

TITLE: Unhealthy dietary patterns established in infancy track to mid-childhood: the EU Childhood Obesity Project.

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- (ii) abbreviations: CORE: Core-foods Dietary Pattern, F&S: Poor Quality Fats and Added Sugars, PROT: High Protein Sources Dietary Pattern; KMO: Kaiser-Meyer-Olkin (test for sample adequacy)
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1 **ABSTRACT**

2 Background: Dietary habits established in infancy could determine long-term health. The aims of this
3 work are to describe dietary patterns, the predictors of adherence to specific dietary patterns and their
4 tracking from 1 to 8y in a cohort of European children.

5 Methods: 3-day food diaries were prospectively collected at ages 1, 2, 3, 4, 5, 6 and 8y. Foods were
6 allocated to one of 29 food groups, which were included in exploratory factor analyses at each
7 children's age. Tracking of patterns through childhood was assessed by an estimated general equation
8 model. Results: At 1y (n=633), two patterns were identified: one labeled as "Core Foods" (CORE), since
9 it positively loaded for vegetables, fish, olive oil, white and red meat, and negatively loaded for ready-
10 to-eat infant products, sugar and confectionary. The second was positively loaded for saturated
11 spreads (e.g. butter), sugar, fruit juices and confectionary, and negatively loaded for olive oil, fish and
12 cow's milk; this was named "Poor Quality Fats and Added Sugar" pattern (F&S). From 2 to 8y, three
13 patterns were repeatedly identified: CORE, F&S, plus a "High Protein Sources" (PROT) pattern that was
14 positively loaded for milk, flavored milks, fish, eggs, white and processed meat, chips and olive oil, and
15 negatively loaded fresh fruits at almost all timepoints.

16 Of those children in the highest quartiles of the CORE, F&S and PROT patterns at 2y, 45%, 72% and
17 36% remained in the highest quartile at 8y, respectively ((OR=2.01 (1.08, 3.8), OR= 3.6 (1.5, 8.4) and
18 OR= 0.80 (0.4,1.6) p=0.510)

19 Conclusions: Dietary patterns are established between one and two years, and track into mid-
20 childhood. A dietary pattern characterized by added sugar, unhealthy fats and poor consumption of
21 fish and olive oil was the most stable throughout childhood. Further analyses will reveal whether those
22 dietary patterns are associated with metabolic disease risk.

23

24 Key words: tracking dietary patterns, exploratory factor analyses, Childhood Obesity Project, infants,
25 toddlers, children, development of dietary habits

26

27 **INTRODUCTION**

28 Non-communicable diseases such as cardiovascular disease, diabetes and cancer are main drivers of
29 morbidity and mortality in many countries and the risk of being impacted by these illnesses can in part,
30 be prevented by having a more healthy diet (1–3).

31

32 Dietary patterns are likely established in the family environment during infancy and childhood (4). A
33 critical window for dietary pattern development is the period just after complementary feeding
34 commences, when the toddler begins to eat family foods. It is of interest to detect when dietary
35 patterns are established as well as whether those dietary patterns track into to older ages, as they may
36 influence later health. This would open a window of opportunity for educational programs at critical
37 ages that could impact health later in life.

38

39 Commonly, single nutrients and foods are analyzed in relation to health and disease. However, foods
40 and nutrients are consumed in combination, and the empirical analyses of dietary patterns support a
41 more comprehensive understanding of dietary intake as a whole. It is a powerful approach to
42 understand diet and its association to health, and to inform food-based recommendations that can be
43 translated to the general population (5). While several studies have described empirically derived
44 dietary patterns in young children (6–8), whether early dietary patterns (in the first 2 years of life) track
45 or predict dietary patterns in later childhood has not been extensively investigated.

46

47 The present work aims to describe major dietary patterns prospectively collected at ages 1, 2, 3, 4, 5
48 and 8y, and their tracking through that period in a cohort of young children from 5 European countries
49 (Germany, Belgium, Italy, Poland and Spain). This work aims as well determining what are the
50 predictors of adherence to specific dietary patterns at ages 2 and 8 years.

51 **METHODS**

52 ***Study design and population***

53 This is a longitudinal analysis of dietary data prospectively collected in children taking part from 1 to 8
54 years in the Childhood Obesity Project (EU CHOP), conducted in Germany, Belgium, Italy, Poland and
55 Spain. More details of the project and results have been previously published (10,11). Briefly, the EU
56 CHOP is the follow-up of an originally double blind randomized clinical trial in which infants were
57 recruited (October 2002 - July 2004) from birth to maximum their first 8 weeks of life (median age 14
58 days (IQR=3-30days)) in neonatal care units of study centers. Inclusion criteria were being born
59 healthy, term, from a singleton pregnancy, with a normal weight for gestational age, and to a mother
60 without health problems nor medication that could influence intrauterine growth. The aim of EU CHOP
61 was to determine whether infant formula with lower protein content during the first year of life
62 reduces later obesity risk. Children were randomly assigned to receive either an infant or a follow-on
63 formula during the first year of life with higher or lower protein contents (maximum and minimum
64 ranges of the recommended protein content at the time that the intervention was conducted). A
65 group of breastfed infants was also recruited for comparison, as a reference of healthy growth and
66 weight gain.

67

68 ***Outcome measures***

69 *Dietary Intake Assessment*

70 Three-day estimated and weighed food diaries were completed by the child's parent or caregiver at
71 the age of 1, 2, 3, 4, 5, 6 and 8y. The diaries were reviewed by trained dieticians during the study visits
72 and parents were asked to provide further details when required. Details about standard operating
73 procedures to assess dietary intake within the project have been published elsewhere (9,12).

74 To support dietary pattern analyses, 7444 individual foods and beverages reported in the food diaries
75 were allocated to 105 groups and then further collapsed to 29 major food groups based on their
76 nutrient profile and industrial processing. The 29 major food groups (e.g. white meat) and single foods

77 included in it (i.e. poultry) are reported as Online Supporting Material (**Supplemental Table 1**). Foods
78 consumed at very low levels were allocated to major food groups to avoid skewed distributions, e.g.
79 whole grain cereals were included in a major group of grains; pulses were included in the vegetables
80 group; and fatty fish was merged with lean fish in a major group. Foods that were rarely consumed
81 and had little to no variance were not included in the dietary pattern analyses (i.e. nuts, processed fish
82 and soft drinks (13). Meats were classified according to fat content: i.e. bovine, pork and lamb were
83 included in the “red meat” food item, while poultry and horse were classified in the “white and low fat
84 meat”. Further details about food items included in each of the 29 food groups could be found as
85 Online Supporting Material (**Supplemental Table 1**).

86

87 *Covariates*

88 Country (Germany, Belgium, Italy, Poland and Spain), gender (male, female), mother’s and father’s
89 education level (Low, Medium, High), mother’s and/or father’s having a different country of origin than
90 the study centre (foreigner, not foreigner), birth order (1, 2, 3, 4, etc.), being single mother (single, not
91 single), mother’s and father’s BMI (kg/m²), being breastfed vs. formula fed, formula intervention (high
92 protein vs. low protein) and maternal age at child’s birth (years) were collected with interviews at
93 baseline. Those covariates were considered as possible predictors of dietary patterns.

94

95 **Statistics**

96 *Dietary Pattern Analysis*

97 Exploratory factor analysis was conducted to extract dietary patterns at 1, 2, 3, 4, 5, 6 and 8y of age.
98 Briefly, this is a statistical method that reduces a large set of predictor variables (intakes of 29 major
99 food groups, g/day), according to correlations between them, into a smaller number of factors (dietary
100 patterns). A factor is the result of successive linear combinations of the predictors. This statistical
101 method is exploratory and extracts dietary patterns that explain as much variation in predictors as

102 possible, without considering any previous knowledge or hypothesis (5). In the present study,
103 exploratory factor analysis was conducted with a varimax rotation resulting in independent dietary
104 patterns. The factors (dietary patterns) extracted were those that accomplished the following criteria:
105 had a Kaiser Meyer Olkin (KMO) test for sample adequacy ≥ 0.6 , had an eigenvalue >1 , and the
106 explained predictors' variation were not below the elbow in the scree plot (19).

107

108 Through the exploratory factor analyses, we provide the amount of variation in predictors explained
109 by each pattern and the factor loadings for each food group for each identified pattern. Factor loadings
110 indicate whether intake of the food group is positively or negatively associated with each dietary
111 pattern and the magnitude of association. Those food groups with a factor loading greater than an
112 absolute value of 0.20 were considered the most important food items associated with each pattern.
113 For each dietary pattern, at each timepoint, each study participant received a standardized z-score
114 (mean=0; SD=1) indicating how closely their intake resembled the dietary pattern, relative to the rest
115 of the cohort. The dietary pattern z-score was calculated using a linear combination of the weighted,
116 standardized food intakes (using scoring weights for each food group estimated by the exploratory
117 factor analysis).

118

119 *Applied dietary pattern scores*

120 Although our exploratory factor analyses identified qualitatively consistent dietary patterns over time,
121 there were some differences in factor loadings between one and other timepoints (**Supplemental**
122 **Table 2**). In order to track dietary pattern z-scores for exactly the same patterns over time, we
123 calculated applied dietary pattern z-scores. These were estimated by applying the scoring coefficients
124 for each dietary pattern identified at 2 years of age, to all subsequent dietary intakes i.e. using the 2y
125 patterns as a template for later ages. This means that all children were scored at 2, 3, 4, 5, 6 and 8y of
126 age for the dietary patterns observed at 2 y of age.

127

128 *Tracking Analysis*

129 The transition from complementary foods to family food is usually completed between one and two
130 years of age, and our factor analyses showed that dietary patterns were most consistent from 2 years
131 of age onwards. For these reasons, we analyzed dietary pattern tracking from 2 to 8 years of age.

132 Tracking coefficients were estimated for each dietary pattern using generalized estimation equation
133 (GEE) models regressing the pattern z-score at 2 years of age on all subsequent pattern z-scores, as
134 suggested by Twisk (20), with a correlation matrix M-dependent ($M=1$). The tracking or stability in
135 dietary patterns analyses were performed with the applied dietary patterns z-scores, in order to track
136 exactly the same patterns over time. In brief, this analysis outputs a coefficient that ranges between 0
137 and 1, which correlates dietary pattern applied scores at all ages for each child. Tracking was
138 considered low if the stability coefficient was <0.4 , moderate if coefficient was 0.4 to 0.6, and strong
139 when it was >0.6 (21,22).

140

141 Although the tracking coefficient describes the longitudinal linear association between dietary pattern
142 scores at a population level, it describes average tracking. It does not specify for example, whether the
143 highest or the lowest scores for a pattern track more strongly. This is important, since higher scores
144 for a specific pattern would indicate a more or less healthy dietary intake. Therefore at each age,
145 children were classified into quartiles using their applied dietary pattern z-scores. The likelihood of
146 staying in the same extreme quartile at 2 and 8 years of age was calculated. For example, the
147 percentage of children in the highest quartile at 2y, remaining in the same extreme quartile at 8y, for
148 each dietary pattern. Binary logistic regression models were used to calculate the odds of being in the
149 same extreme quartile at 2 and 8 years of age, compared to being in any other quartile, adjusting only
150 for country.

151

152 Linear regression models were used to assess the direct and joint effect of those covariates mentioned
153 above on each applied dietary pattern score at 2 and 8y. Effect of individual variables (as for example
154 mother's education level, or birth order) on patterns at 2 and 8y were assessed by separated linear
155 regression models with enter method (only including country adjustment). Variables with a significant
156 effect ($p < 0.05$) on applied dietary patterns scores at 2 or 8y in simple models, were then included in
157 the final multivariate linear regression models for confounding. Therefore, in multivariate linear
158 regression models, all variables with a significant effect on applied dietary patterns scores in simple
159 linear regression models were included. In addition, in the multivariate linear regression models to
160 assess the predictors of dietary patterns scores at 8y, the dietary patterns scores at 2y were included
161 as possible predictors as well.

162

163 Statistical significance was accepted at the level $p < 0.05$.

164 All the statistical analyses were performed using SPSS 23.0 (IBM Corp., Armonk, NY, USA).

165

166 ***Ethics***

167 The study was performed following the principles of the Helsinki Declaration (23). The study was
168 submitted to and approved by local ethical committees. Parents or caregivers received written
169 information and gave signed consent for their child to participate in the study at all different study
170 periods (baseline to 2y, 3 to 6 and 7 to 8).

171

172 **RESULTS**

173 One-thousand six-hundred seventy-eight healthy term infants were recruited in neonatal services of
174 study centers. One-thousand one-hundred infants were still in the study at 1y of life. From these 1100
175 infants, 827 reported dietary intake data, and 633 were included in the exploratory factor analysis, as
176 194 were breastfed and could not be added for analyses at that timepoint (as breastmilk was not

177 quantified). Flow chart of participants, their country of residence, and sex during the study period are
178 reported in **Supplemental Figure 1**. All food diaries from children who were not unwell at the time of
179 the food recording were included for analyses. At all ages, > 85% of the diaries had 3 days recorded,
180 >7% had 2 days, <4.5% had only 1 day recorded.

181 The characteristics of study participants at recruitment and main timepoints are reported in
182 **Supplemental Table 3**. The baseline family characteristics of the study population at main study
183 timepoints (1y, 2y and 8y) are reported in **Supplemental Table 4**. In general, children of more educated
184 parents were more likely to stay in the study.

185 **Supplemental Table 2** shows the food factor loadings for each identified dietary pattern and the
186 proportion of variation explained by those patterns at each time point. At 1 year of age, two major
187 dietary patterns were identified. The first pattern was labelled as Core Foods Dietary Pattern (CORE);
188 food groups having the strongest positive loadings were vegetables, potatoes, fish, olive oil, white and
189 red meat, while ready-to-eat infant products, fruit juices, added sugars and confectionary had negative
190 loadings. The second pattern was characterized by high positive loadings for saturated spreads (such
191 as butter), added sugars, and cakes, biscuits and sweet pastries, and by negative loadings for olive oil
192 and slightly for fish. Therefore, the second pattern was labelled as "Poor Quality Fats and Added Sugar"
193 (F&S).

194
195 At ages 2, 3, 4, 5 and 8y, three major patterns were consistently identified: two had similarities to the
196 CORE and F&S found at 1 year, and a new third pattern was identified. Although there were differences
197 in factor loadings from one timepoint to another, common characteristics of the pattern labelled as
198 CORE across timepoints were: it was always characterized by positive loadings for vegetables, fruits
199 and olive oil and the majority of timepoints it was characterized by positive intakes of fish. Although
200 some timepoints white and red meat loaded positively on that pattern, processed meats never did so.
201 That CORE pattern was never characterized by positive intakes of added sugars, cakes, confectionary

202 nor fruit juices, and unless one timepoint, saturated spreads (such as butter) never loaded for that
203 pattern. One difference in the CORE foods pattern from 1 to 2 years was that cow's milk ceased to load
204 for that pattern at 2 years and 8 years and negatively loaded at 3 and 4 years.

205 Similarly, although loading factors differed between ages, the foods types loading for the pattern were
206 similar across ages for the F&S pattern. This pattern was characterized consistently by the intake of
207 saturated spreads (such as butter), added sugars and other sugar rich products such as cakes or
208 confectionery, or fruit juices. However, olive oil and fish were almost always loading negatively for that
209 pattern F&S. Although the F&S pattern at 2y was roughly similar to that at 1y, cow's milk changed from
210 having a positive loading at 1y to a negative loading at 2y and 3 years, and ceased to load from 4y
211 onwards.

212 The third new pattern found only from 2 years onwards was characterized as almost all timepoints by
213 positive loadings for milk, flavored milks, fish, eggs, white meat, processed meat, potatoes and olive
214 oil, and negative loadings for fresh fruits. Only at two and three years, vegetables loaded positively on
215 that pattern. This third pattern was labelled as "High Protein sources dietary pattern" (PROT). At 6y,
216 no factors were extracted since KMO for sample adequacy was <0.6.

217

218 The dietary pattern tracking coefficients from 2 to 8y were: 0.50 (95% CI: 0.43, 0.57) for CORE, 0.83
219 (95% CI: 0.76, 0.89) for F&S, and 0.53 (95% CI: 0.47, 0.59) for PROT. **Figure 1** shows the percentage of
220 children in each dietary pattern quartile (A for CORE, B for F&S and C for PROT) at 2 and 8 years of age.

221

222 Of those children in the highest quartile for the F&S pattern at 2y, 72% remained in this quartile at 8y
223 (Figure 1, B), translating to an OR= 3.6 (95% CI: 1.5, 8.4) for remaining in the top quartile, compared to
224 being in any other quartile at 8y of age. Of those children in the highest quartile for the CORE (Figure
225 1, A) and PROT (Figure 1, C) patterns at 2y, 45% and 36% remained in the same quartile at 8y,

226 respectively. This translated to an OR=2.01 (95% CI: 1.08, 3.8) for the CORE pattern, and a non-
227 significant odds to remain in the highest PROT pattern (OR= 0.80 (0.40, 1.57) p=0.510)

228 For those children in the lowest quartiles for the CORE, F&S and PROT patterns at 2 y of age, 42%,
229 47.5% and 50% remained in the lowest quartile at age 8y, respectively. The adjusted odds to remain in
230 the lowest quartiles of the dietary patterns from 2 to 8y were not statistically significant neither for
231 CORE (OR= 1.60 (0.73, 3.46) p=0.329), nor F&S (OR=1.11 (0.58, 2.09) p=0.758) nor PROT (OR=1.23
232 (0.59, 2.56) p=0.573).

233

234 **Table 1** shows predictors of dietary patterns scores at 2 and 8 years of age. Only country explained the
235 score for CORE at 2y. However, at 8y, CORE scores were positively associated with the CORE score at
236 2y, country and higher maternal education.

237

238 F&S dietary pattern scores at 2y were explained by country and were positively associated with birth
239 order of the child under study. Second born children had significantly higher F&S scores than first born
240 children. F&S scores at 8y were positively associated with F&S score at 2y and country of origin, only.

241

242 A high maternal education level was inversely associated with PROT dietary pattern score at 2y. In
243 contrast to the CORE and F&S patterns, the PROT pattern score at 8y was not associated with score for
244 the same pattern at 2y. At 8y of age, the PROT scores were explained by country of origin and were
245 inversely associated with having parents born in a different country than that of the study center and
246 a high maternal education level.

247

248 **DISCUSSION**

249 In this longitudinal study of European children from five countries, we identified three major dietary
250 patterns which appear to establish between one and two years of age and track into mid-childhood.

251 Of these, a dietary pattern characterized by added sugar, unhealthy fats and poor consumption of fish
252 and olive oil tracked most strongly in this cohort; children having the highest scores for that pattern at
253 2y, were 3 times more likely to have the highest scores for that pattern at age 8. Similarly, children
254 with the highest scores for a dietary pattern based on healthy core foods at 2y, were twice as likely to
255 continue having at age 8y, a diet still characterized by unprocessed healthy foods such as vegetables,
256 fruits, olive oil and fruits. Those findings indicate that early interventions to encourage healthy eating
257 habits are imperative.

258

259 To our knowledge, this is one of few studies tracking empirical dietary patterns from infancy and across
260 childhood (up to 8y of age). From one year of age, two distinct dietary patterns were prevalent: a “Core
261 Foods Dietary Pattern” and a “Poor Quality Fats and Added Sugar” pattern. From two years, a third
262 dietary pattern appeared beside the first two: a “High Protein Sources” dietary pattern characterized
263 by a high consumption of animal products. This third pattern, was prevalent from 2 to 8 years,
264 however, children with the highest scores for that pattern at 2y, had no increased risk to have the
265 highest scores at age 8y.

266

267 Other studies have reported empirical dietary patterns in very young children. At six and twelve
268 months, two dietary patterns (“infant guidelines” and “adult foods”) were identified using principal
269 components analysis in the Southampton cohort (24). These patterns are comparable to those
270 identified in the current EU cohort study: the “infant guidelines” pattern was characterized by
271 vegetables, fruits, meat and home cooked meals (similarly to the pattern we labelled as “Core foods”);
272 the “adult foods” pattern was characterized by consumption of bread, snacks, biscuits and processed
273 fruits (similar to our “Poor Quality Fats and Added Sugar” pattern that included a high proportion of
274 discretionary or non-core foods). At 14 months of age, a “Health conscious” and a “Western-like”
275 dietary pattern were identified (7) in 2420 toddlers from the Generation R cohort. These dietary

276 patterns were also similar to the CORE and F&S patterns observed in our study, however, it is unknown
277 whether those patterns tracked to later ages.

278

279 Similarly, in more than 9000 children from the Avon Longitudinal Study of Parents and Children
280 (ALSPAC) cohort, three dietary patterns were identified at 2y of age using principal components
281 analysis: “family foods” correlated with a traditional British family diet (e.g. meat, fish, puddings,
282 potatoes and vegetables); “sweet and easy” associated with foods high in sugar and foods requiring
283 little preparation (as sweets, crisps and soups); and “health conscious” associated with fruit,
284 vegetables, eggs, nuts and juices (25). Despite some differences, these dietary patterns had similarities
285 with ours: “family foods” similar to CORE and “sweet and easy” to F&S. Beyond three years of age,
286 three other dietary patterns were observed as: “processed”, ‘traditional’ and ‘health conscious’. These
287 patterns showed moderate stability (with Pearson correlations varying from 0.35 to 0.46) from 3y
288 onwards (26). The different patterns found at 2y and from 3y onwards could be due to the use of a
289 different dietary assessment tool, or a true change in diet in young childhood, as was observed
290 between 1 and 2 years of age in our study

291

292 Between 1 and 2y of age, a transition period typically occurs, during which the infant progresses to
293 family foods and this was reflected in changing dietary patterns in this cohort. Our findings are in
294 general agreement with previously published work, which reports changes in dietary patterns between
295 one and two years of age. This was also shown by an analysis conducted in approximately 500
296 Australian toddlers from two cohorts, the NOURISH and South Australian Infant Dietary Intake (SAIDI)
297 study, identified two dietary patterns at 14 and 24 months. At 14 months, a “core foods” dietary
298 pattern and a “mixture of core and non-core foods” were observed. However, at 2y, they were able to
299 more clearly distinguish two dietary patterns, named as “core foods” and “non-core foods” (28).

300

301 A worth noting finding is that the less healthy pattern, observed from 1y, had the strongest tracking
302 from 2 to 8y. In ~1000 two year old children from the EDEN cohort, two dietary patterns were identified
303 cross-sectionally, and were named as “Processed and fast foods” and “Guidelines” dietary patterns.
304 At 5 years of age, they identified different dietary patterns: a “Processed and fast foods” pattern and
305 a “Protein-rich and diversified” pattern. While these dietary patterns were reported to moderately
306 track from two to five years (8), the “Processed and fast foods” dietary pattern showed the strongest
307 tracking ($r= 0.35$) which concurs with the F&S pattern tracking most strongly in our study.

308

309 There were several factors predisposing to higher or lower pattern scores in this European cohort of
310 young children. The country of origin was a consistent predictor of dietary patterns scoring: children
311 from Spain and Italy had significantly higher CORE scores at 2 and 8y, Poland had the highest scores
312 for F&S pattern at 2 and 8y, and Spain had significantly higher scores for PROT pattern at both ages.

313 In general, mother’s education level predisposed to a healthier dietary patterns, characterized by
314 lower scores for the “Protein sources” pattern at 2 and 8y, and higher scores for the “Core foods”
315 pattern at 8y, independently of the country of origin. This finding was consistent with results from the
316 RAINE study, in which greater maternal education level was associated with a healthier dietary pattern
317 (27). Similarly, in the ALSPAC cohort, mother’s educational attainment was associated with higher
318 scores of healthier dietary patterns extracted through different methods (4). Similarly, two Danish
319 observational cohorts observed an influence of the family characteristics on infants’ dietary patterns
320 at 9 months of age: infants from families with lower social class and higher BMI, already showed lower
321 scores in a “health conscious” dietary pattern (6).

322

323 Interestingly, our multivariate analyses showed that having an older sibling predisposes to a higher
324 F&S score at 2y but not at 8y. This may be due to parents introducing discretionary foods that are
325 typically given to older children, to their younger siblings, at an earlier age e.g. cakes, biscuits and

326 confectionery. However, this association was diminished at 8y of age, suggesting that other factors
327 may have a greater influence on dietary intakes at older ages.

328

329 As this was a longitudinal study, a potential limitation of this analysis is the decreasing number of
330 children participating in each follow up over time, which could introduce some bias due to non-random
331 loss to follow up. Such attrition could introduce bias, through for example, a greater proportion of
332 families with a higher education level and greater interest in diet and health remaining in the study at
333 8y of age, than other families. Our longitudinal analyses included all data available at each time point,
334 rather than restricting the analysis only to those children who completed every follow up. Another
335 potential limitation is that the study sample was not distributed equally in all the countries. As
336 expected, the country of origin was a strong predictor of dietary pattern scores, since every child is
337 scored for all of the patterns, the differences by country do not prevent from being associated with
338 health and disease conditions.

339

340 There are several strengths in the present work. Firstly, the dietary data were collected in five different
341 European countries using 3 day food diaries following standardized methodology, which provides finer
342 detail on dietary intakes than food frequency questionnaires. Secondly, we observed quite consistent
343 dietary patterns in our study population from age 1 year upwards, supporting the fact that the dietary
344 patterns were not a product of chance. Furthermore, we performed a confirmatory analyses to ensure
345 we tracked exactly the same dietary patterns from 2 to 8 y. The methods we used for tracking dietary
346 patterns, has some advantages compared to those used in previously published articles: while others
347 used simple correlations between dietary patterns scores at two timepoints, we used generalized
348 estimating equations, to calculate tracking coefficients using data from over 5 timepoints. Finally, our
349 tracking analysis of extreme dietary pattern quartiles provides important additional information for
350 developing targeted interventions.

351

352 The importance of the present findings, rely on the fact that early diet has been associated with later
353 obesity and related disorders (29). A recent systematic review concluded that dietary patterns that are
354 high in energy-dense, high-fat and low-fiber foods predispose young people to later overweight and
355 obesity (30). Dietary patterns, which hold a comprehensive way of understanding diet overall (rather
356 than single nutrients or foods), are a powerful way of analyzing diet and its effects on health.

357 It is worth highlighting as well, that children within the studied age range did not have full autonomy
358 on food choice, as the parents or caregivers usually exert a control. However, the fact that parents
359 influence children's diet in either a beneficial or detrimental way, it remains to be studied whether this
360 influence remains later in life, when subjects will have more decision autonomy.

361

362 All of the previous studies, together with findings from our present work, support the need for early
363 educational interventions in the complimentary feeding stage, when children are being introduced to
364 adult foods, to establish healthy eating habits early. It is worth mentioning that educational
365 interventions should not only focus on introduction of core foods such as vegetables and fruits, but
366 should also focus on the avoidance of discretionary, low quality foods at early ages.

367

368 In summary, we conclude that dietary patterns may be established during a transition period between
369 one and two years of age, and then track to mid-childhood. Specifically, unhealthy dietary patterns
370 (characterized by added sugars, unhealthy fats and poor consumption of vegetables, fruits, fish and
371 olive oil) track most strongly into midchildhood in this European cohort. These findings highlight the
372 importance of establishing policies of educational programs during this transition period to reinforce
373 healthy dietary habits. Specifically, families with mothers of low educational attainment may need
374 additional support to avoid the introduction of Poor Quality Fats and Added Sugar rich products in

375 their child's diet. Further analyses in this cohort will reveal whether dietary patterns are associated
376 with later obesity and metabolic disease risk.

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Supplemental Table 1. Description of major food groups included in exploratory factor analysis.

FOOD GROUPS FOR FACTOR ANALYSIS	FOOD ITEMS
Refined and whole grains	Bread and rolls, rice, pasta, noodles, flours, infantile cereals, breakfast cereals
Processed cereal products	Convenience food of pasta (filled pasta as tortellini, industrially processed and pizza)
Eggs	Eggs
All fresh fruits	Apple, pear, peach, apricot, plum, nectarine, cherries, strawberry, berries, pineapple, avocado, banana, watermelon, melon, kiwi, orange, etc.
Processed fruit products	Compotes, fruit concentrated products, convenience food of fruits
Leafy green and salads	Spinach, celery, parsley, lettuce, endives, cabbage, lettuce
Vegetables	Cruciferous (cabbage, cauliflower, broccoli), Fruiting vegetables (eggplant, cucumber, pepper, tomato...), Leguminous vegetables (beans green, peas green...), Others (artichoke, asparagus, onion, ...), pulses
Soups and vegetable dishes	Soups, vegetable dishes
Ready-to-eat infant foods	Pureed Infant fruits, vegetables, infant meals
Savory snacks	Chips, crisps, fried potatoes
Potatoes	Potatoes (boiled/ steamed/ smashed), tapioca, starchy plants
Cow's milk & regular yoghurt	Milk and curded milk, soya beverage, milk and cheese dishes
Flavored milk products	Yogurts with added sugars, milk deserts, milk shakes
Processed milk products	Ice cream, cream, puddings
Hard cheese	Hard cheese (hard, semi-hard)
Soft cheese	Soft cheese, cream cheese
Fruit juice	Fruit juices, fruit jelly
Teas	Teas
Olive oil	Olive oil and olives
Saturated spreads	Margarine, butter, vegetable fats
Other oils and oily sauces	Other oils and mayonnaise
Other sauces	Tomato sauce, mustard, soya sauce
Confectionery	Confectionery, sweets, marzipan, nougat, chocolate, chocolate, pralines, sweet products
Added sugars	Sugar, honey, sweet spreads, cocoa, jam
Cakes, biscuits, sweet pastries	Tarts, cakes, pastries, biscuits, crackers, crepes
Lean, fatty fish and sea food	Cod, red fish, whiting, flatfish, flounder, plaice, sole, herrings, mackerel, tuna fish, salmon, trout, crustaceans, shell fish, etc.
Red meat	Beef, veal, pork, lamb, offal
White and low fat meat	Poultry, Horse, goat, rabbit meat, winged game
Processed meat	Animal products, sausages, ham, bacon, cured meat, meat products

Supplemental Table 2. Factor loadings for all dietary patterns, exploratory factor analyses at children's ages 1 to 3 years.

	1 year		2 years			3 years		
KMO sampling adequacy	0.7		0.6			0.6		
% VARIANCE EXPLAINED	15.2%		13.1%			14.3%		
	PATTERNS		PATTERNS			PATTERNS		
	CORE	F&S	CORE	F&S	PROT	CORE	F&S	PROT
% variation for each pattern	9.0%	6.2%	3.8%	5.1%	4.1%	3.5%	6.6%	4.2%
FOOD GROUPS								
Fresh fruits	0.29	0.16	0.25	0.11	-0.12	0.34	0.01	0.01
Fresh leafy greens & salads	0.24	0.05	0.16	-0.01	-0.04	0.19	-0.02	0.01
Fresh vegetables	0.75	-0.07	0.34	0.14	0.27	0.28	0.18	0.26
Soups	-0.13	-0.03	-0.15	0.07	0.03	-0.05	0.32	0.02
Potatoes	0.76	0.10	0.36	0.28	0.35	0.04	0.27	0.21
Grains (refined & whole meal)	-0.17	0.11	-0.02	0.17	0.03	0.05	0.17	0.02
Processed cereal products	0.00	-0.01	0.02	-0.18	-0.21	0.21	-0.13	-0.34
Cow's milk & regular yoghurt	0.17	0.16	-0.09	-0.38	0.09	-0.25	-0.35	-0.03
Flavored milks	0.16	0.03	-0.06	-0.13	0.52	-0.26	-0.18	0.20
Processed milk products	-0.07	-0.35	0.09	-0.04	-0.21	0.09	-0.03	-0.07
Hard cheese	-0.12	-0.57	0.12	-0.15	-0.22	0.20	-0.12	0.02
Soft cheese	-0.15	0.17	0.06	0.44	-0.05	0.04	0.40	-0.06
Fish	0.51	-0.17	0.27	-0.27	0.34	0.02	-0.33	0.36
Eggs	-0.06	0.07	0.07	0.12	0.21	0.06	0.12	0.38
White meat	0.48	-0.01	0.20	-0.13	0.36	-0.06	-0.13	0.35
Red meat	0.32	0.02	0.30	-0.03	0.03	0.11	-0.02	0.15
Processed meat	0.02	0.08	-0.06	-0.01	0.37	-0.35	-0.20	0.35
Processed fruit ¹	-0.23	-0.19	-0.08	-0.08	0.05			
Ready-to-eat IF ¹	-0.56	0.10	-0.26	0.06	-0.02			
Chips and savory snacks	-0.00	0.01	0.04	-0.09	0.24	-0.01	0.00	0.06
Olive oil & olives	0.30	-0.82	0.62	-0.48	0.16	0.48	-0.52	0.44
Saturated spreads	-0.05	0.46	-0.08	0.55	0.04	-0.07	0.71	0.02
Other oils and oily sauces	0.01	0.17	0.04	0.07	0.11	-0.19	-0.10	0.26
Other sauces (not oil based)	0.00	0.18	-0.10	0.00	-0.03	-0.12	0.17	0.17
Added sugars	0.02	0.24	-0.12	0.31	0.02	-0.20	0.20	0.07
Cakes, biscuits, sweet pastries	0.08	0.31	0.01	0.03	-0.05	-0.06	-0.01	0.02
Confectionary	-0.02	0.11	-0.12	0.17	0.00	-0.11	0.27	-0.05
Fruit juices	-0.24	0.16	0.01	0.28	0.03	-0.07	0.23	0.01
Teas	-0.24	0.16	-0.12	0.27	-0.14	0.07	0.19	-0.11

Table 2’. Factor loadings for dietary patterns extracted from exploratory factor analyses at children’s ages 4 to 8 years (Cont.).

	4 years			5 years			8 years		
KMO sampling adequacy	0.6			0.6			0.6		
% VARIANCE EXPLAINED	13.5%			14.2%			16.1%		
	PATTERNS			PATTERNS			PATTERNS		
FOOD GROUPS	CORE	F&S	PROT	CORE	F&S	PROT	CORE	F&S	PROT
% variation for each pattern	3.1%	6.0%	4.4%	6.2%	3.1%	5.0%	5.3%	5.3%	5.5%
Fresh fruits	0.25	-0.20	-0.20	0.17	0.05	-0.16	0.40	-0.10	-0.18
Fresh leafy greens & salads	0.10	-0.14	-0.04	0.20	-0.07	-0.11	0.15	-0.16	-0.09
Fresh vegetables	0.55	-0.15	0.05	0.33	0.41	0.05	0.57	0.09	-0.05
Soups	-0.01	0.28	0.12	-0.27	-0.01	0.08	-0.14	0.04	-0.13
Potatoes	0.19	0.13	0.28	-0.01	0.29	0.05	0.10	0.44	0.05
Grains (refined & whole meal)	0.17	0.04	-0.00	-0.13	0.10	0.10	0.21	-0.24	-0.06
Processed cereal products	-0.16	-0.12	-0.19	0.18	-0.19	-0.23	-0.03	-0.21	-0.11
Cow's milk & regular yoghurt	-0.17	-0.11	0.31	0.05	-0.07	0.35	0.10	0.01	0.38
Flavored milks	-0.10	0.06	0.47	-0.11	0.05	0.40	-0.11	0.10	0.35
Processed milk products	-0.00	-0.04	-0.12	-0.00	0.02	-0.14	-0.22	0.01	-0.04
Hard cheese	-0.06	-0.13	-0.10	0.26	-0.07	-0.02	0.08	-0.09	0.08
Soft cheese	0.31	0.24	-0.12	-0.16	0.42	-0.27	0.09	0.30	-0.31
Fish	0.07	-0.33	0.33	0.29	0.03	0.33	0.26	-0.18	0.26
Eggs	0.12	0.05	0.25	0.01	0.24	0.24	0.27	0.31	0.05
White meat	0.04	-0.15	0.27	0.19	0.07	0.36	0.17	0.03	0.32
Red meat	0.12	-0.22	0.05	0.16	-0.02	0.01	0.00	0.09	0.22
Processed meat	-0.09	-0.03	0.38	-0.14	0.13	0.43	-0.04	0.03	0.25
Processed fruit ¹									
Ready-to-eat IF ¹									
Chips and savory snacks	-0.04	-0.01	-0.19	0.13	-0.03	-0.12	0.01	-0.12	0.24
Olive oil & olives	0.17	-0.71	0.26	0.78	0.09	0.38	0.59	-0.24	0.57
Saturated spreads	0.26	0.59	-0.09	-0.46	0.43	-0.27	-0.02	0.69	-0.37
Other oils and oily sauces	-0.12	0.06	0.35	-0.06	-0.02	0.25	-0.15	0.01	0.42
Other sauces (not oil based)	-0.14	0.07	0.01	-0.29	-0.17	-0.03	-0.31	0.01	-0.04
Added sugars	0.04	0.33	0.06	-0.18	0.13	-0.03	-0.12	0.45	0.05
Cakes, biscuits, sweet pastries	-0.17	0.03	-0.04	-0.04	-0.09	-0.13	-0.24	-0.19	0.01
Confectionary	0.00	0.29	0.04	-0.23	0.13	-0.10	-0.14	-0.00	-0.01
Fruit juices	-0.14	0.25	-0.02	-0.29	-0.03	-0.06	-0.19	0.28	-0.12
Teas	-0.03	0.11	-0.17	-0.05	-0.01	-0.19	-0.11	0.01	-0.20

KMO: Kaiser-Meyer-Olkin Test for suitability of the study sample for factor analysis; CORE: Core Foods Dietary Pattern, F&S: Poor Quality Fats and Added Sugar Dietary Pattern, PROT: High Protein Sources Dietary Pattern. Bold letters for all food factor loadings ≥ 0.2 .¹Processed fruits and ready-to-eat infant products were consumed only in early infancy, as they were commercial products for infants.

Supplemental Table 3. Participants' characteristics at birth and ages 1, 2 and 8 years, for the study sample available (still in study and with dietary data reported) at 1, 2 and 8 years. Values are means and \pm SDs.

	1 year	2 years	8 years
<i>Birth characteristics</i>	<i>n = 633</i>	<i>n = 702</i>	<i>n = 392</i>
Gestational age (weeks)	39.8 [\pm 1.2]	39.8 [\pm 1.2]	39.9 [\pm 1.2]
Birth weight (kg)	3.3 [\pm 0.3]	3.3 [\pm 0.3]	3.3 [\pm 0.3]
Birth length (cm)	50.7 [\pm 2.6]	50.7 [\pm 2.6]	50.6 [\pm 2.6]
Head circumference at birth (cm)	34.2 [\pm 1.3]	34.4 [\pm 1.3]	34.2 [\pm 1.4]
<i>Anthropometry in childhood</i>	<i>n = 624</i>	<i>n = 728</i>	<i>n = 397</i>
Weight (kg)	9.9 [\pm 1.1]	12.4 [\pm 1.4]	28.6 [\pm 6.1]
Length (cm)	75.7 [\pm 2.6]	88.0 [\pm 3.2]	
Height (m)			1.30 [\pm 0.05]
Head circumference (cm)	46.2 [\pm 1.3]	48.6 [\pm 1.5]	52.2 [\pm 1.4]
Body Mass Index (kg/m ²)	17.2 [\pm 1.5]	16.2 [\pm 1.3]	16.9 [\pm 2.6]
Body Mass Index (z-score)	1.51 [\pm 0.91]	0.34 [\pm 0.93]	0.60 [\pm 1.19]

Supplemental Table 4. Baseline family characteristics of the study sample available (still in study and with dietary data reported) at ages 1, 2 and 8 years.

	1 year	2 years	8 years
	<i>n</i> = 633	<i>n</i> = 747	<i>n</i> = 399
Mother's education level, <i>n</i>	631	745	398
<i>Low, n (%)</i>	172 (27.3)	150 (20.1)	60 (15.1)
<i>Medium, n (%)</i>	338 (53.6)	379 (50.9)	200 (50.3)
<i>High, n (%)</i>	121 (19.2)	216 (29)	138 (34.7)†, α
Father's education level, <i>n</i>	621	739	398
<i>Low, n (%)</i>	192 (30.9)	175 (23.4)	82 (20.6)
<i>Medium, n (%)</i>	332 (53.5)	373 (50.5)	198 (49.7)
<i>High, n (%)</i>	97 (15.6)	191 (25.8)	118 (29.6)
Both parents foreigner, <i>n (%)</i>	20 (3.2)	20 (2.7)	4 (1.0)*, α
Single mother, <i>n (%)</i>	36 (5.7)	28 (3.8)	12 (3.0)
Birth order,	633	747	399
<i>First child, n (%)</i>	129 (66.5)	441 (59.0)	229 (57.4)
<i>Second child, n (%)</i>	55 (28.4)	237 (31.7)	138 (34.6)
<i>More than second child, n (%)</i>	10 (5.2)	69 (9.2)	32 (8.0)
Mother's BMI (kg/m ²)	23.9 [±4.4]	23.4 [±4.2]	23.5 [±4.1]
Father's BMI (kg/m ²)	26.0 [±3.7]	26.1 [±3.7]	26.4 [±3.5]

No significant differences in Chi² test for distributions at 24 months irrespective to 12 months.
 ‡ p-value < 0.001, † p-value = 0.006, * p-value = 0.011 compared to 1 year, α p-value = 0.042 compared to 2 years. ANOVA and posthoc bonferroni comparisons for mother's and father's BMI showed no statistical differences between timepoints. BMI = Body Mass Index, values are means ± SDs.

Table 1. Predictors of Dietary Pattern scores at children's ages 2 and 8 years

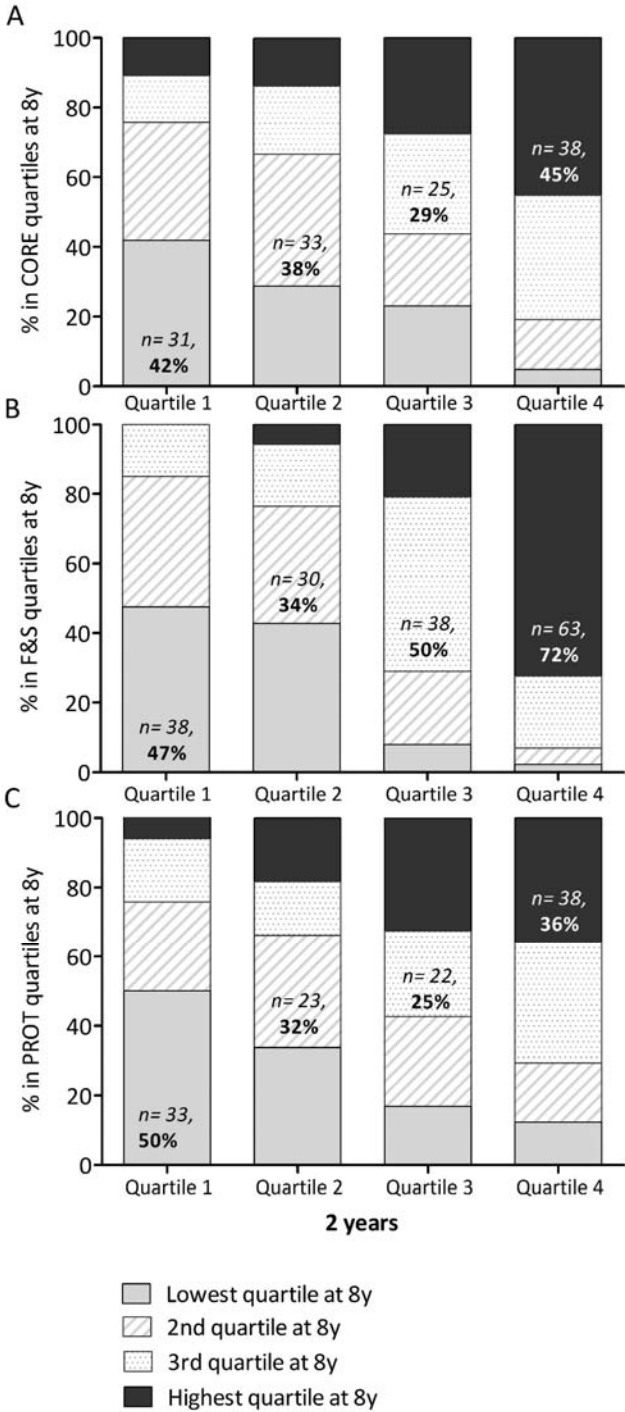
Predictors of Dietary Patterns scores at age 2 years						
Core Foods pattern score at 2y						
		B	95% CI	p-value	R ²	Model p-value
Country ¹	<i>Germany</i>	-0.93	(-1.10, -0.75)	<0.001	23.5%	p<0.001
	<i>Belgium</i>	-0.70	(-0.89, -0.50)	<0.001		
	<i>Italy</i>	0.09	(-0.06, 0.23)	0.23		
	<i>Poland</i>	-0.31	(-0.48, -0.15)	<0.001		
Poor Quality Fats and Added Sugar pattern score at 2y						
		B	95% CI	p-value	R ²	Model p-value
Mother's education level ²	<i>Medium</i>	0.01	(-0.010, 0.12)	0.83	62.3%	p<0.001
	<i>High</i>	-0.07	(-0.21, 0.07)	0.32		
Father's education level	<i>Medium</i>	-0.07	(-0.18, 0.04)	0.20		
	<i>High</i>	-0.13	(-0.27, 0.01)	0.07		
Birth order (compared to first child)						
	<i>Second child in the family</i>	0.09	(0.01, 0.12)	0.04		
	<i>Third (or >) child in the family</i>	0.10	(-0.05, 0.25)	0.19		
Country	<i>Germany</i>	0.94	(0.81, 1.08)	<0.001		
	<i>Belgium</i>	0.97	(0.81, 1.19)	<0.001		
	<i>Italy</i>	0.04	(-0.07, 0.15)	0.47		
	<i>Poland</i>	1.69	(1.57, 1.82)	<0.001		
Protein sources pattern score at 2y						
		B	95% CI	p-value	R ²	Model p-value
Mother's education level	<i>Medium</i>	-0.07	(-0.18, 0.04)	0.20	49.0%	p<0.001
	<i>High</i>	-0.16	(-0.29, -0.03)	0.014		
Country	<i>Germany</i>	-1.19	(-1.33, -1.05)	<0.001		
	<i>Belgium</i>	-0.96	(-1.12, -0.80)	<0.001		
	<i>Italy</i>	-1.40	(-1.52, -1.29)	<0.001		
	<i>Poland</i>	-0.78	(-0.91, -0.64)	<0.001		

Table 1’. Predictors of Dietary Pattern scores at children’s ages 2 and 8 years (*Cont.*).

Predictors of Dietary Patterns scores at age 8 years						
Core Foods pattern score at 8 years						
		B	95% CI	p-value	R ²	Model p-value
Core foods pattern score at 2y		0.23	(0.10, 0.36)	0.001	37.6%	p<0.001
Mother’s education level	<i>Medium</i>	0.17	(-0.11, 0.45)	0.23		
	<i>High</i>	0.37	(0.07, 0.67)	0.014		
Country	<i>Germany</i>	-1.36	(-1.68, -1.03)	<0.001		
	<i>Belgium</i>	-1.39	(-1.80, 0.99)	<0.001		
	<i>Italy</i>	0.10	(-0.17, 0.37)	0.46		
	<i>Poland</i>	-0.61	(-0.87, -0.34)	<0.001		
Poor Quality Fats and Added Sugar pattern score at 8 years						
		B	95% CI	p-value	R ²	Model p-value
Poor Quality Fats and Added Sugar pattern score at 2y		0.26	(0.11, 0.41)	0.001	65.8%	p<0.001
Both parents foreign ³		-0.29	(-1.23, 0.65)	0.54		
Mother’s BMI [kg/m ²]		-0.02	(-0.04, 0.03)	0.10		
Country	<i>Germany</i>	1.29	(1.00, 1.57)	<0.001		
	<i>Belgium</i>	1.18	(0.85, 1.51)	<0.001		
	<i>Italy</i>	0.24	(0.02, 0.45)	0.030		
	<i>Poland</i>	1.70	(1.38, 2.02)	<0.001		
Protein sources pattern score at 8 years						
		B	95% CI	p-value	R ²	Model p-value
Mother’s education level	<i>Medium</i>	-0.17	(-0.40, 0.05)	0.14	39.2%	p<0.001
	<i>High</i>	-0.26	(-0.51, -0.02)	0.031		
Both parents foreign ³		-1.53	(-2.36, -0.71)	<0.001		
Country	<i>Germany</i>	-1.17	(-1.42, -0.93)	<0.001		
	<i>Belgium</i>	-0.74	(-1.06, -0.43)	<0.001		
	<i>Italy</i>	-1.12	(-1.34, -0.91)	<0.001		
	<i>Poland</i>	-0.13	(-0.34, 0.09)	0.24		

Table 1. Each multivariate model separated by a line. Tested predictors in multivariate models were country, gender, mother's and father's education level (*low* for no completed studies and elementary, *medium* for professional studies and intermediate cycles, *high* for university degrees, master and doctorate), mother's and father's BMI, being single mother, mother's age at child's birth, birth order, ³both parents foreign = both parents having a different country of origin than that of the study center, being breastfed vs. formula fed and formula intervention. As variables with effect on patterns at 8y, were also considered dietary patterns scores at 2 years. Effect of all variables were first tested in simple models adjusted by country; only those with a significant effect ($p < 0.05$) in simple models were introduced in the multivariate models. ¹All countries compared to Spain as reference; ²compared to low level of education.

FIGURE 1. Proportion of children at 2 years of age remaining in the same quartile of applied dietary pattern z-scores at 8 years of age.



CORE: Core Foods Dietary Pattern, F&S: Poor Quality Fats and Added Sugar Dietary Pattern, PROT: High Protein Sources Dietary Pattern. Figure shows that 72% of the children in the highest quartile of F&S pattern at 2y remained in this extreme quartile at 8y (OR= 3.6 (95% CI: 1.5, 8.4) for remaining in the top quartile, compared to being in any other quartile at 8y of age); 45% of the children in the highest quartile for the CORE pattern at 2y remained at the same highest quartile at 8y (OR=2.01 (95% CI: 1.08, 3.8)); 36% of the children in the highest PROT pattern quartile at 2y remained in the same quartile at 8y, with a non-significant odds to remain in the highest PROT pattern. The adjusted odds to remain in the lowest quartiles of the dietary patterns from 2 to 8y were not statistically significant.