Breast-milk fat concentrations of rural African women
2. Long-term variations within a community

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1. Long-term variations in breast-milk fat concentration of mothers feeding on demand were studied in 120 rural West African women over a 12-month period.

2. The over-all mean 12 h breast-milk fat concentration was 39·3 g/l.

3. Mean breast-milk fat concentrations were affected by season in a manner which was correlated with seasonal changes in maternal subcutaneous fat stores ($P < 0.05$) but which was unrelated to seasonal variations in maternal energy intake and breast-milk output.

4. Breast-milk fat concentrations were highest in early lactation, decreasing to a constant level during the first year.

5. There was significantly greater between-mother than within-mother variation in breast-milk fat concentrations measured in successive months, after correcting for season and stage of lactation ($P < 0.001$).

6. Breast-milk fat concentrations were highest for primiparous mothers, decreasing to a constant level at parity 4 and higher.

7. A mother's relative breast-milk fat concentration was not correlated with her levels of dietary energy intake and breast-milk output but was positively correlated with her relative subcutaneous fat deposits ($P < 0.01$).

Investigations in rural West Africa into the relationship between maternal lactational performance and infant nutritional status have demonstrated that breast-milk output is influenced by a number of factors, including stage of lactation and season of the year, and that there are consistent, long-term differences in the breast-milk yield of individuals (Whitehead *et al.* 1978; Prentice, 1980). It was possible that such variations in breast-milk volume might be compensated for by reciprocating variations in breast-milk composition. Changes in fat concentration were of prime interest in this respect as fat is the major energy source of breast-milk and is the most variable of the proximate constituents.

The study discussed in this paper was designed to determine whether long-term variations in breast-milk fat concentration occur among underprivileged women feeding on demand and to examine to what extent these variations are related to breast-milk volume. In addition, the influence of maternal energy intake and subcutaneous fat stores on breast-milk fat concentrations was assessed.

EXPERIMENTAL

Experimental design

All lactating women in the rural, farming community of Keneba, The Gambia, were studied from 1 to 18 months post-partum over a 12-month period from January to December 1979. In any 1 month there was an average of seventy-two subjects and there was an approximately even distribution of mothers in different trimesters of lactation and of different parity (1–10). In total 120 mothers took part in the study. All mothers practised feeding on demand. The investigation formed part of a long-term study of nutrition and lactation in this community; all aspects of these studies had the approval of the Ethical Committees of the Dunn Nutrition Laboratory, Cambridge, and the Gambian Government.

Breast-milk fat concentrations of each subject were determined on 1 d in every month as described later. Maternal anthropometry and 12 h breast-milk output measurements were performed on the same day. All these procedures were carried out at the routine 12 h breast-milk estimation sessions which have been described in detail by Prentice (1980). In addition, 24 h maternal energy intakes were determined by direct-weighing on 4 d in each month (Prentice, 1980). A carefully quantitated maternal dietary supplementation programme, which involved all the subjects of the present study, began on 1 May 1979. Full details of this programme have been given by Prentice *et al.* (1980).

Breast-milk fat determinations

Small samples of milk (approximately 0·25 ml) were collected by maternal expression from each breast of the subject before and after the first feed occurring after 13.00 hours. Aliquots (100 μl) from the four samples were pooled and the creamatocrit (Lucas *et al.* 1978) of the resulting mixture was determined in triplicate. The creamatocrit value was converted to fat concentration using the following equation (Prentice *et al.* 1981):

$$\text{fat concentration} = \text{creamatocrit} \times 6{\cdot}48 - 1{\cdot}46 \text{ g/l}.$$

The concentration of fat in the sample collected at the first feed after 13.00 hours has been shown to be related to the 12 h (07.00–19.00 hours) and 24 h mean fat concentrations (Prentice *et al.* 1981). The conversion equations determined for use in this community were as follows:

$$12 \text{ h mean fat level} = \text{mean fat level (13.00 hours)} \times 0{\cdot}66 + 14{\cdot}4 \text{ g/l},$$

$$24 \text{ h mean fat level} = \text{mean fat level (13.00 hours)} \times 0{\cdot}43 + 21{\cdot}0 \text{ g/l}.$$

The continuing validity of these conversion equations was confirmed by determining the 24 h variations in breast-milk fat concentration of sixteen women towards the end of the 12-month period.

Treatment of results

Where appropriate breast-milk fat concentrations were corrected for season of the year by expressing each result as a percentage of the total mean value in the community during the month when the measurement was made. When allowance for stage of lactation was also required, each result was expressed as a percentage of the monthly mean breast-milk fat concentration of those subjects in the same trimester of lactation as the individual concerned.

RESULTS

A considerable variation in breast-milk fat concentration was observed both between mothers and between measurements made in successive months on the same individual. It was possible to identify a number of factors which influenced the variations in breast-milk fat concentration.

Season of the year

Season of the year had a pronounced effect on breast-milk fat concentration (Fig. 1). The monthly mean 12 h fat concentration (g/l) ranged from 36·2 to 42·8 during 1979 with an over-all mean value of 39·3. The corresponding range of 24 h values (g/l) was 35·2–39·5 with a total mean of 37·3. Similar seasonal changes in breast-milk fat concentration were found when mothers at different stages of lactation were considered separately. The variations were most marked during the first trimester, reducing in magnitude as lactation progressed. Within each calendar month there was a wide range of individual fat concentrations; the mean monthly coefficient of variation was 21.

The factors influencing the seasonal variation in breast-milk fat concentration could not be fully elucidated. The changes in monthly mean breast-milk fat concentration followed the seasonal variations in mean maternal subcutaneous fat stores as estimated by triceps

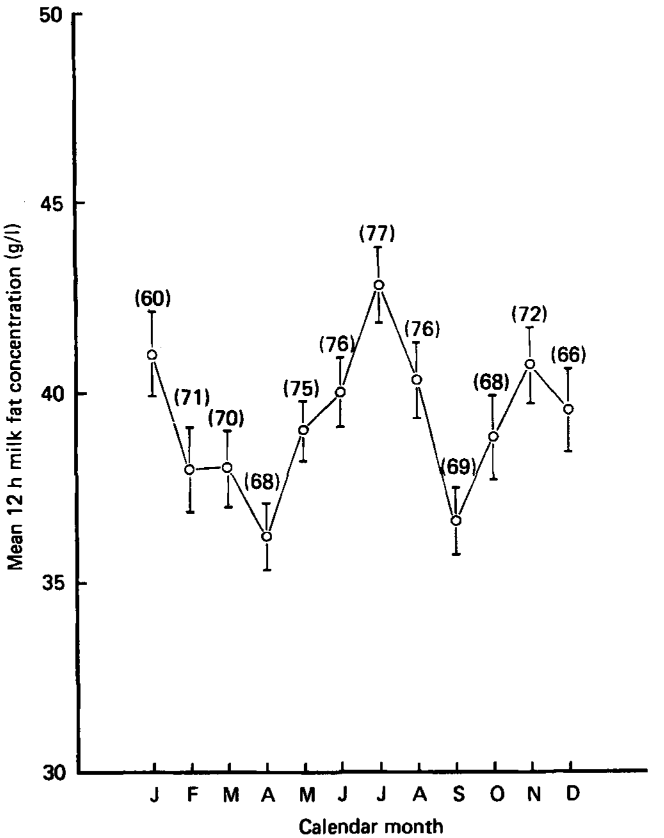

Fig. 1. Seasonal variation in 12 h (07.00–19.00 hours) breast-milk fat concentration (g/l) of rural African women during 1979. Points represent mean values, with their standard errors represented by vertical lines. Number of subjects given in parentheses.

skinfold thickness (r 0·57, n 12, $P < 0{\cdot}05$). No correlation was observed, however, between the monthly mean breast-milk fat concentration and mean maternal energy intake even when the months before and after the start of the supplementation programme were considered separately. Similarly there was no relationship between the seasonal changes in mean breast-milk fat concentration and mean breast-milk output.

There are two distinct seasons in Keneba. The rainy season, lasting from June to October, is accompanied by a period of severe nutritional restriction, a substantial increase in heavy farmwork and an increased incidence of disease (Paul *et al.* 1979; Prentice, 1980). The mean breast-milk output of mothers in this community is markedly reduced during the rainy season, the minimum values occurring in September–October (Whitehead *et al.* 1978; Prentice, 1980). As there was no relationship between monthly mean breast-milk fat concentration and breast-milk output, no mechanism was evident whereby an increase in breast-milk energy content automatically offset the decrease in milk available to the infants during the rainy season. Indeed, in 1979 there was a considerable decrease in mean breast-milk fat concentration in September (Fig. 1) which coincided with the period of minimum breast-milk output. The average 12 h breast-milk fat intake of Keneba infants, therefore, was reduced dramatically at this time falling from 13·5 g in July to 9·7 g in September.

Table 1. *Effect of stage of lactation on breast-milk fat concentration*

(Results are the average difference from the mean fat concentration for all subjects in each calendar month. The standard error of the mean and the number of subjects are shown in parentheses)

	Trimester of lactation					
Calendar month	1	2	3	4	5	6
January	+0·9 (1·6, 22)	+4·1 (4·2, 8)	−2·3 (2·1, 10)	+0·1 (2·2, 10)	−3·7 (2·9, 6)	−3·5 (0·6, 3)
February	+2·9 (3·3, 17)	+1·3 (2·2, 12)	+0·3 (3·8, 8)	−0·7 (1·9, 12)	−3·2 (1·3, 14)	−2·7 (3·7, 7)
March	+1·7 (2·2, 11)	−1·4 (1·3, 18)	+4·0 (4·4, 7)	−1·4 (3·9, 8)	−2·3 (2·3, 14)	+0·9 (3·2, 11)
April	+3·2 (2·8, 9)	+0·8 (2·1, 18)	−3·6 (2·3, 8)	−0·8 (1·9, 9)	+0·9 (1·8, 12)	−1·4 (1·3, 12)
May	+1·8 (1·8, 12)	+2·4 (1·8, 17)	−1·8 (1·5, 10)	+0·3 (3·2, 7)	−1·8 (1·9, 16)	−0·8 (2·0, 13)
June	+4·2 (2·7, 9)	−0·7 (1·7, 16)	−0·7 (1·8, 18)	−0·7 (5·3, 5)	+0·7 (2·2, 13)	−0·5 (2·2, 14)
July**	+6·6 (2·6, 9)	+1·5 (2·4, 14)	−0·6 (1·8, 19)	−3·1 (3·2, 9)	−0·4 (2·8, 11)	−1·9 (1·9, 14)
August	+3·1 (2·4, 10)	+2·4 (2·5, 15)	−1·2 (2·1, 18)	−2·4 (2·2, 11)	−1·8 (3·5, 6)	−0·4 (2·4, 16)
September	+1·8 (2·2, 9)	+2·6 (2·5, 10)	−2·0 (1·4, 13)	−4·8 (1·2, 18)	+0·4 (2·2, 6)	+3·0 (2·4, 15)
October*	+4·1 (1·8, 8)	−2·2 (2·1, 11)	+2·7 (4·1, 13)	−1·1 (1·6, 18)	−3·0 (1·8, 7)	−0·3 (2·5, 11)
November**	+5·9 (3·2, 9)	−0·3 (2·7, 12)	+3·8 (3·0, 11)	−2·0 (1·9, 18)	−4·2 (1·4, 11)	−0·8 (2·0, 9)
December***	+8·9 (2·7, 14)	−0·8 (2·5, 9)	−2·8 (2·5, 10)	−0·9 (2·6, 11)	−4·9 (1·6, 14)	−1·7 (3·7, 8)

t test between trimesters 1 and 4–6: $^*P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$.

Stage of lactation

Within each calendar month there was a tendency for breast-milk fat concentrations to be higher for women in early lactation (Table 1), but this trend rarely reached statistical significance. The decrease in milk fat concentration during the first months of lactation was confirmed by studying a cohort of mothers who were in the first trimester post-partum early in 1979. The mean breast-milk fat concentration of these mothers, corrected for season, decreased steadily throughout the year, falling by 17% between the first and fourth trimester (Fig. 2). This decrease was shown to be significant by a paired *t* test (t 2·94, df 22, $P < 0.01$). When mothers in the third trimester post-partum at the beginning of the year were examined in a similar fashion no significant differences were found between the seasonally corrected mean breast-milk fat concentration in any trimester from 3 to 6.

The mean breast-milk volume of mothers in Keneba declines steadily with increasing stage of lactation after an initial peak at 2–3 months post-partum (Prentice, 1980). Thus the milk fat intakes of the infants were lowered substantially with increasing age as a result of the decrease in both breast-milk fat concentration and breast-milk volume.

Characteristic breast-milk fat levels throughout lactation

When corrected for season and stage of lactation, the breast-milk fat concentration of an individual measured in successive months was found to have a characteristic value that was consistently maintained throughout the year. This was shown by one-way analysis of

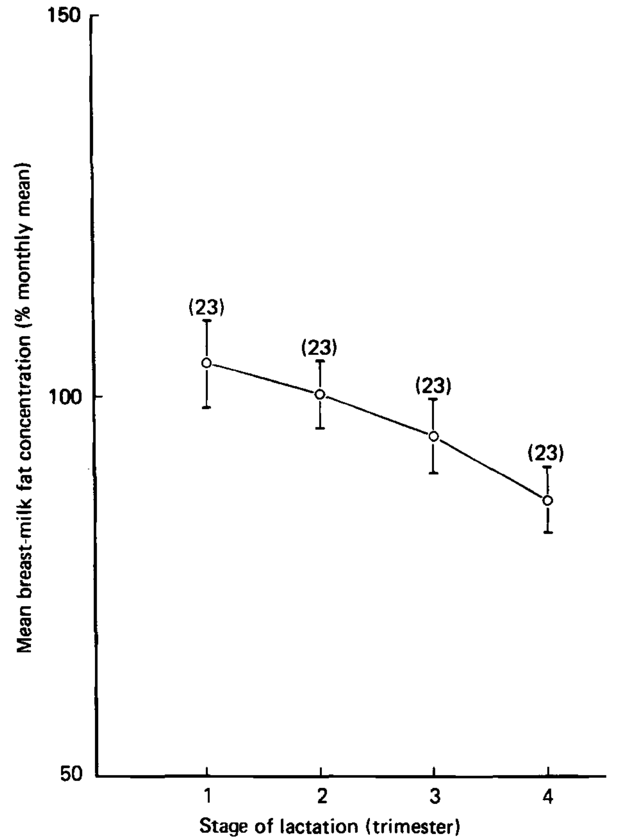

Fig. 2. Variation in seasonally corrected 12 h (07.00–19.00 hours) breast-milk fat concentration (% of monthly mean) with stage of lactation (trimester) during the first year post-partum in rural African women. The points represent mean values of a cohort of mothers who were in the first trimester post-partum early in 1979, with their standard errors represented by vertical bars. Number of subjects indicated in parentheses.

variance which demonstrated a significantly greater between-mother than within-mother variation in corrected breast-milk fat concentration (F 3·72 on 100 and 663 df, $P < 0·001$).

The characteristic breast-milk fat concentration of each mother relative to the community (F_R) was assessed by determining the mean corrected breast-milk fat concentration of the individual over the 12 months of the study. Relative values of other parameters discussed later were estimated in a similar way.

The relationships between an individual's F_R value and a number of other factors were investigated in detail.

Parity. Primiparous women were found to have consistently greater F_R values than mothers of higher parity (Fig. 3). The mean F_R value decreased steadily with increasing parity, reaching a constant level at parities greater than 4. A 25% decrease in mean F_R was noted between primiparous mothers and those of parity 4 and higher; this difference was highly significant (t 7·28, df 68, $P < 0·001$).

Breast-milk volume. The mothers in this study were shown by one-way analysis of variance to have a consistent, characteristic level of breast-milk output during successive months once individual values were corrected for season and stage of lactation (F 6·81 on

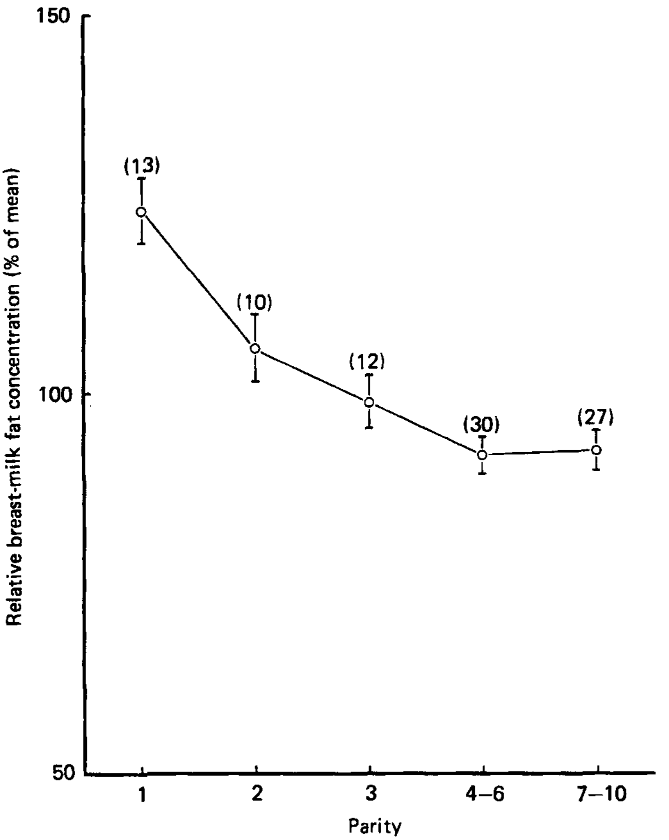

Fig. 3. Variation in relative breast-milk fat concentration (% of monthly mean for women in the same trimester) with parity in rural African women. The points represent mean values, with their standard errors represented by vertical bars. Number of subjects indicated in parentheses.

89 and 586 df, $P < 0.001$). This finding is in agreement with observations made previously in Keneba (Whitehead *et al.* 1978).

No effect of parity on an individual's relative breast-milk volume (V_R) could be demonstrated in this study despite earlier work in this community which had indicated a higher breast-milk output at 6–9 months post-partum for primiparous mothers (Whitehead *et al.* 1978).

No correlation was found between an individual's F_R and V_R values, either when all subjects were considered or when mothers of parity 4 and higher were taken separately to remove the effect of parity on F_R. In addition, there were no significant differences in the monthly mean breast-milk fat concentration of women with low (< 80% of mean), average and high (> 120% of mean) V_R values.

Maternal energy intake. The daily energy intake of the mothers was found to vary considerably with the season of the year but was not affected by stage of lactation. After correcting for seasonal variation, one-way analysis of variance demonstrated a significantly greater between-mother than within-mother variation in daily energy intake (F 6·02 on 70 and 327 df, $P < 0.001$). No effect of parity on maternal energy intake could be shown.

No relationship was found between an individual's relative energy intake (E_R) and F_R value even when mothers of parity 4 and higher were considered separately. There were no significant differences in the monthly mean breast-milk fat concentration of mothers with low (< 80% of mean), average or high (> 120% of mean) E_R values. In addition, no correlation was observed between an individual's E_R and V_R values.

Maternal subcutaneous fat deposits. The triceps skinfold thickness of the mothers showed a marked seasonal variation but no effect of stage of lactation was evident. Using results corrected for season, one-way analysis of variance showed that an individual exhibits a characteristic triceps skinfold thickness relative to other mothers in the community (F 38·11 on 82 and 450 df, $P < 0.001$). The relative triceps skinfold thickness (T_R) of subjects was not related to parity.

A significant, positive correlation was observed between an individual's T_R and F_R values (r 0·27, n 87, $P < 0.01$); the correlation coefficient improved when mothers of parity 4 and higher were considered separately (r 0·38, n 51, $P < 0.01$). No relationships were found, however, between an individual's T_R and either her V_R or E_R values.

DISCUSSION

Previous investigations of factors influencing the fat concentration of breast-milk have been severely handicapped by the difficulty of obtaining milk samples that give a true reflection of an individual's mean breast-milk fat concentration and by the relatively large volumes of milk required for accurate fat analysis. Such problems have resulted in criticism of a number of previous studies and have precluded any extensive investigations of long-term variations in breast-milk fat concentration. The sampling procedure and method of fat analysis developed for use in this study (Prentice *et al.* 1981) provide a simple, reliable measure of individual daily mean breast-milk fat concentrations which requires only very small quantities of milk and which involves the minimum of interference with the normal course of lactation. This method has permitted the detailed long-term study of the breast-milk fat levels of a large group of underprivileged women feeding on demand.

Season of the year and stage of lactation were shown to have a profound effect on breast-milk fat concentration. In both instances fat concentrations decreased substantially at times when breast-milk volumes were declining, thus magnifying the nutritional problem faced by the breast-fed child. This finding conflicts with a number of previous studies which reported either a constant fat concentration throughout lactation after an initial rise (Peters, 1953; Roels & Trout, 1957; Belavady & Gopalan, 1959; Karmarkar *et al.* 1959) or an increase as lactation progresses (Holemans *et al.* 1954; Underwood *et al.* 1970; Abdel Kader *et al.* 1972). However, interpretation of many published results is difficult in view of the small number of subjects involved and the sampling procedures employed.

The over-all mean 12 h breast-milk fat concentration of 39·3 g/l obtained in this community is close to that reported for well-nourished women feeding on fixed schedules (Department of Health and Social Security, 1977). The small difference may be due in part to the higher proportion of primiparous women in privileged communities. The wide range of fat values observed is also similar to that found in Western societies (Morrison, 1952; Hytten, 1954*b*). Several studies of undernourished women have reported considerably lower mean breast-milk fat concentrations than that of well-nourished mothers (Janz & Pinto, 1957; Roels & Trout, 1957; Jansen *et al.* 1960; Venkatachalam, 1962; Bailey, 1965; Lindblad & Rahimtoola, 1974; El Tom Ali & Zaki, 1976) while others have shown no marked differences (Peters, 1953; Holemans *et al.* 1954; Walker *et al.* 1954; Belavady & Gopalan, 1959; Karmarkar *et al.* 1959; Hanafy *et al.* 1972).

In a study of eleven subjects, Hytten (1954*a*) found a close correlation between the breast-milk fat concentrations of samples collected from an individual on the seventh day post-partum and on one occasion later in lactation. The significantly greater between-mother than within-mother variation in corrected breast-milk fat concentrations observed in the present study extends Hytten's finding, demonstrating that an individual maintains a characteristic level of milk fat concentration throughout the whole of a single lactational period once variations due to season and to stage of lactation have been taken into account.

A major influence on an individual's level of breast-milk fat concentration was shown to be the parity of the mother, with primiparous mothers having considerably higher values than multiparous women after correction for season and stage of lactation. This decrease in milk fat concentration with increasing parity is in agreement with the results of other studies (Nims *et al.* 1931; Belavady & Gopalan, 1959; Venkatachalam, 1962). The characteristic level of breast-milk fat concentration was also found to be related positively to the individual's triceps skinfold thickness but not to her dietary energy intake relative to other members of the community. This suggests that a woman's metabolic efficiency is a dominating factor in determining her breast-milk fat concentration.

It was also established that a woman maintains a characteristic level of breast-milk output throughout lactation. No relationship was found between the characteristic levels of milk fat concentration and milk volume produced by an individual. The absence of such a relationship leads to a far wider range of breast-milk fat outputs than would be predicted from breast-milk volume measurements alone. In some mothers the combination of a poor milk volume and a low milk fat concentration results in exceptionally low fat intakes by their infants. Alternatively, a mother whose breast-milk output would normally be considered inadequate may produce milk of a sufficiently high fat concentration to compensate in energy terms for her low milk volume.

In conclusion, therefore, it is not valid to assume that the breast-milk composition of mothers is homogeneous within a community. Thus both the quality and the quantity of breast-milk must be determined in any assessment of the nutrition of breast-fed infants.

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REFERENCES

Abdel Kader, M. M., Bahgat, R., Aziz, M. T., Hefnawi, F., Badraoui, M. H. H., Younis, N. & Hassib, F. (1972). *J. Biosoc. Sci.* **4**, 403.
Bailey, K. V. (1965). *J. trop. Pediat.* **11**, 35.
Belavady, B. & Gopalan, C. (1959). *Ind. J. med. Res.* **47**, 234.
Department of Health and Social Security. (1977). *Rep. Health Soc. Subj.* no. 12.
El Tom Ali, K. & Zaki, J. (1976). *Sudan J. Fd Sci. Technol.* **8**, 12.
Hanafy, M. M., Morsey, M. R. A., Seddick, Y., Habib, Y. A. & El Lozy, M. (1972). *J. trop. Pediat. env. Child Hlth* **18**, 187.
Holemans, K., Lambrechts, A. & Martin, H. (1954). *Revue méd Liège* **9**, 714.
Hytten, F. E. (1954*a*). *Br. med. J.* i, 249.
Hytten, F. E. (1954*b*). *Br. med. J.* i, 253.
Jansen, A. A. J., Luyken, R., Malcolm, S. H. & Williams, J. J. L. (1960). *Trop. geogr. Med.* **2**, 138.
Janz, G. J. & Pinto, G. L. (1957). *Proc. 3rd Inter-African Nutr. Conf. Angola*, p. 133.
Karmarkar, M. G., Kapur, J., Deodhar, A. D. & Ramakrishnan, C. V. (1959). *Ind. J. med. Res.* **47**, 344.
Lindblad, B. S. & Rahimtoola, R. J. (1974). *Acta paediat., Stockh.* **63**, 125.
Lucas, A., Gibbs, J. A. H., Lyster, R. L. J. & Baum, J. D. (1978). *Br. med. J.* i, 1018.
Morrison, S. D. (1952). *Tech. Commun. Commonw. Bur. Anim. Nutr.* no. 18.
Nims, B., Macy, I. G., Brown, M. & Hunscher, H. A. (1931). *Am. J. Dis. Child.* **42**, 569.
Paul, A. A., Muller, E. M. & Whitehead, R. G. (1979). *Trans. R. Soc. trop. Med. Hyg.* **73**, 686.
Peters, F. E. (1953). *Br. J. Nutr.* **7**, 208.
Prentice, A., Prentice, A. M. & Whitehead, R. G. (1981). *Br. J. Nutr.* **45**, 483.
Prentice, A. M. (1980). In *Maternal Nutrition in Pregnancy and Lactation*, p. 167 [H. Aebi and R. G. Whitehead, editors]. Berne: Hans Huber.
Prentice, A. M., Whitehead, R. G., Roberts, S. B., Paul, A. A., Watkinson, M., Prentice, A. and Watkinson, A. A. (1980). *Lancet* ii, 886.

Roels, O. A. & Trout, M. (1957). *Proc. 3rd Inter-African Nutr. Conf. Angola*, p. 805.
Underwood, B. A., Hepner, R. & Abdullah, H. (1970). *Am. J. clin. Nutr.* **23**, 400.
Venkatachalam, P. S. (1962). *Dept. Publ. Hlth Monogr. Port Moresby*, no. 4.
Walker, A. R. P., Arvidsson, U. B. & Draper, W. L. (1954). *Trans. Roy. Soc. trop. Med. Hyg.* **48**, 395.
Whitehead, R. G., Rowland, M. G. M., Hutton, M., Prentice, A. M., Muller, E. & Paul, A. (1978). *Lancet* ii, 178.

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