Opinion of the Panel on nutrition, dietetics products, novel food and allergy of the Norwegian Scientific Committee for Food Safety
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Assessment of infant formulae based on partially hydrolysed proteins

CONTRIBUTORS

Persons working for VKM, either as appointed members of the Committee or as *ad hoc*-experts, do this by virtue of their scientific expertise, not as representatives for his/her employers. The Civil Services Act instructions on legal competence apply for all work prepared by VKM.

Acknowledgements

The Norwegian Scientific Committee for Food Safety (Vitenskapskomiteen for mattrygghet, VKM) has appointed an *ad hoc* group consisting of both VKM members and external experts to answer the request from the Norwegian Food Safety Authority. The members of the *ad hoc* group are acknowledged for their valuable work on this opinion.

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Assessed by

The report from the *ad hoc* group has been evaluated and approved by the Panel on Nutrition, Dietetic Products, Novel Food and Allergy of VKM.

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SUMMARY

The regulation of infant formulas is under revision, including the national restriction on sales of infant formulas with partially hydrolysed proteins to pharmacies only. In December 2007 the Norwegian Food Safety Authority requested an opinion from the Norwegian Scientific Committee for Food Safety on infant formulas with partially hydrolysed protein. The opinion should cover an evaluation of risks associated with the replacement of conventional infant formulae (based on intact protein) by infant formula based on partially hydrolysed protein, including a specification of the risks associated with the consumption of infant formula based on partially hydrolysed proteins among infants with cow's milk allergy. Furthermore, the opinion should include an evaluation of the preventive effect from formula with partially hydrolysed proteins on development of cow's milk allergy.

To complete this task, VKM has established an *ad hoc* group that has prepared this report. The Panel on Nutrition, Dietetic Products, Novel Food and Allergy has discussed and adopted this opinion.

The main conclusions are that although no studies show that partially hydrolysed formulas have negative effect on growth, measures should be taken in order to avoid that partially hydrolysed formulas replace regular infant formulas in the general population. Hydrolysed proteins have a high absorption rate, and ingestion of hydrolysed proteins have been shown to increase gastric emptying and plasma glucose-dependent insulinotropic polypeptide relative to the intact protein forms.

Feeding partially hydrolysed formulas to infants with cow's milk allergy constitutes a risk of eliciting an allergic reaction which in worst case may be fatal. The risk that an infant with cow's milk allergy will receive partially hydrolysed formula may increase if the products are sold with claims such as reduced risk towards cow's milk allergy and reduced risk of developing cow's milk allergy together with ordinary infant formula outsides pharmacies.

The risk of developing cow's milk allergy is not shown to be reduced by introducing partially hydrolysed formula instead of regular formula during the first 4-6 months. There has not been demonstrated any preventive effect from hydrolysed formulas after the age of 6 months on the development of allergic disease.

SAMMENDRAG (IN NORWEGIAN)

Gjeldende regelverk for morsmelkerstatning er under revisjon – herunder begrensning av salg av morsmelkerstatning med delvis hydrolyserte proteiner til apotek. I desember 2007 sendte Mattilsynet en forespørsel til VKM der de ber om en risikovurdering av morsmelkerstatninger med delvis hydrolyserte proteiner. Risikovurderingen skal inkludere utredning av mulige risikoer forbundet med å erstatte vanlig morsmelkerstatning med et delvis hydrolysert produkt, herunder risiko knyttet til barn med kumelkallergi, samt gi en vurdering av om morsmelkerstatning basert på delvis hydrolysert protein vil gi redusert risiko for å utvikle kumelkallergi.

VKM har nedsatt en *ad hoc*-gruppe som har arbeidet frem denne vurderingen. Vurderingen er sluttbehandlet av Faggruppen for ernæring, dietetiske produkter, ny mat og allergi.

Hovedkonklusjonene i vurderingen er at til tross for at studier ikke viser negativ effekt på vekstutvikling hos spedbarn som har fått morsmelkerstatning med delvis hydrolyserte proteiner, bør det iverksettes tiltak for å unngå at vanlig morsmelkerstatning blir erstattet med delvis hydrolysert morsmelkerstatning i den generelle befolkningen. Hydrolyserte proteiner

absorberes raskere, og har vist å øke magesekkstømning samt nivå av glukoseavhengig plasma insulinotropisk polypeptid sammenlignet med intakte proteiner.

Hos kumelkallergikere vil morsmelkerstatning med delvis hydrolysert protein utløse allergiske reaksjoner som i verste fall kan være dødelige. Risikoen for at spedbarn med kumelkallergi skal eksponeres for morsmelkerstatning med delvis hydrolyserte proteiner kan øke dersom disse produktene omsettes med antydninger om allergiforebyggende egenskaper sammen med ordinære morsmelkerstatninger utenfor apotek.

Det er ikke dokumentert at risiko for utvikling av kumelkallergi er redusert ved introduksjon av delvis hydrolyserte proteiner i stedet for ordinær morsmelkerstatning opp til 4-6 måneders alder, og morsmelkerstatninger med delvis hydrolyserte proteiner utløser dessuten allergireaksjon hos kumelkallegikere. Det er forøvrig ikke vist noen allergiforebyggende effekt for noen typer kumelkhydrolysater etter 6 måneders alder.

BACKGROUND

Infant formulas intended for use when breastfeeding is not sufficient may be formulas with intact cow's milk protein, formulas with partially hydrolysed cow's milk protein and formulas with extensively hydrolysed cow's milk protein. Most infants can use formulas with intact cow's milk protein. Hydrolysed formulas are promoted as preventive against food allergy, specifically cow's milk allergy, and allergic diseases like atopic dermatitis and allergic asthma, but few studies document such preventive effect from these formulas. Some well conducted studies document an effect of certain extensively hydrolysed formulas in postponing or even preventing the appearance of allergic symptoms. The effect of partially hydrolysed formulas is more questionable, but there are a few studies demonstrating a minor effect of such formulas on atopic eczema during the first year of life.

According to EU/EEA legislation, infant formulas with partly hydrolysed proteins can bear claims such as *reduced risk towards cow's milk allergy and reduced risk of developing cow's milk allergy*. In partly hydrolysed infant formulas the content of intact protein is so high that the risk of allergic reactions is substantial. Thus such formulas must be labelled with a warning that children with documented cow's milk allergy shall not use these products. This warning is not mandatory for extensively hydrolysed formulas which in clinical tests show that less than 10% of the children with cow's milk allergy have adverse effects/allergic reactions from using the product in question. In the EU/EEA the labelling of infant formulas with partly hydrolysed proteins is regulated in Commission directive of 14 May 1991 on infant formulae and follow-on formulae (EC, 1991). Protein hydrolysates in infant formulas were evaluated by the Scientific Committee on Food in 2003 p 45 – 48 (SCF, 2003).

When this directive was adopted in EU, the Norwegian authorities were against the opportunity to label infant formulas with partly hydrolysed proteins as hypoallergenic etc., but the Norwegian view was not taken into consideration when the directive was adopted in EU. Particularly the risk of allergic reactions related to consumption of partly hydrolysed cow's milk proteins among children with cow's milk allergy (prevalence 1-3% in preschool children) is of concern. There is also a concern that normal population groups will prefer hypoallergenic products to conventional infant formulas. Regular infant formulas are designed to resemble human milk, and human milk does not contain hydrolysed proteins. On the contrary, it contains intact biologically active proteins and petides, several of these with shown immunological activity (e.g. IgA).

According to existing Norwegian legislation, the sale of infant formulas claiming to reduce the risk of developing allergy or to be hypoallergenic is limited to pharmacies (Norwegian Food Control Authority, 2001). The limitation of sales through pharmacies is unique for Norway. This is a market regulation measure attempting to avoid broad sales to normal population groups, and to ensure proper guidance on the use of these products from educated pharmacists to the consumer. The employees at a pharmacy have either a 5 year master degree in pharmacy, a 3 year bachelor degree in pharmacy or a degree as a pharmaceutical technician.

The Norwegian Food Safety Authority is implementing the revised directive on Infant Formulae and Follow-on Formula (Commission Directive 2006/141/EC), and has therefore requested an assessment of the scientific rationale for maintaining sales limitation of infant formulas with partly hydrolysed proteins to pharmacies.

To complete this task, VKM has established an *ad hoc*-group (members listed above). The *ad hoc*-group has had 4 meetings and prepared this report. The Panel on Nutrition, Dietetic Products, Novel Food and Allergy has discussed and adopted this opinion.

TERMS OF REFERENCE

The Norwegian Food Safety Authority has requested VKM to address the following questions:

1. What risks are associated with the replacement of conventional infant formulae (based on intact protein) by infant formula based on partially hydrolysed protein?
2. Specify the risks associated with the consumption of infant formulae based on partially hydrolysed proteins among infants with cow's milk allergy.
3. Will infant formulae based on partially hydrolysed protein reduce the risk of developing cow's milk allergy?

ASSESSMENT

Hazard identification

Infant formulas with partly hydrolysed proteins can be labelled with claims such as *reduced risk towards cow's milk allergy and reduced risk of developing cow's milk allergy*, but few studies document preventive effect from these formulas. There is a concern that normal population groups will prefer hypoallergenic products to regular infant formulas that are designed to resemble human milk.

Prevalence of cow`s milk allergy in preschool children

1. The reported frequency of cow's milk allergy (CMA)/ cow's milk proteinintolerance (CMPI) in the first years of life is between 1 and 7% (Rona *et al.*, 2007). Some breastfed infants (0.5%) have reproducible clinical reactions to human milk due to residual cow's milk protein in human milk (Host *et al.*, 1999).

   It is well documented that most children outgrow their adverse reactions to cow's milk before school age. Tolerance is achieved in 45 – 50% during the first year, 60 – 75% during the second year and 85 – 90% during the third year (Host, 2002).

2. It is difficult to predict the outcome of an allergic reaction in infants. The symptoms may start already during the first weeks of life and may be cutaneous (50-60%), gastrointestinal (50-60%), or respiratory (20-30%) often with symptoms from more than one organ system (Host, 2002).

   Adverse reactions to cow's milk may be either IgE mediated or non IgE mediated, but there are few published reports of the relationship between the two. Some investigators report the frequency of IgE mediated allergy from 26 to 73% of the total adverse reactions to milk (Majamaa *et al.*, 1999; Saarinen *et al.*, 2005).

   The atopy patch test together with skin prick test may be a tool for differentiating between IgE and non IgE allergic reactions while there are no diagnostic tools for non allergic adverse reactions except for food challenges. The level of specific IgE is shown to be important in determining clinical course and prognosis of IgE mediated cow's milk allergy (Saarinen *et al.*, 2005).

Infant formulas with hydrolysed proteins

The optimal peptide length or content of intact protein in infant formula which will induce tolerance is not known. The optimum extent of hydrolysis for induction of tolerance is not known as we do not know what amount of residual allergenicity is necessary to induce tolerance (von Berg *et al.*, 2007; Greer *et al.*, 2008).

It is feasible to differentiate between formula for prevention and formula for therapy.

According to the American Academy of Pediatrics (AAP) Committee on Nutrition, a formula may only be called hypoallergenic if three criteria are fulfilled (Baker *et al.*, 2000).

1. The antigenicity of the protein is reduced
2. The formula does not give symptoms in 90% (with 95% confidence) of cow's milk allergic patients verified by properly conducted elimination – challenge tests.
3. The immunogenicity of the product is reduced

Infant formulas claimed to be hypoallergenic are divided into extensively hydrolysed formula (eHF) and partially hydrolysed formula (pHF). An eHF is defined as a formula where at least 90% of the proteins are < 3 kD whereas a pHF has peptides in the range 3 – 10 kD (Greer *et al.*, 2008).

Only eH formulas fullfill the AAP criteria for hypoallergenicity, and only eHF may be classified as formula for therapy.

An infant formula for prevention must be documented to prevent or postpone symptoms of allergic disease or allergic sensitisation in infants.

Food/formula intended for prevention should have a very low, if any allergenic activity, and should be tested in a high risk population and document significantly lower prevalence of allergy in the population tested (Host *et al.*, 1999).

NAN HA1 is the only pHF on the Norwegian market (April 2008). This is a hydrolysed whey formula. The content of β-lactoglobulin has been analysed by using enzyme-linked immunosorbent assay (ELISA), and was found to be 12 400 ng/ml compared with 8.9 ng/ml in extensively hydrolysed whey formula (Profylac). Breast milk contains 0.9 – 150 ng/ml β-lactoglobulin (Halken *et al.*, 2000).

Except from the hydrolysed proteins, the nutrient content in NAN HA1 is similar to the nutrient content in regular infant formulas.

Development of GI-tract and immune system in infancy

There is substantial immaturity of the intestine at birth. The early neo-natal period is characterised by low IgA-production and increased permeability of the intestinal barrier, and it is generally accepted that early infancy (0-4 months) is a special vulnerable period in view of sensitisation with allergens. The gut has a number of important functions in addition to digestion and absorption of foods including a major immunological function and bacterial colonisation and fermentation.

The small intestine is a major immunological organ and the total number of lymphocytes within the intestine is equivalent to the number in the spleen. There is a rich representation of the innate immune system with cells like macrophages, eosinophils and mast cells.

Maturation of the intestinal immune system continues towards about 2 years of age.

General dietary recommendations for infants (Norway)

The main advice in the general Norwegian dietary recommendations for infants is to breastfeed exclusively the first 6 months of life. In cases where breastfeeding is not possible or sufficient, regular infant formula should be applied as replacement. Breastfeeding should preferably be continued throughout the first 12 months to allow for modulation of the immune response by breast milk when introducing new proteins. From 10 months some ordinary cow's milk may be used in food preparation and also some sour milk. From 12 months the infant may have the same food as the rest of the family, but with less salt (Statens råd for ernæring og fysisk aktivitet, 2002).

Hazard characterisation

Several reviews have examined the hypoallergenicity in different pHF and eHF (Osborn & Sinn, 2003; Hays & Wood, 2005; Zuppa *et al.*, 2005; Osborn & Sinn, 2006; Greer *et al.*, 2008). A few well controlled studies have examined the preventive effect from eHF and pHF in infants with high risk of atopy, and only one well controlled study has examined the preventive effect in an unselected population.

eHf and pHF and prevention of allergy in infants with hereditary risk.

The use of hydrolysed formula (both eHF and pHF) has been suggested as a preventive measure for infants at risk for allergic diseases, especially atopic dermatitis and food allergy.

Both a Cochrane meta-analysis from 2006 and a major review from 2008 conclude that the literature supports a limited although significant atopy-preventing effect of extensively hydrolysed cow's milk formulas compared to adapted cow's milk formulas as supplement or substitute to breast milk during the first 4 to 6 months of life in infants with high risk of atopy. This primarily owing to a preventive effect on the development of atopic dermatitis in the first year of life (Osborn & Sinn, 2006; Greer *et al.*, 2008; Host *et al.*, 2008). A possible preventive effect appears to be limited to immunological problems mainly seen among small children. The literature shows no prevention in respiratory related problems for neither eHF nor pHF.

In their review Greer *et al*. conclude that in studies of high risk infants who were not exclusively breastfed for 4-6 months there is modest evidence that atopic dermatitis may be delayed by the use of hydrolysed formula, and that additional studies are needed to document the long term ability of dietary interventions in infancy to delay or prevent atopic disease (Greer *et al.*, 2008). There are only a few well-controlled studies comparing the preventive effect of extensively hydrolysed with partially hydrolysed cow's milk formula on atopic disease. Greer *et al*. used the inclusion criteria of a 2006 Cochrane review and found only 14 randomised trials in term infants comparing the use of hydrolysed (eHF and pHF) with the use of human milk or an adapted cow's milk formula.

Three studies including altogether 251 infants examined the effect of pHF on reduction of the occurrence of any allergy compared with cow's milk formula in high risk infants (Vandenplas *et al.*, 1992; Willems *et al.*, 1993; de Seta *et al.*, 1994). Two of these found no significant effect; the third found an OR 0.45 (95% CI: 0.22 – 0.94) for pHF versus cow's milk formula (Vandenplas *et al.*, 1992)**.**

One recent study compares eHF and pHF in a high risk population of children followed from birth to 3 years of age (von Berg *et al.*, 2007). This is a prospective, randomised and double blind study in 2252 infants with atopic heredity. Comparison is made of 3 hydrolysates in the interventional arm of the study; partially hydrolyzed whey Beba HA, extensively hydrolyzed whey HIPP HA (Nutrilon Pepti) and extensively hydrolyzed casein formula (Nutramigen). The infants are followed up to 3 years. Outcome measures included are atopic disease and asthma. The authors report preventive effect of both eHf and pHF. The effect of eHf was more pronounced, but only for an eH casein formula, not eH whey formula. The study demonstrates a marginal effect of pHF on the development of atopic eczema, an effect which in an intention to treat analysis was not apparent (von Berg *et al.*, 2007).

In an earlier study however, Halken *et al*. did demonstrate an effect of another eH whey formula. This study shows a small reduction in the development of cow's milk allergy by feeding whey pHF for the first 4 months of life, an effect which was significantly less than the effect of exclusive breastfeeding or eHF (Halken *et al.*, 2000). This supports the view the protein source may be important for an effect on prevention of allergic disease. Oldaeus *et al*. found an effect of eHF on allergic sensitisation and allergic manifestations, but no effect of pHF (Oldaeus *et al.*, 1997).

Studies in infants without hereditary risk

There is no conclusive evidence for the preventive effect of use of hydrolysed formulas in infants without hereditary risk. It is a well recognised limitation that most studies have been conducted in infants with high risk of atopy and thus there is no common knowledge of the effect of preventive measures on the total infant population. Only one well controlled larger study (Exl *et al.*, 2000) has evaluated the possible atopy preventive effect of partially hydrolysed cow's milk formulas in 1130 unselected infants. The study demonstrated no negative effects of a pHF from whey during the first 6 months of life and a statistically positive effect on skin symptoms (when both atopic and non-atopic skin symptoms were included) from using the pHF.

Studies including NAN HA 1 as pHF

As *in vivo and in vitro* studies indicate that the degree of hydrolysis, the resulting molecular weight, and the protein source have little predictive value on the immunogenic or allergenic effect and thus the clinical outcome (von Berg *et al.*, 2007) each type of partially hydrolysed formula has to be independently studied in clinical trials.

Only three studies have so far included NAN HA1 as pHF (Tsai *et al.*, 1991; Vandenplas, 1992; Halken *et al.*, 2000), and only Vandenplas *et al*. and Halken *et al*. showed positive effect on atopy prevention. In the Vandenplas study 67 high risk infants were randomised to exclusive feeding for 6 months with NAN HA 1 or regular NAN and followed to 12 months. A preventive effect against sensitisation to cow's milk was reported. Nestlé provided the formulas and performed statistical analysis (Vandenplas, 1992). Halken *et al*. randomised 595 high risk infants at birth and the effect of eHF was compared to that of pHF (NAN HA 1). The clinical evaluation was blinded and with well defined diagnostic criteria. Total cumulative incidence of CMA at 18 months was 0.6% for eHF versus pHF 4.7%, but $p = 0.05$ (only borderline significance) (Halken *et al.*, 2000). Tsai *et al.* compared two groups of infants from birth; fed regular formula only, or breast milk and/or NAN HA 1, respectively. The two groups showed no significant difference in atopic disease, including moderate to severe eczema, at 6 months (Tsai *et al.*, 1991).

pHF and treatment of infants with known cow's milk allergy

No known specific protein structure or function is associated with allergenicity of milk. Due to the great variability and heterogeneity of the human immune-response, no single allergen or particular structure can account for a major part of milk allergenicity. Furthermore, the available evidence is not sufficient to establish an intake threshold below which allergic reactions are not triggered or to predict reliably the effect of food processing on allergenic potential of milk proteins (Wal, 2004).

According to the AAP criteria only products with highly reduced allergenicity based on extensively hydrolysed protein, or amino acid mixtures, are advisable for the treatment of infants with cow's milk allergy (Baker *et al.*, 2000). Partially hydrolysed formulas contain substantially higher amounts of residual allergens than extensively hydrolysed products and are not to be used by children with documented cow's milk allergy. Use of pHF in infants with cow's milk allergy will lead to allergic reactions. Children with cow's milk allergy may react in different ways and it is not always possible to predict the outcome of an allergic reaction. The most common patterns of reactions in small children are skin symptoms like urticaria, swelling and worsening of atopic dermatitis. Anaphylactic shock is an always dreaded possible outcome of an allergic reaction to cow's milk.

Some infants with cow's milk allergy (about 5 – 10%) may react even against the residual quantities of cow's milk protein in products with highly reduced allergenicity (eHF). Only amino acid-derived formulas are considered to be non-allergenic (Caffarelli *et al.*, 2002; Host & Halken, 2004).

Breast milk and introduction of cow's milk/pHF and other allergens

Small amounts of cow's milk protein may be tolerated while larger amounts may not, especially during the process of tolerance development. Tolerance induction is important during the first year of life. Breast milk with its several known anti-inflammatory properties may also favour the induction of tolerance versus allergic sensitisation in this vulnerable period (van Odijk *et al.*, 2003; Hanson, 2007). Furthermore, tolerance induction appears to be partly associated with the appearance of regulatory T cells (Karlsson *et al.*, 2004).

Simultaneous breastfeeding when introducing pHF, cow's milk and other allergens may function as a shield against sensitisation perhaps primarily owing to the multiple anti-inflammatory properties in breast milk (van Odijk *et al.*, 2003; Hanson, 2007). Thus, the foreign proteins are exposed to the intestinal immune system in an environment more in favour of tolerance induction than unwanted immunologic responses. According to this view, the experience from the Swedish celiac disease epidemic following postponement of the recommended age for introduction of gluten to the infants diet, suggests that the presence of simultaneous breastfeeding when introducing significant amounts of a food antigen is perhaps more important than the age of the infant (Ivarsson, 2005).

Hydrolysates and effects on metabolism/insulin response

Bovine milk contains two major protein fractions, the slowly absorbed caseins and the more rapidly absorbed whey proteins (Mahe *et al.*, 1996). Postprandial digestion rate is an independent factor modulating protein retention, and the slowly absorbed casein induces a positive protein balance relative to the more rapidly absorbed whey proteins (Boirie *et al.*, 1997; Dangin *et al.*, 2001). Furthermore, a high intestinal absorption rate increases the

deamination of ingested amino acids, leading to a higher urinary urea excretion (Lacroix *et al.*, 2006). It might be that a high absorption rate of the dietary protein will influence on protein retention and growth.

Hydrolysed proteins have a high absorption rate, and ingestion of hydrolysed casein or whey proteins has been shown to increase amino acid absorption rates, gastric emptying and glucose-dependent insulinotropic polypeptide relative to the intact protein forms (Calbet & Holst, 2004). Also, hydrolysed soy protein and hydrolysed whey protein have been reported to influence plasma insulin and glucagon levels, possibly by increasing the plasma glucagon to insulin ratio (Claessens *et al.*, 2008), which is important for the regulation of postprandial substrate oxidation (Claessens *et al.*, 2007). It is therefore likely that hydrolysed proteins will influence differently from the intact proteins on energy metabolism.

pHF and growth

Although the literature on cow's milk hydrolysates as nutrients for infants indicates that there may be no measurable difference in growth/ standard anthropometric data between infants fed hydrolysed versus regular cow's milk formula as a substitute or supplement to breast milk, the data is far from conclusive (Vandenplas *et al.*, 1993; Szajewska *et al.*, 2001; Maggio *et al.*, 2005; Osborn & Sinn, 2006; Beyer, 2007; von Berg *et al.*, 2007; Greer *et al.*, 2008).

Vandenplas *et al*. randomised 45 healthy children which were fed either whey intermediate hydrolysed formula (Nutrilon Pepti, Nutricia) or whey-predominant cow's milk formula (Nutrilon premium), and found no difference in nutritional status between the two groups at 3 months (Vandenplas *et al.*, 1993). Szajewska *et al*. randomised 45 preterms to eHF, pHF and standard preterm formula, with blinded intervention and evaluation of effect on growth and plasma amino acid profiles at 3 months, and found that eHF and pHF were nutritionally equivalent (Szajewska *et al.*, 2001).

No negative effect was reported on growth and development in infants fed hydrolysates (eHF or pHF) instead of breast milk or cow's milk formula during the first year of life in the von Berg study (von Berg *et al.*, 2007), a large prospective double blind interventional study including 2252 infants with a hereditary risk of atopy. None of the studies included in the review of Greer *et al*. or the Osborn & Sinn Cochrane meta-analysis report any adverse effects, including adverse effect on infant growth (Osborn & Sinn, 2006; Greer *et al.*, 2008).

However, in a well controlled randomised study of preterm infants (<1750g at birth), those fed a hydrolysed whey protein formula were reported to have slower weight gain and lower increment in head circumference, indicating reduced growth compared to those on standard preterm formula (Maggio *et al.*, 2005).

Exposure

In small children (<6 months), infant formula may represent their only food, and be a substantial part of their diet up to 1 year. Thus, special attention must be given to products intended for use in infants concerning risks and possible benefits.

According to data from nationally representative studies (Sped- og småbarnskost 1998-1999), 36% of the Norwegian infants receive an infant formula to drink at the age of 6 months. The mean daily intake is: 472 g/day in those who receive it daily, 170 g/day when including those who give infant formula less frequently (Lande, 2003).

Studies on increased focus on allergies in the population

Several studies demonstrate the increased awareness of disease in infants and children among young parents (Eggesbo *et al.*, 1999; Eggesbo *et al.*, 2001; Venter *et al.*, 2006). Venter *et al.* studied the incidence of parentally reported food hypersensitivity and objectively diagnosed food hypersensitivity during the first year of life in 969 infants. Adverse reactions to foods were reported by 9.1% parents at 6 months, 5.5% at 9 months, and 7.2% at 12 months. The infants underwent a medical examination and skin prick testing to allergens, and symptomatic children underwent food challenges. Between 6 and 9 months and 9 and 12 months, 1.4% and 2.8% of the infants were diagnosed with food hypersensitivity on the basis of open food challenges, and 0.9% and 2.5% on the basis of double-blind, placebo-controlled food challenges (Venter *et al.*, 2006).

In a population based Norwegian cohort 3623 children were followed from birth until the age of two. The aim of the study was to estimate the prevalence, incidence and cumulative incidences of parentally perceived adverse reactions to food in children younger than 2 years of age, and to study the duration of the reactions. The cumulative incidence of adverse reactions to food was 35% by age two. Milk was the single food item most incriminated, the cumulative incidence being 11.6% (Eggesbo *et al.*, 1999).

In an other study Eggesbo *et al*. estimated the prevalence of adverse reactions to milk. Children with parentally reported reactions to milk were selected for further examination from a population-based cohort of 2721 children. The prevalence of adverse reactions to milk at the age of 2(1/2) years was, based on objective procedures, estimated to be 1.1% (CI 0.8-1.6). However, there was also an underestimation, as unrecognised reactions were detected. Most reactions were not IgE mediated (Eggesbo *et al.*, 2001). This study confirms previous findings that parents overestimate milk as a cause of symptoms in their children.

Many parents avoid giving certain foods to their infants as they are afraid of adverse reactions (either food allergy or food intolerance). The foods that most commonly are avoided are cow's milk, nuts, orange/orange juice, fish and egg (Lande, 2003).

Marketing and availability of pHF

Effects from regulated sales and marketing of pHF on exposure are not discussed in any studies.

The labelling of formulas with *HA* (abbreviation for hypoallergenic) in conjunction with the brand name on pHF products may result in misconception because the term hypoallergenic refers to products suitable for patients with cow's milk allergy.

NAN HA1 is the only pHF product on the Norwegian marked (2008). The product information reads: "NAN HA1 is a hypoallergenic infant formula where a special treatment has reduced most allergenic substances found in cow's milk" (labelling text).

Use of pHF in infants with documented cow's milk allergy will lead to allergic reactions which in worst case may be fatal. This information is on the package but may be overlooked, especially when the product is sold outside pharmacies i.e. in supermarkets without personnel with relevant and qualified education.

Increased availability will most likely result in increased awareness about the product and increased consumption of pHF. The risk that an infant with cow's milk allergy will receive pHF may increase if the products are sold outsides pharmacies.

Risk characterisation

CMA/CMPI is frequent in the first years of life (1-3%) (Rona *et al.*, 2007). The symptoms may start already during the first weeks of life and may be cutaneous, gastrointestinal, or respiratory, often with symptoms from more than one organ system (Host, 2002).

The early neo-natal period is characterised by low IgA-production and increased permeability of the intestinal barrier. It is generally accepted that early infancy (0-4 months) is a special vulnerable period in view of sensitisation with allergens. Maturation of the intestinal immune system continues towards about 2 years of age.

Simultaneous breastfeeding when introducing pHF, cow's milk and other allergens may function as a shield against sensitisation perhaps primarily owing to the multiple anti-inflammatory properties in breast milk (van Odijk *et al.*, 2003; Hanson, 2007).

Atopy-preventing effects of eHF and pHF in early infancy

Infant formulas claimed to be hypoallergenic are divided in eHF and pHF. An eHF is defined as a formula where at least 90% of the proteins are < 3 kD whereas a pHF has peptides in the range 3 – 10 kD (Greer *et al.*, 2008).

It is important to differentiate between formula for prevention and formula for therapy.

There are only a few well controlled studies comparing the preventive effect of extensively hydrolysed with partially hydrolysed cow's milk formula on atopic disease (Vandenplas, 1992; Oldaeus *et al.*, 1997; Halken *et al.*, 2000; Exl *et al.*, 2000; von Berg *et al.*, 2007), and although the effect seems to be comparable, there is still need for more well controlled studies (Osborn & Sinn, 2006). Vandenplas *et al*. found that the incidence of cow's milk protein allergy appeared to be decreased by feeding whey hydrolysate formula for 6 months. Halken *et al*. found a small effect on development of cow's milk allergy by feeding pHF the first 4 months of life, an effect which was significantly less than the effect of exclusive breastfeeding or eHF (Halken *et al.*, 2000). Oldæus *et al*.found an effect of eHF on allergic sensitisation and allergic manifestations, but no effect of pHF (Oldaeus *et al.*, 1997). Although these studies generally are well controlled, one main weakness with such prospective interventional studies is, however, that a randomisation to breast feeding/extent of breastfeeding is of course not obtainable because of ethical reasons.

In the von Berg study, the authors report preventive effect of both eHF and pHF. The effect of eHF was more pronounced, but only for an eH casein formula, not eH whey formula. The study demonstrates a marginal effect of pHF on the development of atopic eczema, an effect which in an intention to treat analysis was not apparent (von Berg *et al.*, 2007). The results from the von Berg study indicate a real disease reduction, not only postponement of symptoms. Because of strict diagnostic criteria in this study (DBPCC), CMA is probably under diagnosed.

Almost all of the referred studies are in infants with high risk of atopy, and only one well controlled larger study (Exl *et al.*, 2000) has reported a possible atopy preventive effect of partially hydrolysed cow's milk formulas in unselected infants.

There are no firm criteria available for the design of hypoallergenic foods for prevention (Host & Halken, 2004). According to the AAP criteria, prevention studies should be

randomised from birth and the infants should be fed the formula (when supplements are needed) for at least the first 4-6 months of life. A formula for prevention should be tested in a high risk population and document significantly lower prevalence of allergy in the tested population. Follow up should be at least 18 months (Baker *et al.*, 2000). Validated clinical criteria including controlled challenges should be used for diagnosis. Very few studies (only the von Berg study from 2007) meet these criteria. Because of great variations in study design and diagnostic criteria, the relative efficacy of the different interventions tested in the various studies cannot be compared directly with each other.

As *in vivo* and *in vitro* studies indicate that the degree of hydrolysis, the resulting molecular weight, and the protein source have little predictive value on the immunogenic or allergenic effect and thus the clinical outcome (von Berg *et al.*, 2007), each type of partially hydrolysed formula has to be independently studied in clinical trials.

Furthermore, unfortunately the Exl study and some of the other studies referred to were performed in a company setting by Nestlé employees. The same comment must be made as to a meta-analysis performed by Tiffani Hays, a nutrition consultant for Nestlé USA, a meta-analysis which concludes with a general recommendation to use hydrolysates as a preventive measure when breast milk is not sufficient (Hays & Wood, 2005).

The conclusion can be made that firm evidence of a preventive effect of hydrolysed formula (both eHF and pHF) on the development of allergic disease is lacking although a modest effect on atopic dermatitis has been shown by some authors (von Berg *et al.*, 2007; Greer *et al.*, 2008), and a more pronounced effect of eHF than pHF (von Berg *et al.*, 2007). Almost all of these studies are in high risk infants. Only very few studies have been carried out in an unselected infant population and thus there is little or no knowledge of prevention in the total population.

There is no evidence for a preventive effect of feeding hydrolysed formulas beyond 4 months of age (Osborn & Sinn, 2006; Beyer, 2007; Greer *et al.*, 2008).

Adverse effects from pHF and eHF

No adverse effect on growth by feeding hydrolysed formula compared to intact cow's milk has been demonstrated, but most studies have not focused on safety. Special attention must be given in terms of safety or possible adverse effects to products intended for use in infants 0-4 months when the immune and gastro-intestinal systems are especially vulnerable.

Only products with highly reduced allergenicity based on extensively hydrolysed protein, or amino acid mixtures, are advisable for the treatment of infants with cow's milk allergy (Baker *et al.*, 2000). Partially hydrolysed formulas contain substantially higher amounts of residual allergens than extensively hydrolysed products and are not to be used by children with documented cow's milk allergy. Use of pHF in infants with cow's milk allergy will lead to allergic reactions. Children with cow's milk allergy may react in different ways and it is not always possible to predict the outcome of an allergic reaction. The most common patterns of reactions in small children are skin symptoms like urticaria, swelling and worsening of atopic dermatitis. Anaphylactic shock is an always dreaded possible outcome of an allergic reaction to cow's milk.

Some infants with cow's milk allergy (about 5 – 10%) may react even against the residual quantities of cow's milk protein in products with highly reduced allergenicity (eHF).

Lack of documentation both on effect and safety of pHF in general, including NAN HA 1

As the evidence for even a minor atopy-preventing effect of NAN HA 1 in unselected infants is far from convincing, the potential hazards of a widespread use of this and similar products in the general infant population demands serious attention. A general shift towards less allergenic and perhaps also less tolerogenic peptides in the diet during the first months of life in the unselected infant population may have unwanted effects.

Little information is available with regard to the importance of molecular weight distribution in the hydrolysates (i.e. extent of hydrolysis) on metabolic parameters. For example, if extensively hydrolysed proteins actually increase the plasma glucagon level, one could expect increased hepatic gluconeogenesis, which would not be beneficial for the diabetic infants. Also, little is known about dietary effects of hydrolysates from different raw materials, such as meat, plant protein or fish. There is a need for further studies to clarify how various protein hydrolysates influence growth and metabolic pathways in infants as well as in humans in general.

Adverse consequences of more widespread use of pHF in the general infant population

The rate of parental perception of food hypersensitivity is higher than the prevalence of atopic sensitisation to main food allergens or objectively assessed food hypersensitivity (Eggesbo *et al.*, 1999; Eggesbo *et al.*, 2001; Venter *et al.*, 2006). It is generally accepted that illness awareness is increasing and that children are victimised when special diets are instituted. If infant products are marketed as allergy preventive (as pHF) and are generally available, parents might think these products will spare their child of symptoms and diseases. The available literature on preventive effect of pHF is scarce and where such an effect is demonstrated, it is marginal (von Berg *et al.*, 2007; Greer *et al.*, 2008). This information is not given to the public, and will be even less available if the products were sold outsides pharmacies, e.g. in supermarkets.

Furthermore, one possible adverse effect of pHF is the possibility of the product being given to infants with cow's milk allergy. The risk that an infant with cow's milk allergy will receive pHF may increase if the products are sold outsides pharmacies.

There is a lack of data on the nutritional quality and physiologic effects of hydrolysed versus regular cow's milk formula. Further investigation regarding bioavailability of minerals and trace elements, protein and mineral metabolism, as well as long term effects, including neurodevelopment, are needed (Zuppa *et al.*, 2005) to make qualified recommendations for the use of partially hydrolysed cow's milk formula in preference to standard cow's milk formula in an unselected infant population (prematures not included), especially as there are no proven positive health effects, e.g. such as atopy prevention from pHF.

Allergies and allergic diseases are highly prevalent and much focus is on possibilities to prevent these conditions. When infant products are marketed as preventive (as pHF) and generally available, parents might think it is necessary to use these products in order to spare their infant of symptoms and diseases. It is generally accepted that illness awareness is increasing and that children are victimised when special diets are instituted.

Increased availability will most likely result in increased awareness about the product and increased consumption of pHF.

CONCLUSIONS

1. No adverse effect on growth and development by feeding hydrolysed formula compared to intact cow's milk has been demonstrated in term infants, but most studies have not focused on safety (Greer *et al.*, 2008). In small children (<6 months) infant formula may represent their only food, and be a substantial part of their diet up to 1 year. Special attention must be given to risks and possible benefits of products intended for use in early infancy (0-4 months) when the immune and gastro-intestinal systems are especially vulnerable. Measures should be taken in order to avoid that pHF replace regular infant formulas (when breastfeeding is insufficient) in the general population.

   Hydrolysed proteins have a high absorption rate, and ingestion of hydrolysed casein or whey proteins have been shown to increase amino acid absorption rates, gastric emptying and plasma glucose-dependent insulinotropic polypeptide relative to the intact protein forms (Calbet & Holst, 2004). Also, hydrolysed soy protein and hydrolysed whey protein have been reported to influence in plasma insulin and glucagon levels, possibly increasing the plasma glucagon to insulin ratio (Claessens *et al.*, 2008), which is important for the regulation of postprandial substrate oxidation (Claessens *et al.*, 2007). It is therefore likely that hydrolysed proteins will influence differently from the intact proteins on energy metabolism.

2. Feeding pHF to infants/children with cow's milk allergy constitutes a risk of eliciting an allergic reaction which in worst case may be fatal. This information is given on pHF products, but may easily be overlooked if the products are generally available and not confined to pharmacies.

   Parental perceived adverse food reactions are much higher than documented food reactions. Increased availability will most likely result in increased awareness about the product and increased consumption of pHF. The risk that an infant with cow's milk allergy will receive pHF may increase if the products are sold outsides pharmacies.

3. There is limited evidence that formulae based on partially hydrolysed protein reduce the risk of developing cow's milk allergy. Some studies found a marginal preventive effect, especially on atopic eczema. Almost all of these studies are in high risk infants. The risk of developing cow's milk allergy does not seem to be substantially reduced by introducing pHF during the first 4-6 months. There has not been demonstrated any preventive effect of hydrolysed formula used after the age of 6 months on the development of allergic disease.

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