

Do combined elimination diet and prolonged breastfeeding of an atopic infant jeopardise maternal bone health?

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Summary

Background Prolonged breastfeeding, and intervention in the maternal diet during breastfeeding, have been suggested as a means to treat atopic symptoms of infants. The impact of these actions on the mother's skeletal health has remained unclear.

Objective The purpose of this study was to determine factors that are associated with bone mineral density (BMD) and serum lipid fatty acid profile in mothers of atopic infants.

Methods BMD at the lumbar spine, right femoral neck and dominant distal radius of 24 mothers of atopic infants (study group) and 25 mothers of healthy infants (control group) were measured with dual energy X-ray absorptiometry postpartum after the resumption of menses. In addition, the fatty acid profile of their serum cholesteryl esters was determined.

Results A systematic trend for 4–6% lower BMD was observed in the study group, but this difference was mainly attributable to a difference in body weight between the groups. In addition, the omega (ω)-3 fatty acid metabolite EPA showed a positive association with femoral neck BMD. Mothers of atopic infants had a significantly lower proportion of ω -6 fatty acid parent LA and a higher proportion of ω -6 fatty acid metabolite GLA than the control mothers. These differences were associated not only with dietary factors but also with the duration of postpartum amenorrhea and total lactation.

Conclusion Neither the breastfeeding nor the mother's short-term elimination diet *per se* seemed to be associated with low BMD in the mothers of atopic infants. Thus, if an elimination diet benefits the infant, it can be recommended for a limited period. However, it may be advisable to give special dietary counselling, especially to low body weight mothers, in order to ensure adequate nutrient intake and to minimize the possible risk of potential bone loss.

Keywords: breastfeeding, bone mineral density, elimination diet, essential fatty acid, postpartum

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Introduction

Lactation is a demanding process in terms of nutritional requirements. It has been recommended that breastfeeding women should increase their energy and nutrient intake above the non-pregnant levels [1]. Lactating mothers secrete about 200 mg calcium into breast milk every day

[2]. In principle, an inadequate calcium supply may therefore result in maternal bone loss or reduced breast milk calcium secretion, with effects on the growth and bone development of the breast-fed baby as well. The relationship between diet and maternal bone changes during lactation is inconclusive [3].

This issue arises frequently in clinical practice as both the prevalence and severity of allergic diseases are increasing [4]. Prolonged breastfeeding is recommended

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for infants genetically predisposed to allergic disease [5,6]. In addition, elimination diets designed for breastfeeding mothers of atopic children form a common strategy to prevent and treat allergy of the child [7,8]. Unfortunately, allergies to foods constituting the major source of daily nutrients (i.e. milk, egg, fish and wheat) predominate in early childhood [9]. Thus, their elimination from the diet may increase the risk of general nutritional deficiency of essential nutrients (e.g. dietary fatty acids) and possibly jeopardise the mother's health [10–12].

A recent increase in the prevalence of atopic diseases has been linked to altered consumption of polyunsaturated fatty acids. For example, reduced consumption of omega (ω)-3 fatty acids in conjunction with increased consumption of ω -6 fatty acids may have contributed to this change in prevalence [11]. The maternal diet during breastfeeding may well modify the fatty acid composition of the breast milk [13], and in fact, the breast milk of mothers of children with newly developed atopic disease is known to have changed fatty acid profiles [14]. Fatty acids not only take part in the regulation of cell-mediated immune mechanisms [15,16], but they are also recognized as important regulators of calcium metabolism [17]. In this respect, fish as a rich source of ω -3 fatty acids [10] and as a moderate source of calcium, other minerals and vitamin D, may play some role. All these nutrients are central to bone health.

The purpose of this study was therefore to evaluate the potential association between elimination diets and prolonged breastfeeding with maternal bone mineral density (BMD) and fatty acid profile of serum cholesteryl esters. The relevant clinical question of this study is: can infants sensitized during exclusive breastfeeding be treated with an elimination diet in the mother without compromising her bone health?

Materials and methods

Subjects

Twenty-five mothers of atopic infants (study group) were recruited from the Tampere University Hospital to which their infants had been admitted for atopic dermatitis and food allergy. Eighteen of 24 atopic infants had a hereditary risk for allergy. One of the mothers used medication (corticosteroids) and was excluded from the study. In attempts to relieve atopic symptoms in the infants during exclusive lactation, 19 of 24 mothers temporarily modified their diets; six mothers eliminated common allergenic foods such as fish, eggs and peanuts, six mothers eliminated cow's milk in addition to the above, and seven mothers eliminated cow's milk and wheat. By resumption

of menstruation, only five mothers had not been on an elimination diet at all.

A group of 25 mothers of healthy infants formed the control group. These women were recruited from the mothers' health care unit in Kangasala Health Centre close to Tampere region.

All mothers in both groups were ambulant with no history of excessive exercise, caffeine or alcohol consumption, or periods of immobilization. None of the study mothers smoked, whereas four control mothers were smokers.

The study protocol was approved by The Ethical Committee of Tampere University Hospital and UKK Institute in Tampere, and written informed consent was obtained from the subjects.

Methods

According to protocol, BMD was to be measured during the second postpartum bleeding after the resumption of menses. In fact, it was measured so in 37 mothers, during the first postpartum bleeding in three study mothers, and during the third bleeding in three control mothers and six study mothers. BMD was determined at the lumbar spine (L2–4), right femoral neck and dominant distal radius with dual energy X-ray absorptiometry (DXA, XR-26, Norland Corp., Fort Atkinson, WI, USA) by the same experienced laboratory technician according to standard procedures [18]. In the laboratory, the *in vivo* precision of the BMD measurements is 0.7% for the lumbar spine, 0.5% for the femoral neck and 0.8% for the distal radius [18].

The mothers' body height and weight were measured during the DXA visit. The mothers also completed a questionnaire on health, lactation history, dietary habits and physical activity, and a one-week calcium intake diary [19]. The duration of exclusive lactation was not obtained from one mother in the study group. Data on calcium intake were obtained from 22 of 25 mothers in the control group, and from 18 of 24 mothers in the study group. Calcium intake was analysed with Micro-Nutrica software (Social Insurance Institution, Helsinki, Finland).

At the DXA visit, venous blood samples were collected for fatty acid analysis. Fatty acids of chain lengths ranging from 14:0 to 24:1 in serum cholesteryl esters were identified. Fatty acids of a special class of polyunsaturated fatty acids known as ω -3 fatty acids [α -linolenic acid 18 : 3n3 (ALA), eicosapentaenoic acid 20 : 5n3 (EPA)] and ω -6 fatty acids [linoleic acid 18 : 2n6 (LA), γ -linolenic acid 18 : 3n6 (GLA), dihomo- γ -linolenic acid 20 : 3n6 (DGLA), arachidonic acid 20 : 4n6 (AA)] were determined. The method used in the fatty acid analysis has been described elsewhere [20]. In short, lipids were extracted from the serum sample with chloroform–methanol (2:1).

Lipid fractions (cholesteryl esters, triglycerides and phospholipids) were separated with an aminopropyl column. Fatty acids of lipid fractions transmethylated with boron-trifluoride were analysed with gas chromatography (Hewlett-Packard 5890 series II, Hewlett-Packard Company, Waldbronn, Germany) with FFAP-column helium as carrier gas. Fatty acids were presented as molar percentages of total fatty acids.

The extent and severity of the infants' atopic eczema was assessed by the SCORAD method [21], the severity being the sum of the individual scores for extent of eczema (A), intensity of eczema (B), and the subjective manifestations including pruritus and sleep loss based on parents' evaluation (C). The growth of all infants was followed during the first year of life from birth until 12 months of age.

Statistical analysis

Mean and standard deviations (SD) and 95% confidence intervals (95% CI) are given as descriptive statistics. The clinical characteristics, BMD and fatty acid data were normally distributed in both groups. The differences in clinical characteristics between the groups were tested with unpaired *t*-test and χ^2 -test, as appropriate.

The difference in BMD between the study group and control group was defined as a z-score [(study patient BMD – mean control BMD)/control SD]. The z-scores for the fatty acid proportions were calculated in an analogous way. Preliminary calculation suggested that the total sample of 50 would provide an 80% statistical power to detect a z-score difference of about –0.8 at a significance level of $P = 0.05$.

To find predictors for significant z-scores (zero not included in the 95% CI) in site-specific BMD values, multiple regression analyses based on all possible subsets

regression using criterion of Mallows' C_p were performed. The z-score was used as a dependent variable, and continuous variables [age, weight, duration of exclusive lactation (= duration of lactation without introduction of formula or solid foods), duration of total lactation (= duration of any lactation until the DXA visit), duration of postpartum amenorrhea, and serum fatty acid levels] and categorical variables [consumption of fish (1 = not weekly, 2 = weekly) and trial of the elimination diet (0 = no, 1 = yes)] were used as independent variables. Due to missing data, a separate analysis was performed including maternal dietary calcium in the regression analysis in addition to the above-mentioned variables. However, the predictors for significant z-scores in fatty acid proportions were determined as described above, excluding the use of fatty acid levels as independent variables.

Results

Clinical characteristics

The study group and the control group were comparable in age, parity, weight, height, BMI and age at menarche (Table 1). As could be expected, the duration of exclusive lactation was significantly ($P = 0.003$) longer in the mothers of atopic infants. The birth weights of the babies were similar, being 3722 (± 359) g for atopic infants and 3732 (± 603) g for healthy infants. Half of the infants were boys. Likewise, the growth of the atopic and healthy infants during the first year of life was virtually the same, the body weights at the age of one year being 9809 (± 1090) g and 10 188 (± 1303) g, respectively.

Allergic symptoms (eczema) of atopic infants had begun at the mean age of 2.4 (range 0.2–6) months, and during

Table 1. Descriptive clinical characteristics [mean (SD)] of mothers of atopic infants (study group) and mothers of healthy infants (control group)

	Mothers	
	Study group ($n = 24$)	Control group ($n = 25$)
Age (years)	30.9 (3.9)	31.3 (4.8)
Weight (kg)	61.3 (11.3)	65.7 (12.2)
Height (cm)	165.4 (5.9)	166.6 (6.3)
BMI (kg/m^2)	22.3 (3.3)	23.7 (4.0)
Age at menarche (years)	13.2 (1.3)	13.0 (1.3)
Parity	1.0 (0.9)	1.0 (0.8)
Duration of lactation until DXA visit (months)	7.2 (3.0)	6.2 (3.1)
Duration of exclusive lactation (months)	4.1 (1.5)	2.8 (1.2)
Duration of amenorrhea (months)	7.5 (2.6)	6.0 (3.1)

Table 2. Serum cholesteryl ester fatty acid proportions [mean (SD)] in mothers of atopic infants (study group) and of healthy infants (control group) and corresponding z-scores ([mean (95% confidence interval)])

Fatty acid, mol %	Study group	Control group	Z-score
ω -3 fatty acids			
ALA	0.86 (0.23)	0.80 (0.20)	+ 0.3 [−0.2 to + 0.8]
EPA	1.01 (0.28)	0.89 (0.30)	+ 0.4 [−0.01 to + 0.8]
ω -6 fatty acids			
LA	52.72 (4.44)	54.78 (3.86)	−0.5 [−1.0 to −0.05]
GLA	0.76 (0.33)	0.57 (0.21)	+ 0.9 [+ 0.2 to + 1.5]
DGLA	0.57 (0.14)	0.52 (0.13)	+ 0.4 [−0.2 to + 0.9]
AA	4.93 (1.16)	5.25 (1.53)	−0.2 [−0.5 to + 0.1]

ALA (α -linolenic acid 18 : 3n3), EPA (eicosapentaenoic acid 20 : 5n3), LA (linoleic acid 18 : 2n6), GLA (γ -linolenic acid 18 : 3n6), DGLA (dihomo- γ -linolenic acid 20 : 3n6), and AA (arachidonic acid 20 : 4n6).

Table 3. Predictive models for the significant Z-scores of LA (linoleic acid 18 : 2n6) and GLA (γ -linolenic acid 18 : 3n6) proportions

Fatty acid	Adjusted R ²	Variable	Regression coefficient	Standard error	Contribution to R ²
LA	0.42	Fish	+ 1.412	0.451	0.26
		Total lactation	+ 0.170	0.061	0.20
GLA	0.34	Amenorrhea	−0.009	0.003	0.22
		Elimination diet	−1.862	0.721	0.20
		Weight	+ 0.054	0.024	0.15

Table 4. Crude BMD data [mean (SD)] in mothers of atopic infants (study group) and of healthy infants (control group) and corresponding z-scores (mean [95% confidence interval])

BMD (g/cm ²)	Study group	Control group	Z-score
Lumbar spine	0.945 (0.117)	1.003 (0.096)	−0.6 [−1.1 to −0.09]
Femoral neck	0.817 (0.102)	0.866 (0.114)	−0.4 [−0.8 to −0.1]
Distal radius	0.334 (0.039)	0.348 (0.053)	−0.3 [−0.6 to + 0.04]

Table 5. Predictive models for the significant Z-scores of lumbar spine and femoral neck BMD

Skeletal site	Adjusted R ²	Variable	Regression coefficient	Standard error	Contribution to R ²
Lumbar spine	0.25	Weight	+ 0.058	0.020	0.29
Femoral neck	0.49	Weight	+ 0.049	0.012	0.38
		EPA*	+ 1.048	0.491	0.11

* Eicosapentaenoic acid 20 : 5n3.

exclusive breastfeeding in 20 infants. The mean age of infants at paediatric consultation was 6.0 (2.1–13.9) months. Their mean SCORAD A (extent of eczema), B (intensity of eczema) and C (subjective manifestations) scores were 19.1 (95% CI 8.4–29.7), 3.0 (2.0–3.9) and 4.3 (2.5–6.1), respectively.

Dietary factors

The body weight in the study mothers who had been on elimination diets did not differ from those who had never been on a diet [62.7 kg ($\pm$ 11.5 kg) vs 64.1 kg ($\pm$ 12.2 kg), $P = 0.7$]. Only nine of the 19 mothers on dietary elimination took concurrent calcium supplementation. The calcium intake in the study group was significantly lower than in the control group ($P = 0.05$). The mean calcium intake of the control group [1220 (SD 490) mg/day] was well within the recommended dietary allowance (1000 mg calcium/day), in contrast to the substantially lower intake of the study group [880 (480) mg/day]. Consumption of fish was similar in both groups ($\chi^2 = 1.4$, $P = 0.2$). Thirty-five mothers ate fish less than once a week, while the remaining 14 mothers ate fish weekly, five of them being mothers of the atopic infants.

Fatty acids

The fatty acid profile of serum cholesteryl esters in the study and control groups, and corresponding z-scores, are given in Table 2. Mothers in the study group had a significantly lower proportion of ω -6 fatty acid parent LA and a higher proportion of ω -6 fatty acid metabolite GLA than the control mothers. The consumption of fish and duration of lactation accounted for 42% of the variability in the z-score of the LA level, whereas the duration of postpartum amenorrhea, trial of elimination diet and body weight explained 34% of the variability in the z-score of the GLA level (Table 3).

Bone mineral density

BMD data at the lumbar spine, femoral neck and distal radius in the study and control groups, and corresponding z-scores, are given in Table 4. There was a systematic trend for a 4–6% lower BMD in the study group compared with the control group, the differences being significant at the lumbar spine and femoral neck. The body weight chiefly explained this difference (Table 5), but it is noteworthy that the ω -3 fatty acid metabolite EPA showed a positive association with the femoral neck BMD. The regression models remained virtually the same when dietary calcium was included in the regression analysis ($N = 18$).

Discussion

Elimination of responsible antigens from breastfeeding mothers' diets may help to alleviate atopic symptoms in the infant [8]. However, the maternal elimination diet may not only impair the nutrition and growth of the infant [22], but it may also disturb the delicate nutritional balance of the mother and have consequences on the mother's health.

Prolonged amenorrhea and hypoestrogenemia associated with lactation, and increased maternal demand for calcium, concern the great majority of women and impose on their skeletons subsequent periods of apparent bone loss, the recovery being either partial [23,24] or complete [25–28] after weaning. Maternal physiology apparently meets the increased calcium demand via increased uptake of calcium from the skeleton, a process which is apparently modulated by inherent hormonal changes postpartum. In the present study of postpartum women, BMD was determined at three skeletal sites soon after the resumption of menses, at the time when the skeletal status was anticipated to reflect the combined effects of both the postpartum amenorrhea and the exclusive lactation and subsequent calcium loss into breast-milk during the first months postpartum. A systematic trend was observed towards lower BMD in mothers of atopic infants compared with mothers of healthy infants, but this difference was mainly attributable to differences in body weight. Despite the fact that the mothers of atopic infants breastfed their babies without supplementation for significantly longer (4.1 months vs 2.8 months on average) than the control mothers, the duration of exclusive lactation was not associated with the observed difference in BMD. In general, a long unsupplemented lactation is known to be associated with a higher bone loss than a short lactation [29].

As far as is known there have been no previous studies assessing the impact of a maternal elimination diet during lactation on the mother's skeletal health. The present results indicate that short-term elimination diets do not have a considerable adverse effect, if any, on BMD in postpartum lactating mothers. It should be noted that the empirical elimination diets of the mothers in this study were uncontrolled and unstructured, as typically applies in clinical practice. In general, empirical elimination diets are aimed at restricting the consumption of fish, egg, citrus fruits and cow's milk [8]. These products are excellent sources of many vital nutrients (i.e. calcium, vitamin D), and it is thus possible that these dietary restrictions may lead to an inadequate intake of several nutrients, including calcium [12]. It is commonly believed that loss of protein, fat and calcium due to an elimination diet should be compensated [30,31]. Recent studies, however, have not found a clear relationship between calcium intake or calcium supplementation and bone mineral changes in lactating women [32–37].

Under normal circumstances, the most important influence on bone mass evidently results from the mechanical loading which the bone experiences, i.e. the combined effects of muscle activity and weight-bearing [38]. The present results showing a clear association between body weight and BMD is in line with previous studies in postpartum women [33,39]. However, the possibility that a lower body weight would also reflect some indirect effects of elimination diets on the nutritional balance of the mother cannot be totally ruled out. This disturbance may have been manifested as a trend towards lower BMD in the study group. Particularly with regard to the mothers with a low body weight, it is possible that proportionately greater additional dietary energy [40] and/or nutrient requirements are needed during lactation compared with normal weight mothers. If this is so, and if the condition is not adequately met, some loss in body weight and BMD may occur.

Elimination diets modify the consumption of fats which is reflected in the serum fatty acid profile. Fatty acids affect not only the immune system [15,16], but ω -3 and ω -6 fatty acids in particular are also known to be important regulators of calcium metabolism [17]. It has previously been reported that an EPA-enriched diet can prevent bone loss caused by oestrogen deficiency or inadequate nutrition in rats [41]. The present finding showing the association of EPA level with femoral neck BMD gives some support to the relationship between fatty acids and bone mass. The potential interaction between fatty acids and calcium metabolism thus deserves further investigation. Lower LA levels and higher GLA levels were observed in mothers of atopic infants compared with mothers of healthy infants. Interestingly, despite lower linoleic acid levels in the study group, they had significantly higher levels of its metabolic and elongation products, namely γ -linolenic acid. This could be due to changes in fatty acid metabolism during lactation which may be different in mothers of atopic infants. Not only dietary factors but also incident hormonal factors postpartum may play a role in fatty acid metabolism.

In conclusion, a systematic 4–6% lower BMD in mothers of atopic infants was observed, but this difference was mainly attributable to a lower body weight in these mothers compared with mothers of healthy babies. Whether this lower body weight reflects some modulation of the elimination diets, or other related aspects, is not yet known. The association of the ω -3 fatty acid metabolite, EPA, level with the femoral neck BMD is worth noting. The ω -6 fatty acid metabolite LA and GLA levels, in turn, were associated not only with dietary factors but also with the duration of postpartum amenorrhea and total lactation. The main limitation in the study was the small sample size which apparently led to a limited statistical power to detect more potential and relevant predictors (e.g. exclusive lactation) for the intergroup difference, and the findings should

therefore be interpreted with caution. From the clinical point of view, a mother's short-term elimination diet *per se* seemed not to be associated with maternal BMD, but it may be advisable to give special dietary counselling, especially to low body weight mothers, in order to ensure adequate nutrient intake and minimize the risk of potential bone loss.

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