet}$ in high-energy availability conditions, except for an increased breath release of $NO^{\bullet}$[81] and the consequent formation of nitrite and nitrate[93,94]. Perhaps the high levels of nitrite and the easy interconversion of nitrite and $NO^{\bullet}$[99,119], a powerful vasodilator[120], may be related to the 'obesity paradox', i.e. a decreased severity of the consequences of heart failure in the obese[121,122]. The large presence of NO_x in the alimentary canal and its profound influence on the microbiota has to produce, necessarily, changes in their properties and functions: at least a different way to cope with unused substrates and different relationships with the immune system-controlled intestinal barrier. The latter may be related to the higher levels of circulating lipopolysaccharide observed in the metabolic syndrome[123], also linked to the maintenance of low-key inflammation[124,125] caused by increased intestinal bacteria activity[126]. These findings hint to the postulated excess 2-amino-N, in agreement with the higher availability of amino acids and energy to increase protein turnover[127,128] and to maintain a fully functional immune system[129] observed in the metabolic syndrome.

Conclusions

Humans are fairly well prepared for amino-N scarcity: dietary protein utilisation is maximised, and amino acid catabolism is restricted in order to preserve body protein, and, with that, to maintain the ability to function and survive. However, the same mechanisms that make possible sparing amino acid catabolism for energy seriously hamper the metabolism of excess dietary amino-N under conditions of overfeeding and excess available energy. Insulin resistance limits the use of glucose when fats are readily available, and ample glucose (energy) availability practically blocks the removal of amino-N to form ammonia, the only, and narrow, canonical way to dispose of excess N. The obese excrete less urea than the lean, high-energy diets inhibit the urea cycle function, but also alter the glucose–alanine cycle and the operation of the purine nucleotide cycle; the path of conversion of amino-N to ammonium is severely restricted. This creates a surplus amino acid availability which enhances growth and protein synthesis, but protein turnover simply stores, and transamination changes, the hydrocarbon skeletons, preserving the amino groups. Consequently, non-conventional mechanisms are necessarily activated (there is no body storage of this surplus N). We do not know how this is accomplished, and only can suggest the possible implication of $NO^{\bullet}$-increased synthesis, followed by higher nitrite (and nitrate) secretion/excretion, and including the production of N_2 gas, through a mechanism so far unsolved.

The metabolic consequences of the imbalance between amino- and ammonia-N are far-reaching and should be studied in detail, since probably a number of unexplained phenomena of the metabolic syndrome sink their roots in the profound alteration of N homeostasis. The consequences of excess dietary protein and our inability to dispose of it have not been studied, but the indications obtained from animal studies and the few data available suggest that excess protein is harmful in the long term for humans.

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