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Short Report

# Effect of hydrolysed formula feeding on taste preferences at 10 years. Data from the German Infant Nutritional Intervention Program Plus Study☆

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SUMMARY

*Background & aims:* There is some evidence that the type of infant formula received in early infancy has an influence on later food preferences. How long potential effects of taste programming persist is however not clear. Therefore, the aim of present study was to investigate whether feeding with different kinds of infant formula is associated with the acceptance of infant formula at 10 years of age.
*Methods:* Preference testing was conducted in 833 10-year old children being part of the interventional subgroup of the German Infant Nutritional Intervention Program Plus Study (GINIplus). If they were not exclusively breastfed, these children have been fed with either one of three hydrolysed formulas or a cow's milk formula during the first four months of life.
*Results:* Feeding with any kind of hydrolysed formula in infancy was positively associated with a higher acceptance of extensively hydrolysed casein formula (aOR: 1.88; 95% CI 1.08, 3.29) after adjusting for sex and study centre.
*Conclusions:* Although this study did not show consistent associations and thus leaves some questions unanswered, it might stimulate further research on the potential programming effect of early infant feeding.


## 1. Introduction

Food acceptance patterns are individually different as soon as the child experiences a particular food. There is some evidence that the type of infant formula received in early infancy has an influence on food preferences in later childhood.

A previous study by *Mennella et al.* demonstrated that regular exposure to hydrolysed infant formula during the first 7 months of life resulted in a greater acceptance of this formula at 7.5 months of age than if it was first offered to infants who were never fed hydrolysed formula before.[1] The authors also reported that children who had experienced the sour and bitter tasting hydrolysed formula early in life preferred higher levels of citric acid in juice at 4–5 years of age when compared to children who were fed whole cow's milk protein-formulas.[2] Children who had been fed soy formula, which is bitter and astringent, were more likely to prefer bitter-flavoured apple juice.[3] However, by 7–8 years of age this effect had disappeared. How long potential effects of taste programming persist is thus not yet clear. Therefore, we investigated within the framework of the EU research project EARNEST (Early Nutrition Programming Project, www.metabolic-programming.org), whether feeding with different kinds of infant formula is associated with the acceptance of infant formula at 10 years of age. In particular, we hypothesise that children who were fed with extensively hydrolysed casein formula in infancy reject this kind of formula at 10 years of age less strongly than children who were fed with cow's milk formula.

## 2. Methods

Preference testing was conducted in 10-year old children being part of the interventional subgroup of the German Infant

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Nutritional Intervention Program Plus Study (GINIplus). This is a prospective, randomised, double-blind intervention trial initiated to assess the allergy-preventive effect of three different hydrolysed formulas compared with cow's milk formula in infants at high risk of atopy. Details of design, recruitment and exclusion criteria have been described.[4,5] In brief, between September 1995 and June 1998, a total of 2252 newborns with at least one family member (parents or biological sibling) with an allergic disease were recruited in obstetric clinics in the two German study areas Munich and Wesel. Infants were randomly assigned to one of three hydrolysed infant formulas or a regular cow's milk formula by means of a computer generated list:

(a) a partially hydrolysed whey formula (pHF-W; Beba HA);
(b) an extensively hydrolysed whey formula (eHF-W; Hipp HA [at that time identical with Nutrilon pepti];
(c) an extensively hydrolysed casein formula (eHF-C; Nutramigen);
(d) a standard cow's milk formula (CMF; Nutrilon Premium).

All mothers received detailed recommendations for feeding their infants. They were encouraged to exclusively breastfeed for at least four months (strict intervention period) and not to feed solid foods during this period. Only if breastfeeding was not possible, insufficient or refused, the randomised study formula was advised to be fed.

Of the 2252 newborns recruited at baseline, 1475 participated in the follow-up at 10 years (65.5 percent) and of those 833 children agreed to participate in the taste preference test (57.5%). To measure preferences of infant formulas, we used a continuous line-scale ranging from "extremely bad" to "extremely good". Subjects were instructed to taste the blinded samples in a given order and to mark the line according to how much they like or dislike the taste of each sample. A randomised complete block design with five treatment levels (five samples) was calculated in advance to warrant random assignment of samples to each child. The companies provided their adequate infant formulas with an almost identical composition to the formulas used during the intervention period 10 years before. The following infant formulas were tested:

(a) cow's milk formula (Nutrilon Standaard 1®, Nutricia, The Netherlands) (CMF_2),
(b) extensively hydrolysed casein formula (Nutramigen®, Mead Johnson, Germany (eHF-C_2),
(c) extensively hydrolysed whey formula (Nutrilon pepti 1, Nutricia, Netherlands) (eHF-W_2);
(d) partially hydrolysed whey formula (Beba H.A. 1, Nestlé, Germany (pHF-W_2) and
(e) conventional ultra heat treated milk (UHT) as control.

This study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving human subjects were approved by the Bavarian General Medical Council and the Medical Council of North-Rhine-Westphalia. Written informed consent was obtained from all participating families.

For statistical analysis, the distance between the left end of the scale and the mark on the line was measured and thus, liking scores (0–10.5) were generated which were then used as continuous outcome variable. Differences in liking scores between groups with different randomised study formulas during the intervention period were assessed by Kruskal–Wallis test.

The association between study formula feeding and acceptance of extensively hydrolysed casein infant formula at age 10 years was analysed by multiple logistic regression models. Due to the extremely skewed distribution, a dichotomous variable was constructed defining a 'positive' liking score as a value of above 0.1 (in addition 0.0 and 0.2). Sex and study area were included as basic covariates. All computations were performed by using the statistical analysis package SAS for Windows version 9.1 (SAS Institute, Cary, NC, USA). Two-sided $p$-values $<0.05$ were considered statistically significant for all analyses.

## 3. Results

Of 833 10-year old children who participated in the taste preference test, 565 (67.8%) were tested in Munich and 268 (32.2%) in Wesel. These children were equally distributed over the four randomised groups of infant formula, 26.1% to pHF-W, 25.1% to eHF-W, 25.3% to eHF-C and 23.5% to CMF. No significant differences in breastfeeding characteristics (exclusively/partially/not breastfed, breastfeeding duration) and timing of first exposure to the study formula were observed.

The median (Q1; Q3) liking score given by all subjects ($n = 833$) was 3.2 (0.2; 5.4) for CMF_2, 0.0 (0.0; 0.2) for eHF-C_2, 1.2 (0.0; 4.3) for eHF-W_2, 0.5 (0.0; 3.4) for pHF-W_2 and 9.4 (6.2; 10.4) for the control UHT.

Only subjects who were ever exposed to the randomised study formula ($n = 410$; 49.2%) were relevant for our study question why exclusively breastfed infants were excluded for all analyses (Intention to treat 2 analysis (ITT-2)).

Systemic differences in median liking scores between the randomised study formula groups were not observed for any of the test formulas (Table 1). There were further no major differences in liking scores according to sex, study centre nor were they dependent on the duration or the beginning of formula feeding during the intervention period (data not shown).

We further analysed whether a 'positive' liking score (>0.1) for extensively hydrolysed casein formula (eHF-C_2) is dependent on the formula children were fed with during early infancy (data not shown). We observed a positive effect for former feeding with pHF-W compared to feeding with CMF and acceptance of extensively hydrolysed casein formula (eHF-C_2) at age 10 years (aOR 2.20; 95% CI 1.15, 4.19) after adjusting for sex and study centre. No statistically significant associations were seen for former feeding with eHF-C (aOR 1.77; 95% CI 0.91, 3.47) or eHF-W (aOR 1.68; 95% CI 0.86, 3.27). After further adjustment for the amount of bottles which were fed during the intervention period, the effect estimates did not change substantially (data not shown). When the cut-off was shifted either to 0.0 or to 0.2, the effect estimates decreased, indeed showing the same direction but without statistical significance.

When combining all hydrolysed formula groups, the effect of exposure to any hydrolysed infant formula during the first 4 months of life was statistically significantly associated with a positive liking score for the bitter tasting casein formula (eHF-C_2) at 10 years of age (aOR: 1.88; 95% CI 1.08, 3.29).

## 4. Discussion

The findings of this study showed a positive association between feeding with any kind of hydrolysed formula in infancy and the acceptance of extensively hydrolysed casein formula at age 10 years. This effect was in particular due to higher liking scores given by children fed with partially hydrolysed whey formula.

Since the results were not robust against shifting the cut-off and there is no plausible explanation for a relationship with extensively hydrolysed casein formula only, these findings should not be over-interpreted. One may think of a familiarisation with any kind of hydrolysed formula in infancy that affects the acceptance of extensively hydrolysed casein formula at a later age, which has probably the most distinctive taste. The reason for this single result might be due to a lack of statistical power. Since most children

**Table 1**
Distribution of liking scores of four infant formulas and one control according to the randomised study formula during the intervention period (ITT-2 analysis; $n = 410$).

| | *n* | CMF_2[a] | eHF-C_2[b] | eHF-W_2[c] | pHF-W_2[d] | UHT[e] |
|---|---|---|---|---|---|---|
| | | Median (Q1; Q3) | Median (Q1; Q3) | Median (Q1; Q3) | Median (Q1; Q3) | Median (Q1; Q3) |
| CMF[f] | 107 | 3.2 (0.2; 5.6) | 0.0 (0.0; 0.1) | 0.7 (0.0; 4.2) | 0.3 (0.0; 3.5) | 9.9 (7.8; 10.4) |
| eHF-C[g] | 97 | 1.8 (0.1; 5.3) | 0.0 (0.0; 0.2) | 0.6 (0.0; 4.4) | 0.3 (0.0; 2.3) | 9.3 (6.6; 10.4) |
| eHF-W[h] | 101 | 3.2 (0.2; 5.5) | 0.0 (0.0; 0.2) | 1.3 (0.0; 4.7) | 0.3 (0.0; 3.8) | 9.9 (6.0; 10.4) |
| pHF-W[i] | 105 | 2.7(0.1; 5.3) | 0.0 (0.0; 0.2) | 0.7 (0.0; 3.2) | 0.4 (0.0; 3.0) | 9.4 (5.9; 10.3) |
| *p*-Value[j] | | 0.485 | 0.335 | 0.926 | 0.70 | 0.582 |

[a] CMF_2: Cow's milk formula (test formula)
[b] eHF-C_2: Extensively hydrolysed casein formula (test formula)
[c] eHF-W_2: Extensively hydrolysed whey formula (test formula)
[d] pHF-W_2: Partially hydrolysed whey formula (test formula)
[e] UHT: ultra heat treated milk (control)
[f] CMF: Cow's milk formula (randomised study formula)
[g] eHF-C: Extensively hydrolysed casein formula (randomised study formula)
[h] eHF-W: Extensively hydrolysed whey formula (randomised study formula)
[i] pHF-W: Partially hydrolysed whey formula (randomised study formula)
[j] Kruskal–Wallis-test

evaluated hydrolysed formulas as extremely bad, the data had to be dichotomised, which was not planned a priori, and we cannot assure that a positive evaluation defined as a value of above 0.1 actually reflects a better acceptance than children ranking the formula with a value of zero. Therefore, we cannot exclude that the reported effect is purely coincidental. In addition, this negative evaluation implies that extensively hydrolysed infant formula cannot be reliably used as test food in older children. Furthermore, no information on taste exposure and dietary habits during the first 10 years of life could be considered so that we cannot exclude residual confounding by any dietary influence during this period.

However, the prospective study design, the respectable number of participants in the trial and the approach of using infant formula in the preference test were basically appropriate for studying the effect of early infant feeding on later taste preferences.

We are aware of the fact that this study has some major limitations but think that it might be worthwhile to conduct further studies on the potential programming effect of early infant feeding also to verify whether there is any influence on food preferences and diet variety later in life.

## Conflict of interest

Berthold Koletzko is the recipient of a Freedom to Discover Award of the Bristol Myers Squibb Foundation, New York, NY, USA. None of the authors had any conflict of interest.

## Author agreement

All authors have made substantial contributions and approved the conception, drafting, and final version of the manuscript.

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SS was responsible for data analysis, interpretation of data and manuscript preparation. SK, BK and JH assisted in the interpretation of results and critical revision of the manuscript. DR, UK, AB, DB, CPB, AG, HEW and JH were responsible for data collection. SS, BK and JH developed the design and analysis plan of this study. None of the authors had any conflict of interest.

## Appendix. GINIplus – Study Group

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