

CONCLUSION

Growth, as measured by weight gain, and reproductive performance are the most common factors used in assessing a diet. Often reproductive performance is given on the results with one or two litters. The desirability of rapid growth has been a dominant idea in current nutritional thought, and recent discoveries have resulted in feeding practises that have accelerated the growth rate as measured by weight gain. However, some animals with a promising growth performance fail to come into profitable production, or have a short reproductive life. It cannot be said that all factors for growth are optimal until their influence on the life-time performance has been studied as thoroughly as their effects during the growth period itself.

It would seem that in the past too much emphasis has been put on weight and size in judging the value of a diet for promoting growth with less consideration of the actual composition of the changes in form. With breeding stock the life-time performance is the final measure of success and it is important to consider several aspects of growth and their influence on the life-time performance. Therefore before a diet can be described as being adequate for maintenance and reproduction it should be tested through all these aspects.

I should like to thank Dr W. Lane-Petter and Professor B. S. Platt for the help and advice they have given me in the preparation of this paper and during the actual course of the work.

REFERENCES

- Bender, A. E. & Miller, D. S. (1953). *Biochem. J.* **53**, vii.
 Brody, S. (1945). *Bioenergetics and Growth*. New York: Reinhold Publishing Corp.
 Bruce, H. M. & Parkes, A. S. (1949). *J. Hyg., Camb.*, **47**, 202.
 Dean, E. W. & Stark, D. D. (1920). *J. Indust. Engng Chem.* **12**, 486.
 Hald, J., Giddings, G. & Wynn, W. (1941-2). *Amer. J. Physiol.* **135**, 392.
 Howie, J. W. (1948-9). *Brit. J. Nutr.* **2**, 331.
 Howie, J. W. & Porter, G. (1950). *Brit. J. Nutr.* **4**, 175.
 Kabat, E. A. & Mayer, M. M. (1948). *Experimental Immunoochemistry*. Springfield, Ill.: Charles C. Thomas.
 Kaye, I. A., Leibner, I. W. & Connor, E. B. (1940). *J. biol. Chem.* **132**, 195.
 Mackay, D. H. (1952). *Trans. R. Soc. trop. Med. Hyg.* **46**, 135.
 McCay, C. M., Crowell, M. F. & Maynard, L. A. (1935). *J. Nutr.* **10**, 63.
 Orr, J. B. & Richards, M. B. (1934). *Biochem. J.* **28**, 1259.
 Sengupta, S. R. & Howie, J. W. (1948-9). *Brit. J. Nutr.* **2**, 313.
 Tomlin, D. H., Henry, K. M. & Kon, S. K. (1953). *Brit. J. Nutr.* **7**, 235.

Nutrient requirements of rats and mice

By W. F. J. CUTHBERTSON, *Glaxo Laboratories Ltd, Greenford, Middlesex*

Nutrient requirements may be assessed in several ways: the quantity of a specific nutrient needed to abolish or prevent the appearance of a specific deficiency disease

may be estimated or the amount needed to ensure the maximum growth rate or essential for a full life-cycle may be determined. Most data in the literature relate to the quantities needed to prevent abnormal phenomena or to ensure maximum growth rates.

Analysis of diets known to be satisfactory in practical use may also give a guide to nutrient requirements, though the method may frequently overestimate the quantities essential.

The needs of an animal depend not only on its species, and even strain, but also on its physiological state and other factors, such as the nature of the diet and the external environment; a margin of safety has therefore to be allowed for these variables. The suggestions made in this paper refer to those amounts thought to be required for the full life-cycle and are expressed as required concentrations in the diet. This method has shortcomings, because the requirements per unit weight of food will depend on the calorific value of the diet, but data presented in this way are simpler to use in designing diets than are daily requirements or figures based on surface area or body-weight. The requirements of the rat and mouse for their known dietary essentials are so presented in Tables 1-4 (most of the published data are for the minimum need for maximum growth). Quantities in bold type in the table are suggestions thought to prove optimal in most circumstances for most strains of these species.

Table 1. *Nutrient requirements* of the rat and the mouse*

Vitamin	Rat		Mouse	
	Reference	Quantity/ kg diet	Reference	Quantity/ kg diet
(a) Fat-soluble vitamins				
Vitamin A	Rubin & De Ritter (1954)	3000 i.u. 3000 i.u.	McCarthy & Cerecedo (1953)	300 i.u. 1000 i.u.
Vitamin D	300 i.u. sufficient unless Ca:P ratio < 1.0 or > 2.0 or P < 0.5% McCoy (1949)	1000 i.u.		1000 i.u.
Vitamin E	Rose & György (1950) (α -tocopherol)	30 mg 50 mg	Cerecedo & Vinson (1944) Lee, King & Visscher (1953)	20 mg 80 mg 60 mg
Vitamin K	Not essential unless intestinal synthesis inhibited	1 mg	Not normally essential	1 mg
Essential fatty acids (as linolenic acid)	Deuel & Reiser (1955) Thomasson (1953) Greenberg, Calvert, Savage & Deuel (1950)	2-20 g 10 g	Decker, Fillerup & Mead (1950)	Essential 5-10 g?

* Quantities in bold type are suggestions thought to prove optimal in most circumstances for most strains of these species.

Table 1 (*continued*)

Vitamin	Rat		Mouse	
	Reference	Quantity/ kg diet	Reference	Quantity/ kg diet
(b) Water-soluble vitamins				
Thiamine	Brown & Sturtevant (1949)	1.25 mg 2 mg	Morris (1947)	3.0 mg 3 mg
Riboflavin	Brown & Sturtevant (1949) El Sadr, Macrae & Work (1940)	2.5 mg 5.0 mg 5 mg	Morris (1947) Fenton & Cowgill (1947a)	1.5 mg 4.0-6.0 mg 8 mg
Calcium D- pantothenate	Brown & Sturtevant (1949)	10 mg 12 mg	Fenton, Cowgill, Stone & Justice (1950)	6-8 mg 10 mg
Vitamin B ₁₂	Cuthbertson & Thornton (1952) Emerson (1949)	15-20 µg 20-25 µg 30 µg	Jaffe (1950) Bosshardt, Paul & Barnes (1950) Bosshardt, Paul, O'Doherty, Huff & Barnes (1949)	5 µg 10-20 µg 15-30 µg 40 µg
Nicotinic acid (amide)	Not needed unless diet deficient in tryptophan Hundley (1947)	10-100 mg 10 mg	Probably not needed unless diet deficient in tryptophan	10 mg
Pteroyl- glutamic acid	Not needed unless gut synthesis prevented Asenjo (1948) Darke & White (1950)	0.3-2.0 mg	Essential, Nielsen & Black (1944) Essential for lactation, Fenton & Cowgill (1947b)	1 mg
Biotin	Not needed except in presence of raw egg white or if gut synthesis prevented Nielsen & Elvehjem (1941)	100-400 µg	Essential for lactation, Fenton & Cowgill (1947b)	80 µg 200 µg
Pyridoxine	McCoy (1949) Brown & Sturtevant (1949) Sarma, Snell & Elvehjem (1946)	1-2 mg 1.0 mg 1.5 mg 2 mg	Morris (1947) Fenton & Cowgill (1947b)	1 mg 5 mg 5 mg
Inositol	Not normally needed Cunha, Kirkwood, Phillips & Bohstedt (1943) Engel (1942)	3 g 0.3 g	Wooley (1941) Spitzer & Phillips (1946)	10-1000 mg
Choline	Hale & Schaefer (1951) Glynn, Himsorth & Lindan (1948) Conger & Elvehjem (1941) Engel (1942)	1-2 g 1 g	Mirone (1954)	Essential 1.5 g ?
p-amino- benzoic acid	Not normally required		Not normally required	

Table 2. *Trace-element and mineral requirements* of the rat*

Element	Reference	Requirement/ kg diet
Ca	Hubbell, Mendel & Wakeman (1937) McCoy (1949)	4.5 g 5.6 g 6.0 g
P	McCoy (1949) Hubbell <i>et al.</i> (1937)	3.5–4.5 g 3.5 g 4 g
Ca:P ratio	McCoy (1949)	1:2 1.5
K	Grijns (1938) Grunert, Meyer & Phillips (1950)	0.5 g 1.8 g 5 g
Na	Grunert <i>et al.</i> (1950)	0.5 g 5 g
Cl	McCoy (1949)	500 mg 3 g
Mg	Kunkel & Pearson (1948)	200 mg 500 mg
Fe	Waddell, Elvehjem, Steenbock & Hart (1928)	10–50 mg 50 mg
Cu	Hart, Steenbock, Waddell & Elvehjem (1928) Hundley (1950)	5 mg 15–20 mg 20 mg
Mn	Hill, Holtkamp, Buchanan & Rutledge (1950)	3–10 mg 20 mg
Zn	Hubbell & Mendel (1927) Todd, Elvehjem & Hart (1934)	2 mg 1.6 mg 4 mg
I	Remington (1932)	100–200 µg 200 µg
Br	Probably essential †	50 µg
F	Doubtful requirement	100 µg
Mo	De Renzo (1953)	40 µg 100 µg

* Quantities in bold type are suggestions thought to prove optimal in most circumstances for most strains of the rat.

† Shown to be essential for the mouse (Huff, Bosshardt, Miller & Barnes, 1956).

The nutrition of the rat and mouse has been reviewed by Farris (1950), McCoy (1949), Brown & Sturtevant (1949), Morris (1944, 1947) and Worden (1947).

Comparatively little is known of the requirements of the mouse for mineral elements and amino-acids, but the ability of mice to grow on diets primarily designed for rat nutrition suggests that their amino-acid needs are similar to those of the rat, and the work of Hubbell & Mendel (1927) and of Kemmerer, Elvehjem & Hart (1931) on zinc and manganese requirements suggests that the trace-mineral needs are also of the same order for the two species.

In certain circumstances (when animals are given high-calorie diets or thyroxine or receive certain drugs, such as sulphonamides, by mouth) the nutrient requirements for vitamins may be greatly increased. In Glaxo Laboratories the vitamin mixture shown in Table 4 has been found satisfactory for growth of the rat, for addition of this mixture in quantities beyond those indicated gave no further

Table 3. *Amino-acid requirements of the rat for growth and nitrogen balance*

Amino-acid	Reference	Requirement g/100 g diet	mg/rat/day
L-valine	Rose, Smith, Womack & Shane (1949)	0.7	
L-leucine	Rose <i>et al.</i> (1949)	0.8	
L-isoleucine	Rose <i>et al.</i> (1949)	0.5	
	Womack, Harlin & Lin (1953)	—	11
D- or L-	Forbes, Vaughan & Norton (1955)	0.4	
methionine	Rose <i>et al.</i> (1949)	0.6	
	Schweigert & Guthneck (1954)	0.22*	
	Womack <i>et al.</i> (1953)	—	14.6
			3.2*
L-threonine	Rose <i>et al.</i> (1949)	0.5	
	Womack <i>et al.</i> (1953)	—	10
D- or L-	Rose <i>et al.</i> (1949)	0.9	
phenylalanine	Armstrong (1955)	1.2	
	Armstrong (1955)	0.6†	
L-tryptophan	Rose <i>et al.</i> (1949)	0.2	
	Hundley (1947)	0.1‡	
L-lysine	Rose <i>et al.</i> (1949)	1.0	
L-histidine	Rose <i>et al.</i> (1949)	0.4	
L-arginine	Rose <i>et al.</i> (1949)	0.2	
Non-essential amino-N	Rose, Oesterling & Womack (1948)	To make 12.5% protein (N × 6.25)	

* Requirement in the presence of ample cystine.

† Requirement in the presence of ample tyrosine.

‡ Requirement in the presence of ample nicotinic acid (amide).

All these amino-acids have been shown to be essential for the mouse (Morris, 1944) though quantitative needs have not been determined.

increase in growth rate of animals subjected to a variety of nutritional stresses and maintained on 'semi-synthetic' rations based on casein and starch or glucose.

Fenton & Cowgill (1947b) have described a 'semi-synthetic' ration that is almost adequate for the whole life-cycle of the mouse and would probably wholly be so if vitamin B₁₂ were included.

Frequently the presence of a particular food component may greatly modify the nutrient need for a specific essential. Some of the more important interactions of this type are reviewed by Coates, Kon & Porter (1956).

Several diets have been proposed for use in the breeding and maintenance of rats and mice and are used for this purpose in different laboratories, and so would appear

Table 4. *Vitamin additions used in Glaxo Laboratories per kg semi-synthetic ration for the rat*

Vitamin A	4000 i.u.	Nicotinic acid	100 mg
Vitamin D (calciferol)	2000 i.u.	Vitamin B ₁₂	50 µg
Vitamin E (α -tocopherol acetate)	280 mg	Choline	1000 mg
Vitamin K (menaphthone)	2 mg	Pteroylglutamic acid	1.0 mg
Thiamine	30 mg	Biotin	0.2 mg
Riboflavin	30 mg	Inositol	220 mg
Pyridoxine	8 mg	p-aminobenzoic acid	75 mg
Ca D-pantothenate	100 mg		

Table 5. *Amount of certain major nutrients in some commonly used rat and mouse diets as calculated from food tables (mainly McCance & Widdowson, 1942 and Morrison, 1951)*

Nutrient	RBSS10*	Diet 41B (Bruce & Parkes, 1956)	Thompson (1936)
Protein (%)	16.8	15.5	21
Fat (%)	7.6	3.4	4
Calories (Cal./kg)	3739	3280	3100
Ca (%)	0.27	0.43	1.3
P (%)	0.43	0.58	1.0
Vitamin A (i.u./kg)	2700	9000	5000
Carotene (mg/kg)	0.0	0.05	0.5
Vitamin D (i.u./kg)	Almost none	2000	1000
Riboflavin (mg/kg)	6.7	2.7	5.3
Calcium D-pantothenate (mg/kg)	20	12	15
Nicotinic acid (amide) (mg/kg)	63	45	51
Vitamin B ₁₂ (μg/kg)	10	7.5	9
Choline (mg/kg)	1100	1100	1100
L-lysine (%)	0.98	0.85	1.1
Methionine and cystine (%)	0.7	0.6	0.75
Tryptophan (%)	0.25	0.2	0.26

* Stock-colony diet, Glaxo Laboratories Ltd.

to be satisfactory in practice, although it is not possible to say which, if any, of them allow maximum reproduction and growth. The quantities of some of the more important essential nutrients in these diets have been calculated from food tables (McCance & Widdowson, 1942; Morrison, 1951). The results of these calculations, presented in Table 5, show that the nutrient needs as quoted in the other tables are to a large extent satisfied by these diets, though in some of them the dietary supply may be marginal.

In estimating needs, no account has been taken of nutrient losses from the food. These can be important. Vitamin E deficiency has been occasionally noted to follow the use of diet 41 devised by Bruce & Parkes (1949). This is probably due to destruction of vitamin E by the cod-liver oil included. For this reason Bruce & Parkes, (1956) now recommend that diet 41B should be employed. In it a dry stabilized vitamin A and D mixture is used to eliminate risk of destruction of vitamin E by the unsaturated fish oil.

REFERENCES

- Armstrong, M. D. (1955). *J. biol. Chem.* **213**, 408.
 Asenjo, C. F. (1948). *J. Nutr.* **36**, 601.
 Bosshardt, D. K., Paul, W. J. & Barnes, R. H. (1950). *J. Nutr.* **40**, 595.
 Bosshardt, D. K., Paul, W. J., O'Doherty, K., Huff, J. W. & Barnes, R. H. (1949). *J. Nutr.* **37**, 21.
 Brown, R. A. & Sturtevant, M. (1949). *Vitam. & Horm.* **7**, 171.
 Bruce, H. M. & Parkes, A. S. (1956). *J. Anim. Tech. Ass.* **7**, 54.
 Bruce, H. M. & Parkes, A. S. (1949). *J. Hyg., Camb.* **47**, 202.
 Coates, M. E., Kon, S. K. & Porter, J. W. G. (1956). *Brit. med. Bull.* **12**, 61.
 Cerecedo, L. R. & Vinson, L. J. (1944). *Fed. Proc.* **3**, 55.
 Conger, T. W. & Elvehjem, C. A. (1941). *J. biol. Chem.* **138**, 555.
 Cramer, J. W. & Steenbock, H. (1956). *Arch. Biochem.* **63**, 9.
 Cunha, T. J., Kirkwood, S., Phillips, P. P. & Bohstedt, G. (1943). *Proc. Soc. exp. Biol., N.Y.*, **54**, 236.

- Cuthbertson, W. F. J. & Thornton, D. M. (1952). *Brit. J. Nutr.* **6**, 170.
- Darke, S. J. & White, C. (1950). *Brit. J. Nutr.* **4**, 9.
- Decker, A. B., Fillerup, D. L. & Mead, J. F. (1950). *J. Nutr.* **41**, 507.
- De Renzo, E. C. (1953). *Naturwissenschaften*, **40**, 251. *Nutr. Abstr. Rev.* **23**, 862.
- Deuel, H. J. Jr. & Reiser, R. (1955). *Vitam. & Horm.* **13**, 29.
- El Sadr, M. M., Macrae, T. F. & Work, C. E. (1940). *Biochem. J.* **34**, 601.
- Emerson, G. A. (1949). *Proc. Soc. exp. Biol., N.Y.*, **70**, 392.
- Engel, R. W. (1942). *J. Nutr.* **24**, 175.
- Farris, E. J. (1950). *The Care and Breeding of Laboratory Animals*. New York: Wiley.
- Fenton, P. F. & Cowgill, G. R. (1947a). *J. Nutr.* **34**, 273.
- Fenton, P. F. & Cowgill, G. R. (1947b). *J. Nutr.* **33**, 703.
- Fenton, P. F., Cowgill, G. R., Stone, M. A. & Justice, D. H. (1950). *J. Nutr.* **42**, 257.
- Forbes, R. M., Vaughan, L. & Norton, H. W. (1955). *J. Nutr.* **57**, 593.
- Glynn, L. E., Himsworth, H. P. & Lindan, O. (1948). *Brit. J. exp. Path.* **29**, 1.
- Greenberg, S. M., Calvert, C. E., Savage, E. E. & Deuel, H. J. Jr. (1950). *J. Nutr.* **41**, 473.
- Grijns, G. (1938). *Hoppe-Seyl. Z.* **251**, 97.
- Grunert, R. R., Meyer, J. H. & Phillips, P. H. (1950). *J. Nutr.* **42**, 609.
- Hale, O. M. & Schaefer, A. E. (1951). *Proc. Soc. exp. Biol., N.Y.*, **77**, 633.
- Hart, E. B., Steenbock, H., Waddell, J. & Elvehjem, C. A. (1928). *J. biol. Chem.* **77**, 797.
- Hill, R. M., Holtkamp, D. E., Buchanan, A. R. & Rutledge, E. K. (1950). *J. Nutr.* **41**, 359.
- Hubbell, R. B. & Mendel, L. B. (1927). *J. biol. Chem.* **75**, 567.
- Hubbell, R. B., Mendel, L. B. & Wakeman, A. J. (1937). *J. Nutr.* **14**, 273.
- Huff, J. W., Bosshardt, D. K., Miller, O. P. & Barnes, R. H. (1956). *Proc. Soc. exp. Biol., N.Y.*, **92**, 216.
- Hundley, J. M. (1947). *J. Nutr.* **34**, 253.
- Hundley, J. M. (1950). *Proc. Soc. exp. Biol., N.Y.*, **74**, 531.
- Jaffe, W. G. (1950). *Arch. Biochem.* **27**, 464.
- Kemmerer, A. R., Elvehjem, C. A. & Hart, E. B. (1931). *J. biol. Chem.* **92**, 623.
- Kunkel, H. O. & Pearson, P. B. (1948). *Arch. Biochem.* **18**, 461.
- Lee, Y. C. P., King, J. T. & Visscher, M. B. (1953). *Amer. J. Physiol.* **173**, 456.
- McCance, R. A. & Widdowson, E. M. (1942). *Spec. Rep. Ser. med. Res. Coun., Lond.*, no. 235.
- McCarthy, P. T., & Cerecedo, L. R. (1953). *J. Nutr.* **49**, 357.
- McCoy, R. H. (1949). In *The Rat in Laboratory Investigation*, 2nd ed., p. 68. [E. J. Farris and J. Q. Griffiths, editors.] Philadelphia: J. B. Lippincott Co.
- Mirone, L. (1954). *Amer. J. Physiol.* **179**, 49.
- Morris, H. P. (1944). *J. nat. Cancer Inst.* **5**, 115.
- Morris, H. P. (1947). *Vitam. & Horm.* **5**, 175.
- Morrison, F. (1951). *Feeds and Feeding*. Ithaca: Morrison Publishing Co.
- Nielsen, E. & Black, A. (1944). *J. Nutr.* **28**, 203.
- Nielsen, E. & Elvehjem, C. A. (1941). *Proc. Soc. exp. biol., N.Y.*, **48**, 349.
- Remington, R. E. (1932). *J. biol. Chem.* **97**, ci.
- Rose, C. S. & György, P. (1950). *Proc. Soc. exp. Biol., N.Y.*, **74**, 411.
- Rose, W. C., Oesterling, M. J. & Womack, M. (1948). *J. biol. Chem.* **176**, 753.
- Rose, W. C., Smith, L. C., Womack, M. & Shane, M. (1949). *J. biol. Chem.* **181**, 307.
- Rubin, S. H. & De Ritter, E. (1954). *Vitam. & Horm.* **12**, 101.
- Sarma, P. S., Snell, E. & Elvehjem, C. A. (1946). *J. biol. Chem.* **165**, 55.
- Schweigert, B. S. & Guthneek, B. T. (1954). *J. Nutr.* **54**, 333.
- Spitzer, R. R. & Phillips, P. H. (1946). *Proc. Soc. exp. Biol., N.Y.*, **63**, 10.
- Thomasson, H. (1953). *Int. Rev. Vitam. Res.* **25**, 62.
- Thompson, W. (1936). *J. Hyg., Camb.*, **36**, 24.
- Todd, W. R., Elvehjem, C. A. & Hart, E. B. (1934). *Amer. J. Physiol.* **107**, 146.
- Waddell, J., Elvehjem, C. A., Steenbock, H. & Hart, E. B. (1928). *J. biol. Chem.* **77**, 777.
- Womack, M., Harlin, H. A. & Lin, P. H. (1953). *J. Nutr.* **49**, 513.
- Wooley, D. W. (1941). *J. biol. Chem.* **139**, 29.
- Worden, A. N. (editor) (1947). *The UFAW Handbook on the Care and Management of Laboratory Animals*. London; Ballière, Tindall & Cox.