Clinical Nutrition 38 (2019) 2333–2341

Contents lists available at ScienceDirect

Clinical Nutrition

journal homepage: http://www.elsevier.com/locate/clnu

Original article

No independent associations between preconception paternal dietary patterns and embryonic growth; the Predict Study

Elsje C. Oostingh [a], Iris de Vos [b], Annelies C. Ham [a], Elske M. Brouwer-Brolsma [b], Sten P. Willemsen [a, c], Alex J. Eggink [a], Eric A.P. Steegers [a], Régine P.M. Steegers-Theunissen [a, *]

[a] Department of Obstetrics and Gynecology, Erasmus MC, University Medical Centre Rotterdam, PO Box 2040, 3000 CA Rotterdam, the Netherlands
[b] Division of Human Nutrition, Wageningen University, PO Box 17, 6700 AA Wageningen, the Netherlands
[c] Department of Biostatistics, Erasmus MC, University Medical Centre Rotterdam, PO Box 2040, 3000 CA Rotterdam, the Netherlands

ARTICLE INFO

Article history:
Received 1 March 2018
Accepted 18 October 2018

Keywords:
Nutrition
Virtual reality
3D ultrasound
Crown-rump length
Embryonic volume
Periconception period

SUMMARY

Background & aim: Several studies show the importance of periconceptional maternal dietary patterns on human embryonic growth. Healthy paternal nutrition has been associated with better semen quality and fecundability, however, evidence on the impact on pregnancy outcome is limited. Therefore, the aim of this study was to investigate the association between preconception paternal dietary patterns and first trimester embryonic growth using the parameters longitudinal crown-rump length (CRL) and embryonic volume (EV).

Methods: A total of 638 couples were enrolled in the Rotterdam Periconceptional Cohort and received longitudinal three dimensional transvaginal ultrasound scans from 7^{+0} up to 12^{+0} weeks of gestation. Virtual reality software was used to perform offline measurements of the embryonic CRL and EV. Food frequency questionnaires (FFQ) were used to estimate habitual food intake in couples. Principal component analysis (PCA) was performed to identify paternal and maternal dietary patterns. Linear mixed models adjusted for potential confounders were applied to analyze associations between paternal and maternal dietary patterns and embryonic growth parameters.

Results: The paternal dietary patterns retrieved were identified as “Whole wheat grains and Vegetables”, “Sauces and Snacks Refined Grains”, “Fish and Legumes” and explained 27.5% of the total variance of the dietary intake. No significant additional effects, independent of maternal dietary patters and other maternal and paternal potential confounders, were shown of these paternal dietary patterns on embryonic growth in spontaneous or IVF/ICSI pregnancies.

Conclusion: No significant effects of paternal dietary patterns independent of maternal dietary patters and other parental potential confounders on embryonic growth parameters could be established in spontaneous or IVF/ICSI pregnancies. The biological importance of paternal nutrition on semen quality, however, supports the need of periconceptional tailored nutritional counselling of couples trying to conceive.

1. Introduction

The global epidemic of overweight and obesity affects approximately 2 billion men and women aged over 18 years [1]. A poor balance between nutrition and physical exercise is the main cause of overweight and obesity affecting short- and long-term health, but also reproduction.

The periconception period is a critical timespan in reproduction covering 14 weeks before up to 10 weeks after conception [2]. Previous research shows that periconceptional parental characteristics and lifestyle factors influence embryonic growth. For example,

List of abbreviations: BMI, body mass index; CRL, crown-rump length; EV, embryonic volume; IUI, intrauterine insemination; IVF, in vitro fertilization; ICSI, intracytoplasmic sperm injection; LMP, last menstrual period; FFQ, food frequency questionnaire; 3D-US, three dimensional ultrasound.

* Corresponding author. Department of Obstetrics and Gynecology, Erasmus MC, University Medical Centre Rotterdam, PO Box 2040, 3000 CA Rotterdam, the Netherlands.
E-mail address: r.steegers@erasmusmc.nl (R.P.M. Steegers-Theunissen).

https://doi.org/10.1016/j.clnu.2018.10.011

higher maternal age and folic acid supplement use are associated with increased embryonic growth while periconception smoking and alcohol consumption are associated with decreased embryonic growth trajectories [3,4]. Moreover, paternal birthweight is positively associated with embryonic growth trajectories [5].

Current scientific evidence strongly suggests that nutrition during this early period of life influences gametogenesis, fertilization, embryogenesis and placentation with consequences for growth and development of the fetus, neonate and even health in later life [3]. However, most evidence of the impact of periconceptional nutrition on fertility and pregnancy outcomes has been investigated in women. Strong adherence to a healthy-, Mediterranean- or one-carbon-rich dietary pattern have been associated with an increased chance of pregnancy and reduced risks of several congenital malformations [6–9].

Research on the involvement of paternal nutrition on reproduction has been mainly focused on semen quality parameters. A review by Salas-Huetos et al., showed that healthy nutrition contributes to improved semen quality, whereas high intake of red meat, processed meat, and caffeine were inversely associated with fecundability [10]. This was substantiated by another review which also showed that the intake of trans and saturated fats was consistently related to poor semen quality [9].

We hypothesize that paternal nutrition is not only important for semen quality, but also for embryonic development as a consequence of contributions to epigenetic reprogramming and influencing nutritional behavior of the pregnant woman. This is relevant because a small embryo is associated with an increased risk of miscarriage, congenital malformations, fetal growth restriction and even with features of cardiovascular- and metabolic diseases in childhood [3,11,12].

So far the evidence is limited to animal studies providing evidence for a transgenerational impact of the paternal diet on offspring health by showing that offspring of male mice fed a low protein diet or a folate deficient diet had compromised cardiovascular and metabolic functioning in later life [13,14]. One of the possible explanations is the influence of the preconception paternal diet on the epigenetic programming of male gametes and subsequent transmission to the embryo [15,16].

Nowadays, in human studies embryonic growth and development can be investigated with high precision of crown-rump length (CRL) (Fig. 2a) and embryonic volume (EV) (Fig. 2b) measurements using transvaginal 3-dimensional ultrasound (3D-US) with virtual reality (VR) techniques [17–19].

From this background the aim of this study was to investigate whether periconceptional paternal dietary patterns contribute to embryonic growth independent of the influence of maternal dietary patterns.

2. Materials and methods

2.1. Study population

The present study was embedded in the Rotterdam Periconceptional Cohort (Predict Study), an ongoing prospective hospital-based birth cohort study, conducted at the department of Obstetrics and Gynecology of the Erasmus University Medical Centre, Rotterdam, the Netherlands [20]. The protocol was approved by the Central Committee on Research in The Hague and by the local Medical Ethical Committee of the Erasmus MC in Rotterdam. All participants were informed about the study and signed a written informed consent. Details of this study have previously been described elsewhere [20].

For the current study, couples were recruited before 8^{+0} weeks of pregnancy. We selected pregnant women of at least 18 years of age and their male partners who were included in the study between November 2010 and October 2016 and were able to speak and write in the Dutch language. Couples were excluded when the pregnancy was conceived after sperm or oocyte donation, when ultrasound data was unreliable due to incomplete embryonic measurements or technical problems, or when nutritional data was missing or unreliable (i.e. total energy intake <500 kilocalories (kcal)/day for women and <800 kcal/day for men) [21] (Fig. 1).

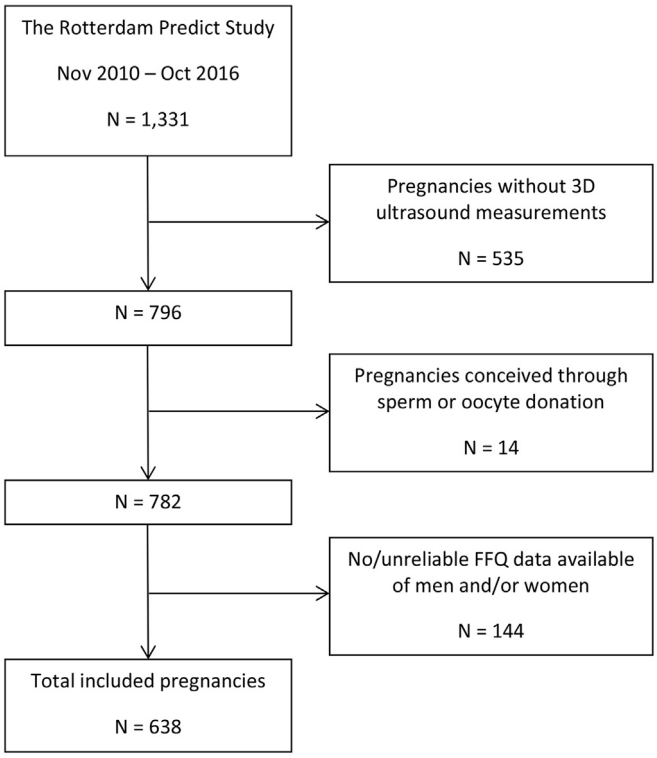

Fig. 1. Flowchart of the study population. FFQ = Food Frequency Questionnaire, IVF = in vitro fertilization, ICSI = intracytoplasmic sperm injection.

2.2. Pregnancy dating

For spontaneously conceived pregnancies, gestational age was determined by the first day of the last menstrual period (LMP) if there was a regular cycle (25–35 days). If the menstrual cycle was prolonged (32–35 days), gestational age was adjusted for the duration of the menstrual cycle. If the gestational age deviated more than six days from the measured CRL, or when the LMP was unknown, the first CRL measurement was used to determine gestational age. For pregnancies conceived through intrauterine insemination (IUI), gestational age was calculated using the LMP or insemination date plus 14 days. For pregnancies derived from in vitro fertilization (IVF) with or without intracytoplasmic sperm injection (ICSI), gestational age was determined on the day of oocyte retrieval plus 14 days. For pregnancies derived from cryo-embryo transfer, gestational age was calculated as the day of the embryo transfer plus 17 or 18 days [20,22].

2.3. Dietary assessment

Habitual food intake was assessed using a validated 166-item semi-quantitative food frequency questionnaire (FFQ), with a reference period of four weeks. This FFQ was designed to cover >96% of the absolute level of food intake and >95% of the between-

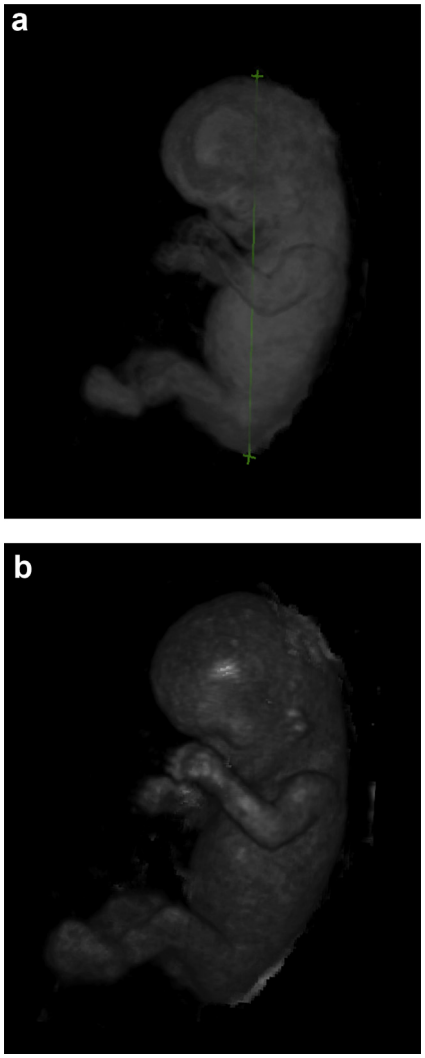

Fig. 2. a. 3D measurement of crown-rump length (CRL), as depicted by the green line, of an embryo of 9 weeks of gestational age in the Barco I-space (with permission). b. 3D measurement of embryonic volume (EV) of an embryo of 9 weeks of gestational age in the Barco I-space (with permission).

person variability of each nutrient under study assessed in the DNFCS from 1998 [23]. Participants answered questions relating to frequency by selecting answers ranging from 'never' to '6–7 days per week'. Portion sizes were estimated using natural portions and commonly used household measures. Average daily nutrient intakes were calculated by multiplying the consumption frequency by the portion size and nutrient content in grams as indicated in the most recent Dutch food composition table (2011) [24]. Intake levels for energy, macronutrients, dietary fiber, and selected vitamins were validated [25–27].

2.4. Embryonic growth

To determine embryonic growth, transvaginal 3D-US scans were performed in the 7th, 9th, and 11th week of gestation [4,28]. The ultrasound scans were executed by experienced researchers using a 6–12 MHz transvaginal probe and using GE Voluson E8 equipment and 4D View software (General Electrics Medical Systems, Zipf, Austria). Subsequently, the 3D-US scans were stored as Cartesian (rectangular) volumes and transferred to the BARCO I-Space (Barco N.V., Kortrijk, Belgium) at the Department of Bioinformatics, Erasmus University Medical Centre, Rotterdam. V-scope software was used to perform offline measurements of the CRL and EV. The CRL was measured three times in three dimensions, the average of these three measurements was used for analysis. The EV was measured with a semi-automated volume measuring application based on gray-scale differences.

2.5. General data

At enrolment all participating men and women filled out a general questionnaire covering details on age, geographical background, level of education, obstetric and medical history and periconceptional lifestyle behaviors (smoking, use of alcohol, folic acid supplement use) which was thoroughly checked by an experienced research nurse. At primary visit anthropometric measurements were conducted to obtain data on height and weight and blood pressure [20].

2.6. Statistical analysis

First, we compared the baseline characteristics between included and excluded participants to investigate whether our study sample was a representative reflection of the entire Predict cohort. These characteristics are reported as medians with interquartile ranges or as absolute numbers with percentages. Mann–Whitney U and Chi-square tests were used to compare respectively continuous variables or categorical data.

Principal component analysis (PCA) was used to derive paternal and maternal dietary patterns separately from the nutritional data obtained from the FFQ. First, the 166 food items were reduced to 24 predefined food groups based on their origin and nutrient content [29]. Thereafter, PCA was performed. High correlations between food groups were identified and factor loadings were calculated for each food group, indicating the extent to which each food group contributes to the specific dietary pattern [30]. Based on visual inspection of the scree plot, three dietary patterns were retrieved with eigenvalues >1.75). All participants received factor scores for each dietary pattern, which represented their adherence to the specific pattern.

Maternal dietary patterns were expected to show a stronger association with embryonic growth than paternal dietary patterns. Therefore, linear mixed model analyses were conducted to assess the additional independent effect of each paternal dietary pattern on embryonic growth in the presence of each maternal dietary pattern. An increase of one factor score (represented by the beta (β)) is equal to an increase of one standard deviation and can thus be interpreted as such. To start, a first analysis was performed, adjusting the maternal dietary factor scores for gestational age and maternal total energy intake *(model 1)*. Thereafter, we additionally adjusted for maternal BMI, maternal age, maternal smoking, nulliparous and fetal gender *(model 2)*, paternal BMI, paternal smoking *(model 3)*, and lastly paternal dietary factor scores and paternal total energy intake *(model 4)*. Subsequently, model 1 and model 2 were used to determine the association between maternal dietary patterns and embryonic growth. Model 3 and model 4 were used to determine the additional association between paternal dietary patterns and embryonic growth. Subsequently, model 3 and model 4 were compared using a likelihood ratio test.

As linear mixed model analyses require a normal distribution of the data, square root transformations of CRL data and third root transformations of EV data were performed. Analyses were performed in two subgroups; 1. strictly dated spontaneous pregnancies, defined as known first day of LMP and regular menstrual cycle, and 2. IVF/ICSI pregnancies.

P-values of $P \leq .05$ were considered significant. Data analyses within this project were performed using SPSS Statistics for Windows, Version 21.0 (IBM Corp. Armonk, NY) and R version 3.4.1 (The R foundation for Statistical Computing).

3. Results

3.1. Study population

A total of 1331 couples participated in the Predict study. After excluding pregnancies who did not match our inclusion criteria, 638 pregnancies were further analyzed. This included 94 pregnancies conceived through IVF, 104 from ICSI, and 440 spontaneously conceived, whether or not in combination with IUI (Fig. 1).

Paternal and maternal characteristics of included and excluded pregnancies are shown in Table 1. Median age of included men was 34 [31–38] years and they had a median BMI of 25.9 (23.8–28.3) kg/m^2. No significant differences were observed between included and excluded men. However, included women had a significantly lower BMI (24.4 vs. 24.7 kg/m^2; $P = .025$), were more often from Dutch origin (82.9 vs. 77.7%; $P = .027$) and were more likely to consume alcohol in the periconceptional period (34.3 vs. 27.3%; $P = .010$) compared to excluded women. In addition, included pregnancies were more often conceived after IVF/ICSI treatment (31.0 vs. 26.9%; $P < .001$) compared to excluded pregnancies.

The three paternal dietary patterns retrieved from the PCA, explained 27.5% of the total variance of the dietary intake. Paternal factor loadings for these dietary patterns are presented in Table 2. The first paternal dietary pattern explained 11.5% of the total variance and was labelled “Whole wheat grains and Vegetables”, reflecting high intakes of whole wheat grains, vegetables, and margarine and could therefore be labelled healthy. The second paternal dietary pattern explained 8.5% of the total variance and was labelled “Sauces and Snacks”, reflecting high intakes of alcohol, sauces, and snacks and could therefore be labelled unhealthy. The third paternal dietary pattern explained 7.4% of the total variance and was labelled “Refined Grains, Fish and legumes”, reflecting high intakes of refined grains, legumes, and fish.

Linear mixed model analyses neither showed an additional independent effect of paternal dietary patterns on embryonic growth in spontaneous pregnancies (strictly and not strictly dated) nor IVF/ICSI pregnancies (Table 3). Additionally, we compared the likelihood of model 4 with the likelihood of model 3 using the likelihood ratio test. No significant differences were found between these two models, for CRL as well as EV, in strictly dated spontaneous pregnancies (CRL: $P = .964$; EV: $P = .953$), not strictly dated spontaneous pregnancies (CRL: $P = .175$; EV: $P = .676$), and IVF/ICSI pregnancies (CRL: $P = .379$; EV: $P = .306$) *(data not shown)*.

The three maternal dietary patterns retrieved from the PCA, explained 27.2% of the total variance in dietary intake. Maternal factor loadings for these dietary patterns are presented in Table 2. The first maternal dietary pattern explained 11.0% of the total variance and was labelled “Nuts and Fruits”, reflecting high intakes of fish, fruits, legumes, and nuts. The second maternal dietary pattern explained 8.7% of the total variance and was labelled “Whole wheat Grains and Margarine”, reflecting high intakes of margarine, potatoes, and whole wheat grains. The third maternal

Table 1
Baseline characteristics of in- and excluded participants of the Rotterdam Periconception Cohort (Predict study).

	Paternal		Maternal	
	Included	Excluded	Included	Excluded
	(n = 638)	(n = 693)	(n = 638)	(n = 693)
Age (years)	34 (31–38)	34 (31–38)	32 (29–35)	32 (29–35)
missings	*104*	*247*	*51*	*55*
BMI (kg/m^2)	25.9 (23.8–28.3)	26.0 (23.8–28.3)	24.4 (21.9–28.0)*	24.7 (22.3–29.1)
missings	*77*	*213*	*46*	*53*
Geographic background				
Dutch	519 (83.7%)	350 (85.0%)	520 (82.9%)*	421 (77.7%)
Other Western	23 (3.7%)	10 (2.4%)	31 (4.9%)	25 (4.6%)
Non-Western	78 (12.6%)	52 (12.6%)	76 (12.2%)	96 (17.7%)
missings	*18*	*281*	*11*	*151*
Education level				
Low	84 (13.6%)	62 (15.3%)	51 (8.2%)	51 (9.5%)
Intermediate	228 (36.8%)	140 (34.6%)	211 (33.7%)	209 (38.8%)
High	307 (49.6%)	203 (50.1%)	364 (58.1%)	279 (51.7%)
missings	*19*	*288*	*12*	*154*
Smoking (yes)	186 (30.7%)	117 (30.0%)	98 (15.7%)	90 (17.1%)
missings	*32*	*303*	*14*	*168*
Alcohol (yes)	455 (75.2%)	280 (72.0%)	214 (34.3%)*	144 (27.3%)
missings	*33*	*304*	*15*	*166*
Folic acid supplement use (yes)	49 (8.0%)	28 (7.2%)	616 (98.2%)	518 (97.2%)
missings	*28*	*304*	*11*	*161*
Multivitamin use (yes)	116 (19.2%)	74 (19.2%)	432 (69.0%)	392 (73.7%)
missings	*33*	*308*	*12*	*161*
Nulliparous (yes)	NA	NA	283 (46.5%)	236 (44.9%)
missings			*29*	*167*
Fetal gender (male)	NA	NA	311 (52.9%)	240 (50.8%)
missings			*50*	*48*
Mode of conception				
Spontaneous	NA	NA	440 (69.0%)*	122 (65.6%)
IVF/ICSI			198 (31.0%)	50 (26.9%)
Sperm/Egg donation			0 (0.0%)	14 (7.5%)
missings			*0*	*507*

Note: Data are represented as median with interquartile range (IQR) or as number with percentage.
Abbreviations: BMI = Body Mass Index; IF = In vitro fertilization; ICSI = Intracytoplasmic sperm injection.
*$P \leq .05$.

Table 2
Factor loadings of 24 food groups within three paternal and three maternal dietary patterns identified through principle component analysis.

Food groups	Paternal dietary patterns			Maternal dietary patterns		
	Whole wheat Grains and Vegetables	Sauces and Snacks	Refined Grains, Fish and Legumes	Nuts and Fruits	Grains and Margarine	Solid fat and Sugars
Variance explained (%)	11.5	8.5	7.4	11.0	8.7	7.5
Alcohol	−0.042	**0.646**	−0.137	−0.001	−0.058	−0.024
Cereals	0.225	−0.133	0.296	0.108	−0.118	−0.090
Dairy	0.210	0.073	0.102	−0.028	0.207	0.101
Eggs	−0.167	0.126	0.120	0.308	−0.355	−0.097
Fat	−0.158	0.111	−0.033	−0.048	−0.097	−0.022
Fish	0.009	0.098	**0.610**	**0.562**	−0.083	−0.075
Fruits	**0.418**	−0.254	0.212	**0.589**	0.118	0.285
Grains (refined)	−0.188	0.222	**0.626**	−0.073	−0.357	0.083
Grains (whole wheat)	**0.739**	−0.012	−0.199	0.163	**0.768**	−0.021
Legumes	0.052	−0.146	**0.549**	**0.578**	−0.150	−0.093
Liquid fat	0.045	−0.023	−0.019	−0.166	0.142	0.040
Margarine	**0.529**	0.164	−0.284	−0.208	**0.676**	−0.078
Mayonnaise	−0.055	**0.548**	0.189	0.123	0.077	0.112
Met	0.151	0.366	−0.005	−0.021	0.222	0.004
Nuts	0.201	0.043	0.052	**0.649**	0.092	−0.027
Olive oil	**0.429**	0.231	0.228	0.049	0.071	0.109
Potatoes	0.324	0.113	−0.081	−0.045	**0.412**	0.388
Sauces	0.169	**0.594**	0.174	0.044	0.218	0.065
Snacks	0.044	**0.571**	−0.074	0.020	0.091	0.107
Solid fat	−0.045	−0.032	0.021	0.000	−0.114	**0.746**
Soup	0.127	0.208	−0.027	0.099	−0.020	0.093
Sugars	0.094	0.140	−0.157	−0.085	0.040	**0.738**
Sodas/Preserved Juices	−0.153	0.109	0.116	0.138	−0.034	**0.420**
Vegetables	**0.692**	0.041	0.216	0.367	0.278	0.302

Note: Factor loadings indicate the extent the food group is correlated to a specific dietary pattern and are presented as correlation coefficients. Factor loadings of significant relevance (>0.400 or < −0.400) are presented in bold.

Table 3
Effect estimates from the linear mixed model analysis for associations between paternal dietary patterns and embryonic crown-rump length (CRL) and embryonic volume (EV), stratified by strictly dated spontaneous pregnancies, not strictly dated spontaneous pregnancies, and IVF/ICSI pregnancies.

Paternal dietary patterns			Strictly dated spontaneous pregnancies	Not strictly dated spontaneous pregnancies	IVF/ICSI pregnancies
			(n=251)	*(n=139)*	*(n=171)*
Whole wheat Grains and Vegetables	CRL	β	−0.006	−0,048	−0.015
		95% CI	−0.069, 0.058	−0,095; −0,000	−0.061, 0.031
	EV	β	0.001	−0,015	−0.006
		95% CI	−0.022, 0.021	−0,036; 0,006	−0.025, 0.013
Sauces and Snacks	CRL	β	0.001	−0,015	0.006
		95% CI	−0.052, 0.052	−0,064; 0,034	−0.037, 0.050
	EV	β	−0.007	0,004	0.002
		95% CI	−0.025, 0.011	−0,017; 0,025	−0.016, 0.020
Refined Grains, Fish and Legumes	CRL	β	−0.005	−0,019	0.012
		95% CI	−0.051, 0.041	−0,060; 0,022	−0.030, 0.054
	EV	β	−0.003	−0,002	−0.010
		95% CI	−0.020, 0.014	−0,020; 0,017	−0.028, 0.008

Note: effect estimates (β) represent the amount of change in square root CRL ($\sqrt{mm}$) and third root EV ($\sqrt[3]{cm^3}$) per unit increase of the dietary factor score. For example for the 'Whole wheat grains and vegetables' dietary pattern; 1 unit increase in dietary factor score means an increase in CRL of $(-0.006)^2$ mm = 0.000036 mm.
In this table, only the fully adjusted model 4 is presented (adjusted for maternal and paternal dietary factor scores, maternal and paternal total energy intake, maternal en paternal BMI, maternal and paternal smoking, maternal age, nulliparous and fetal gender).
Abbreviations: IVF = in vitro fertilization, ICSI = intracytoplasmic sperm injection, CI = confidence interval.

dietary pattern explained 7.5% of the total variance and was labelled "Solid fat and Sugars", reflecting high intakes of sodas/preserved juices, sugars and solid fat.

Strong adherence to the "Nuts and Fruits" maternal dietary pattern was associated with a larger CRL of 0.0032 mm per unit of increase in factor score (β = 0.057, 95%CI 0.015; 0.099) (model 1) but this association was limited to strictly dated spontaneous pregnancies only (Table 4). After adjustment for maternal factors, linear mixed model analyses showed a significant decrease in EV when there is a high adherence to the "Solid fat and Sugars" maternal dietary pattern (β = −0.026, 95%CI −0.051;−0.001) (model 2) (Table 4). Transformation to the original scale showed that adherence to the "Solid fat and Sugars" dietary pattern was associated with a decrease in EV of 0.000018 cm^3 per unit of increase in factor score. This inverse association was limited to strictly dated spontaneous pregnancies. Furthermore, high adherence to the "Nuts and Fruits" or "Whole wheat Grains and Margarine" dietary pattern showed a trend toward an increase in EV, although not significant (model 2) (Table 4). For not strictly dated spontaneous pregnancies and IVF/ICSI pregnancies no significant associations were found between maternal dietary patterns and EV.

4. Discussion

In the present study, no significant associations were shown between adherence to periconceptional paternal dietary patterns and embryonic growth independent of maternal dietary patterns in spontaneous pregnancies or IVF/ICSI pregnancies. Although for

Table 4
Effect estimates from the linear mixed model analysis for associations between maternal dietary patterns and embryonic crown-rump length (CRL) and embryonic volume (EV), stratified by strictly dated spontaneous pregnancies, not strictly dated spontaneous pregnancies, and IVF/ICSI pregnancies.

Maternal dietary patterns			Model 1			Model 2			Model 3			Model 4		
			Strictly dated	Not strictly dated	IVF/ICSI	Strictly dated	Not strictly dated	IVF/ICSI	Strictly dated	Not strictly dated	IVF/ICSI	Strictly dated	Not strictly dated	IVF/ICSI
			(n = 280)	*(n = 160)*	*(n = 198)*	*(n = 257)*	*(n = 147)*	*(n = 177)*	*(n = 251)*	*(n = 139)*	*(n = 171)*	*(n = 251)*	*(n = 139)*	*(n = 171)*
Nuts and Fruits	CRL	β	0.057*	−0.008	0.005	0.038	−0.010	−0.002	0.039	−0.009	−0.001	0.040	−0.000	−0.003
		95% CI	0.015, 0.099	−0.045, 0.030	−0.025, 0.035	−0.012, 0.087	−0.057, 0.036	−0.040, 0.035	−0.011, 0.088	−0.057, 0.038	−0.039, 0.037	−0.010, 0.090	−0.049, 0.048	−0.043, 0.037
	EV	β	0.014	0.008	−0.003	0.001	−0.006	−0.006	0.002	−0.006	−0.006	0.002	−0.003	−0.002
		95% CI	−0.002, 0.030	−0.025, 0.010	−0.015, 0.010	−0.017, 0.018	−0.026, 0.015	−0.021, 0.009	−0.015, 0.019	−0.027, 0.015	−0.021, 0.009	−0.016, 0.019	−0.024, 0.018	−0.019, 0.014
Whole wheat Grains and Margarine	CRL	β	0.022	0.004	−0.009	0.033	0.010	−0.012	0.032	0.011	−0.011	0.032	0.024	0.004
		95% CI	−0.021, 0.064	−0.032, 0.041	−0.041, 0.024	−0.015, 0.080	−0.034, 0.054	−0.049, 0.026	−0.045, 0.079	−0.033, 0.055	−0.049, 0.026	−0.026, 0.090	−0.026, 0.075	−0.038, 0.046
	EV	β	0.005	−0.016	−0.001	0.004	−0.015	−0.005	0.003	−0.014	−0.005	0.0019	−0.008	−0.001
		95% CI	−0.012, 0.021	−0.032, 0.000	−0.013, 0.012	−0.013, 0.020	−0.034, 0.005	−0.019, 0.009	−0.013, 0.020	−0.034, 0.001	−0.019, 0.009	−0.019, 0.023	−0.031, 0.014	−0.017, 0.015
Solid fat and Sugars	CRL	β	−0.012	−0.012	−0.027	−0.022	−0.009	−0.044	−0.022	−0.010	−0.038	−0.026	−0.007	−0.029
		95% CI	−0.075, 0.050	−0.067, 0.042	−0.058, 0.005	−0.089, 0.045	−0.074, 0.059	−0.089, 0.002	−0.089, 0.045	−0.075, 0.055	−0.084, 0.009	−0.094, 0.043	−0.074, 0.060	−0.078, 0.020
	EV	β	−0.021	−0.013	0.002	−0.026*	−0.011	0.01	−0.023	−0.005	0.01	−0.025	−0.002	0.007
		95% CI	−0.046, 0.0034	−0.038, 0.012	−0.012, 0.015	−0.051, −0.001	−0.040, 0.018	−0.008, 0.028	−0.048, 0.001	−0.034, 0.025	−0.008, 0.028	−0.050, 0.001	−0.032, 0.028	−0.012, 0.027

Note: effect estimates (β) represent the amount of change in square root CRL ($\sqrt{mm}$) and third root EV ($\sqrt[3]{cm^3}$) per unit increase of the dietary factor score. For example for the 'Nuts and Fruits' dietary pattern; 1 unit increase in dietary factor score means an increase in CRL of (0.057)2 mm = 0032 mm *P ≤ .05.
Abbreviations: IVF = in vitro fertilization, ICSI = intracytoplasmic sperm injection, CI = confidence interval.
Model 1; adjusted for maternal dietary factor scores, total energy intake, and gestational age.
Model 2; model 1 and additionally adjusted for maternal BMI, smoking, age, nulliparous and fetal gender.
Model 3; model 2 and additionally adjusted for paternal BMI and smoking.
Model 4; model 3 and additionally adjusted for paternal dietary factor scores and total energy intake.

maternal dietary patterns, a significantly inverse association between the periconceptional maternal "Solid fat and Sugars" dietary pattern and embryonic volume was found in strictly dated spontaneous pregnancies only, the association was weak and only shown in model 2.

The absence of the association in the not strictly dated group pregnancy can be explained by the fact that dating is performed by using CRL thereby ignoring all (patho)physiological variation in embryonic growth. The most likely explanation supported by the likely hood test between Model 3 and 4 that no independent associations were found between paternal dietary patterns and embryonic growth is that the maternal dietary pattern and other unmeasured factors predominate the potential impact of the paternal dietary pattern.

The contributions of the paternal diet to embryonic growth and offspring health have mostly been investigated in animal studies, providing evidence for a transgenerational impact by showing that offspring of male mice fed a low protein diet or a folate deficient diet had compromised cardiovascular and metabolic functioning in later life [14,31]. Although animal studies provide valuable insights in the mechanism underlying paternal inheritance, these studies can often not be extrapolated to humans.

Several other paternal characteristics however have been associated with embryonic growth and birth outcomes in humans. Van Uitert et al. found a positive association between paternal birth weight and embryonic growth in the first trimester of pregnancy among 81 Dutch men participating in the Rotterdam Periconception Cohort between 2009 and 2010. One point increase in paternal birthweight Z-score was associated with an increased CRL of 0.0019 $\sqrt{}$mm/day [5]. Furthermore, paternal obesity has shown to affect fecundability and birth outcomes. Ramlau-Hansen et al. showed that overweight and obese men had an increased risk of subfecundity, defined as waiting time to pregnancy of more than 12 months, compared to normal weight men (OR 1.15 and 1.49, for overweight and obese respectively) [32]. Additionally, in 305 couples undergoing assisted reproductive technology (ART) paternal obesity was associated with a decreased chance of clinical pregnancy (25.8% obese vs. 42.9% normal weight) and live birth rates (22.6% obese vs. 41.3% normal weight) [33]. The effects of paternal smoking have been studied by Morales-Suárez-Varela and colleagues [34]. A total of 87,930 pregnancies were included from the population-based Danish National Birth Cohort. Paternal smoking was associated with a 10% higher risk of fetal deaths and a 46% higher risk of still births compared to non-smoking fathers.

Although we found no significant association between paternal dietary patterns and embryonic growth, there are many indications that paternal nutrition affects semen quality and fecundability [10]. In the reviewed studies, high intakes of fish, shellfish, seafood, poultry, cereals, vegetables, fruits and low-fat dairy products were positively associated with semen quality. High intakes of red meat, processed meat, caffeine and tea were inversely associated with fecundability, including rates of fertilization, pregnancy, or miscarriage. Besides, Vujkovic et al. observed that couples ($n = 161$) undergoing IVF/ICSI treatment with a higher adherence to the "Mediterranean" dietary pattern had a 40% increased probability of pregnancy [35]. However, as paternal adherence to the "Mediterranean" diet and IVF/ICSI outcomes were not studied in absence of maternal adherence to the "Mediterranean" diet, no conclusions could be drawn about the single impact of paternal adherence to the "Mediterranean" diet and IVF/ICSI outcomes.

Although paternal dietary patterns have not been examined in relation to embryonic growth yet, Parisi et al. showed that maternal adherence to a 'high fish, and olive oil, low meat' dietary pattern has been associated with increased embryonic CRL and EV per unit of increase in factor score [22]. Likewise, our results indicated an inverse association between high adherence to the maternal "unhealthy" dietary pattern and EV. These results suggest that the intake of healthy food groups, containing essential nutrients, is positively associated with embryonic growth, whereas the intake of unhealthy food groups, lacking essential nutrients, is inversely associated with embryonic growth. In conjunction, these associations are not only reflected in embryonic growth, but also in offspring health. A Dutch case-control family study showed that maternal high intakes of fish and seafood were associated with a 70% reduced risk of congenital heart diseases in offspring [6].

This study does not show significant associations, however, because lifestyle behaviours of couples strongly correlate, it is important for clinical care that those who are contemplating pregnancy are aware and encouraged to adopt healthy behaviours. This is a responsibility of these women and men as well as for health professionals.

An important strength of our study is its longitudinal prospective design, in particular addressing the periconceptional period, the large number of included men and women, and the use of comprehensive questionnaires with detailed information about the participants. Moreover, the validated FFQ measured participant's habitual food intake of the previous four weeks, minimizing day to day variation in food intake, and the derived dietary patterns reflect the overall nutritional intake of participants covering a wide range of nutritional factors instead of investigating single nutrients. Finally, the longitudinal 3D ultrasound scans were analyzed using the BARCO I-Space and V-scope software, providing 3D holograms of the embryos. Consequently, embryonic growth could be measured with high accuracy and precision [17]. The CRL was measured three times, of which the inter- and intra-observer agreement had been shown to be very high [18,36]. Furthermore, the 3D imaging technique enabled us to measure embryonic volumes and use it as a second parameter of embryonic growth. Embryonic volume has been proposed to be a predictive measure of embryonic growth restrictions [36,37].

Some limitations of the study have to be addressed as well. The cohort is embedded in a tertiary hospital, which limits external validity of our study because of the higher proportion of high risk pregnancies compared to the general population. In addition, a large proportion of the pregnancies were conceived through IVF/ICSI, compared to the general Dutch population (31.0% vs. 2.5%) [38]. Although, the growth trajectories of embryos conceived through IVF/ICSI pregnancies are comparable to embryos conceived spontaneously, analyses were performed separately for spontaneously conceived pregnancies and IVF/ICSI pregnancies to minimize confounding by mode of conception [28]. Embryonic growth is highly dependent on gestational age and an incorrect estimation of gestational age could have attenuated the potential association between paternal dietary patterns and embryonic growth. However, the differentiation of spontaneous pregnancies in strictly dated and not strictly dated spontaneous pregnancies reduces confounding by gestational age. A total of 535 pregnancies was excluded because of missing CRL and EV measurements, which was mainly due to not fully visualized embryos at the beginning of the study. As a result, the CRL and EV of these embryos could not be measured. It was not known whether larger embryos were more often missed than smaller embryos. If this was the case, it may have diluted potential positive associations between paternal dietary patterns and embryonic growth. Since the FFQ was self-administered and filled out in retrospect, it is susceptible for recall bias. Furthermore, the FFQ was only validated for women at reproductive age, not for men. There are some indications that bias related to social desirability and social approval differ for men and women, possibly leading to misclassification of dietary exposure [39,40]. However, other studies observed that fathers and mothers

have qualitatively comparable dietary patterns [41,42]. By using a semi-quantitative FFQ, differences in portion size between men and women were taken into account. Finally, although we have adjusted for many covariates in our analyses, residual confounding cannot be excluded.

5. Conclusion

In this study no significant associations between adherence to periconceptional paternal dietary patterns and embryonic growth independent of the maternal dietary patterns could be shown. Previous studies however revealed associations between paternal nutrition and semen quality, fecundability, and offspring health, which can be explained by preconceptional epigenetic influences of the paternal dietary pattern on male gametes and subsequent transmission to the embryo. Therefore, we emphasize that more research, should be focused in particular on the influence of paternal dietary patterns on prenatal growth and pregnancy outcome. This will enhance the awareness of the importance of healthy nutrition of couples contemplating pregnancy and health care professionals beyond pregnancy as an investment for health of current and future generations.

Statement of authorship

RST conceived, designed the study, contributed to all versions of the manuscript and supervised all aspects of the study. IdV, EO and AH conducted the statistical analyses. IdV, EO and AH drafted the first version of the manuscript. SW supported with the statistical analyses. All authors contributed to the critical revision of the manuscript and approved the final version.

Conflict of interest

All authors have completed and submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. No disclosures were reported.

Funding sources

This study was funded by the Department of Obstetrics and Gynecology of the Erasmus University Medical Center, Rotterdam, the Netherlands and an additional grant from ZonMW; the Netherlands organization for health research and development (project number 209040003).

Acknowledgements

The authors would like to thank all the patients for participating in this study, and the research team of the Rotterdam Periconceptional Cohort (Predict Study) for their contribution to the data collection.

References

[1] WHO. Obesity and overweight. 2017 [updated October 2017. Available from: www.who.int/mediacentre/factsheets/fs311/en.
[2] Steegers-Theunissen RP, Twigt J, Pestinger V, Sinclair KD. The periconceptional period, reproduction and long-term health of offspring: the importance of one-carbon metabolism. Hum Reprod Update 2013;19(6):640–55.
[3] Mook-Kanamori DO, Steegers EA, Eilers PH, Raat H, Hofman A, Jaddoe VW. Risk factors and outcomes associated with first-trimester fetal growth restriction. JAMA 2010;303(6):527–34.
[4] van Uitert EM, van der Elst-Otte N, Wilbers JJ, Exalto N, Willemsen SP, Eilers PH, et al. Periconception maternal characteristics and embryonic growth trajectories: the Rotterdam Predict study. Hum Reprod 2013;28(12):3188–96.
[5] EM van Uitert ES, GJ Bonsel, GJJM Borsboom, AHJ Koning, JSE Laven, N Exalto, RPM Steegers-Theunissen. Human embryonic growth trajectories: does the father matter? The Rotterdam Predict study.
[6] Obermann-Borst SA, Vujkovic M, de Vries JH, Wildhagen MF, Looman CW, de Jonge R, et al. A maternal dietary pattern characterised by fish and seafood in association with the risk of congenital heart defects in the offspring. BJOG 2011;118(10):1205–15.
[7] Vujkovic M, Ocke MC, van der Spek PJ, Yazdanpanah N, Steegers EA, Steegers-Theunissen RP. Maternal Western dietary patterns and the risk of developing a cleft lip with or without a cleft palate. Obstet Gynecol 2007;110(2 Pt 1):378–84.
[8] Vujkovic M, Steegers EA, Looman CW, Ocke MC, van der Spek PJ, Steegers-Theunissen RP. The maternal Mediterranean dietary pattern is associated with a reduced risk of spina bifida in the offspring. BJOG 2009;116(3):408–15.
[9] Gaskins AJ, Chavarro JE. Diet and fertility: a review. Am J Obstet Gynecol 2018 Apr;218(4):379–89.
[10] Salas-Huetos A, Bullo M, Salas-Salvado J. Dietary patterns, foods and nutrients in male fertility parameters and fecundability: a systematic review of observational studies. Hum Reprod Update 2017;23(4):371–89.
[11] Bukowski R, Smith GC, Malone FD, Ball RH, Nyberg DA, Comstock CH, et al. Fetal growth in early pregnancy and risk of delivering low birth weight infant: prospective cohort study. BMJ 2007;334(7598):836.
[12] Leunissen RW, Kerkhof GF, Stijnen T, Hokken-Koelega A. Timing and tempo of first-year rapid growth in relation to cardiovascular and metabolic risk profile in early adulthood. JAMA 2009;301(21):2234–42.
[13] Watkins AJ, Sirovica S, Stokes B, Isaacs M, Addison O, Martin RA. Paternal low protein diet programs preimplantation embryo gene expression, fetal growth and skeletal development in mice. Biochim Biophys Acta 2017;1863(6):1371–81.
[14] Lambrot R, Xu C, Saint-Phar S, Chountalos G, Cohen T, Paquet M, et al. Low paternal dietary folate alters the mouse sperm epigenome and is associated with negative pregnancy outcomes. Nat Commun 2013;4:2889.
[15] Sinclair KD, Watkins AJ. Parental diet, pregnancy outcomes and offspring health: metabolic determinants in developing oocytes and embryos. Reprod Fertil Dev 2013;26(1):99–114.
[16] Lane M, Robker RL, Robertson SA. Parenting from before conception. Science 2014;345(6198):756–60.
[17] Verwoerd-Dikkeboom CM, Koning AH, Hop WC, Rousian M, Van Der Spek PJ, Exalto N, et al. Reliability of three-dimensional sonographic measurements in early pregnancy using virtual reality. Ultrasound Obstet Gynecol 2008;32(7):910–6.
[18] Rousian M, Koning AH, van Oppenraaij RH, Hop WC, Verwoerd-Dikkeboom CM, van der Spek PJ, et al. An innovative virtual reality technique for automated human embryonic volume measurements. Hum Reprod 2010;25(9):2210–6.
[19] Verwoerd-Dikkeboom CM, Koning AH, Hop WC, van der Spek PJ, Exalto N, Steegers EA. Innovative virtual reality measurements for embryonic growth and development. Hum Reprod 2010;25(6):1404–10.
[20] Steegers-Theunissen RP, Verheijden-Paulissen JJ, van Uitert EM, Wildhagen MF, Exalto N, Koning AH, et al. Cohort profile: the Rotterdam periconceptional cohort (Predict study). Int J Epidemiol 2016;45(2):374–81.
[21] Willet WC. Nutritional epidemiology. 3rd ed. Oxford University Press; 2013.
[22] Parisi F, Rousian M, Huijgen NA, Koning AH, Willemsen SP, de Vries JH, et al. Periconceptional maternal 'high fish and olive oil, low meat' dietary pattern is associated with increased embryonic growth: the Rotterdam Periconceptional Cohort (Predict Study). Ultrasound Obstet Gynecol 2017 Dec;50(6):709–16.
[23] Centre TDN. Zo eet Nederland: Resultaten van de Voedselconsumptiepeiling 1997–1998 (Results of the Dutch food consumption survey 1997–1998). Den Haag, the Netherlands: Voedingscentrum; 1998.
[24] (RIVM) TDNIfPHatE. NEVO-tabel; Nederlands Voedingsstoffenbestand 2011. Den Haag: Voedingscentrum; 2011.
[25] Feunekes GI, Van Staveren WA, De Vries JH, Burema J, Hautvast JG. Relative and biomarker-based validity of a food-frequency questionnaire estimating intake of fats and cholesterol. Am J Clin Nutr 1993;58(4):489–96.
[26] Siebelink E, Geelen A, de Vries JH. Self-reported energy intake by FFQ compared with actual energy intake to maintain body weight in 516 adults. Br J Nutr 2011;106(2):274–81.
[27] Streppel MT, de Vries JH, Meijboom S, Beekman M, de Craen AJ, Slagboom PE, et al. Relative validity of the food frequency questionnaire used to assess dietary intake in the Leiden Longevity Study. Nutr J 2013;12:75.
[28] van Uitert EM, Exalto N, Burton GJ, Willemsen SP, Koning AH, Eilers PH, et al. Human embryonic growth trajectories and associations with fetal growth and birthweight. Hum Reprod 2013;28(7):1753–61.
[29] Slimani N, Fahey M, Welch AA, Wirfalt E, Stripp C, Bergstrom E, et al. Diversity of dietary patterns observed in the European Prospective Investigation into Cancer and Nutrition (EPIC) project. Publ Health Nutr 2002;5(6B):1311–28.
[30] Hu FB. Dietary pattern analysis: a new direction in nutritional epidemiology. Curr Opin Lipidol 2002;13(1):3–9.
[31] Watkins AJ, Sinclair KD. Paternal low protein diet affects adult offspring cardiovascular and metabolic function in mice. Am J Physiol Heart Circ Physiol 2014;306(10):H1444–52.
[32] Ramlau-Hansen CH, Thulstrup AM, Nohr EA, Bonde JP, Sorensen TI, Olsen J. Subfecundity in overweight and obese couples. Hum Reprod 2007;22(6):1634–7.
[33] Bakos HW, Henshaw RC, Mitchell M, Lane M. Paternal body mass index is associated with decreased blastocyst development and reduced live birth rates following assisted reproductive technology. Fertil Steril 2011;95(5):1700–4.
[34] Morales-Suarez-Varela M, Nohr EA, Bech BH, Wu C, Olsen J. Smoking, physical exercise, BMI and late foetal death: a study within the Danish National Birth Cohort. Eur J Epidemiol 2016;31(10):999–1009.

[35] Vujkovic M, de Vries JH, Lindemans J, Macklon NS, van der Spek PJ, Steegers EA, et al. The preconception Mediterranean dietary pattern in couples undergoing in vitro fertilization/intracytoplasmic sperm injection treatment increases the chance of pregnancy. Fertil Steril 2010;94(6):2096–101.
[36] Rousian M, Hop WC, Koning AH, van der Spek PJ, Exalto N, Steegers EA. First trimester brain ventricle fluid and embryonic volumes measured by three-dimensional ultrasound with the use of I-Space virtual reality. Hum Reprod 2013;28(5):1181–9.
[37] Baken L, Benoit B, Koning AHJ, van der Spek PJ, Steegers EAP, Exalto N. First-trimester crown-rump length and embryonic volume of fetuses with structural congenital abnormalities measured in virtual reality: an observational study. BioMed Res Int 2017;2017:1953076.
[38] (NVOG) DSoOaG. Landelijke IVF cijfers 2015 2017 [Available from: http://www.nvog.nl/Sites/Files/0000005105_IVFlandelijk2015.pdf.
[39] Hebert JR, Ma Y, Clemow L, Ockene IS, Saperia G, Stanek 3rd EJ, et al. Gender differences in social desirability and social approval bias in dietary self-report. Am J Epidemiol 1997;146(12):1046–55.
[40] Lee H, Kang M, Song WO, Shim JE, Paik HY. Gender analysis in the development and validation of FFQ: a systematic review. Br J Nutr 2016;115(4):666–71.
[41] Northstone K, Emmett PM. Dietary patterns of men in ALSPAC: associations with socio-demographic and lifestyle characteristics, nutrient intake and comparison with women's dietary patterns. Eur J Clin Nutr 2010;64(9):978–86.
[42] Lioret S, McNaughton SA, Crawford D, Spence AC, Hesketh K, Campbell KJ. Parents' dietary patterns are significantly correlated: findings from the melbourne infant feeding activity and nutrition trial program. Br J Nutr 2012;108(3):518–26.