

Supplementary File

Study Outcomes

Supplemental Table 1: Comparison of current study cohort to remainder of ALSPAC cohort

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Study Outcomes

We used the ALSPAC pre-school development assessments completed by the mothers between 6 and 42 months which provided scores for four domains, i.e. fine motor, gross motor, social development, and communication; items for this test were developed by ALSPAC based on those listed by Frankenburg¹.

The Strengths and Difficulties Questionnaire (SDQ)² was completed by the mothers when the child was 81 months old. Published cut-offs², as used previously in ALSPAC research³ were used to define suboptimal development. The cut-off and percentage in the suboptimal group for the subscales are: pro-social behaviour (≤ 5 ; 9.8%), peer problems (≥ 3 ; 13.5%), hyperactivity (≥ 6 ; 18.7%), emotional symptoms (≥ 4 ; 12.2%) and conduct problems (≥ 3 ; 24.3%) together with a total behavioural difficulties score (≥ 14 ; 10.5%).

Child IQ was assessed at age 8 years by trained psychologists, with an abbreviated form of the Wechsler Intelligence Scale III for Children (WISC-III). The verbal, performance and total IQ scores were calculated using standard procedures, as previously described⁴.

At age 9 years, the children were asked to read 10 real words, with the sum of total correct responses comprising their "Reading Score"⁵. In addition by reading pieces of prose the reading speed, accuracy, and comprehension were assessed by trained psychologists, using the revised Neale Analysis of Reading Ability (NARA II)⁶.

As described in the online ALSPAC data dictionary, (<http://www.bristol.ac.uk/alspac/researchers/access/>) (Child Focussed Questionnaire F08 and F09; Built files) every effort was made to ensure inter-rater reliability. For WISC, Dr Clare Bell (who is experienced in psychometric testing) managed and observed the team of testers to ensure inter-rater compatibility, including how the tests were being administered and scored (See Child Focussed Questionnaire F08). For the reading tests staff were trained in order to reduce inter-observer variability, with regular quality control measurements and strict protocols for recording reading scores (See Child Focussed Questionnaire F09).

Supplemental Table 1: Comparison of current study cohort to remainder of ALSPAC cohort

Confounder	This study cohort			Remaining ALSPAC cohort†			p value‡
	n	Mean	SD	n	Mean	SD	
BMI (kg/m²)	6221	22.9	3.8	5016	23.0	3.9	0.41
Maternal age (years)	7065	28.3	4.8	6551	27.7	5.2	<0.0001
Breastfeeding (% yes)	5264	78.7		4036	75.6		<0.0001
Crowding (% 1+ person/room)	404	6.0		454	7.9		<0.0001
Ethnicity (% white)	6515	97.9		5185	96.7		0.002
Gender (% male)	3687	52.2		3336	50.9		0.14
	n	%		n	%		p value§
Housing							
Owned/mortgaged (%)	5192	76.3		4116	69.8		<0.0001
Other rented (%)	410	6.0		507	8.6		
Council rented (%)	1199	17.6		1278	21.7		
Iodine status							
Sufficient	227	28.8		115	32.4		0.23
Deficient	560	71.7		240	67.6		
Maternal Education							
Low (%)	1911	28.6		1724	31.8		<0.0001
Medium (%)	2369	35.5		1815	33.5		
High (%)	2398	35.9		1884	34.7		
Oily fish consumption							
Never/rarely	2756	42.0		2242	42.7		0.43
Once a fortnight or more	3808	58.0		3007	57.3		
Parity							
Zero (%)	3039	45.1		2620	44.7		0.61
One or more (%)	3695	54.9		3245	55.3		
Social class							
Manual (%)	1047	19.0		900	20.9		0.02
Non-manual (%)	4455	81.0		3401	79.1		
Smoking in pregnancy							
Non-smoker (%)	5219	76.3		4407	73.6		<0.0001
Smoking at 18 weeks (%)	1621	23.7		1577	26.4		

n: number of subjects † Remaining ALSPAC cohort defined as singletons from women who enrolled in the core ALSPAC sample with children surviving to 1 year (n13616); ‡ p value from independent t-test; § p value for χ^2 test.

Supplemental Table 2: Odds of suboptimum outcomes according to maternal vitamin D status (< 50.0 vs ≥ 50.0 nmol/L): sensitivity analysis by adjusting for additional confounders

		Model 3 with causal pathway†				Model 3 with iodine ‡			
		Age	OR (95% CI)	p value	n	OR (95% CI)	p value	n	
ALSPAC early development tests	Gross Motor Skills	6 mo§	0.94 (0.80, 1.12)	0.49	4329	0.80 (0.51, 1.25)	0.33	627	
		18 mo	1.16 (1.00, 1.35)	0.05	4336	1.33 (0.90, 1.96)	0.15	635	
		30 mo	1.18 (1.01, 1.39)	0.04	4084	1.47 (0.98, 2.20)	0.06	620	
		42 mo	1.08 (0.91, 1.27)	0.39	4022	1.20 (0.79, 1.80)	0.39	621	
	Fine Motor Skills	6 mo§	1.05 (0.90, 1.23)	0.52	4092	0.76 (0.47, 1.21)	0.25	597	
		18 mo	1.02 (0.88, 1.17)	0.84	4336	1.50 (1.02, 2.23)	0.04	636	
		30 mo	1.20 (1.03, 1.40)	0.02	4089	1.61 (1.06, 2.46)	0.02	631	
		42 mo	1.10 (0.94, 1.29)	0.26	4023	1.20 (0.78, 1.86)	0.41	620	
	Social Development	6 mo§	1.01 (0.85, 1.20)	0.90	4157	0.85 (0.54, 1.33)	0.47	598	
		18 mo	1.15 (0.97, 1.35)	0.10	4334	0.98 (0.63, 1.52)	0.92	636	
		30 mo	1.07 (0.91, 1.27)	0.43	4078	1.19 (0.79, 1.81)	0.41	620	
		42 mo	1.18 (0.99, 1.39)	0.06	4018	1.21 (0.78, 1.89)	0.39	619	
Communication	6 mo§	0.99 (0.81, 1.20)	0.89	4235	0.90 (0.53, 1.55)	0.71	619		
	18 mo	1.12 (0.95, 1.31)	0.17	4341	0.73 (0.47, 1.13)	0.16	636		
Behaviour	Prosocial	7 yr	1.00 (0.77, 1.31)	0.97	3466	1.10 (0.57, 2.11)	0.77	580	
	Peer problems	7 yr	1.04 (0.83, 1.31)	0.74	3463	1.07 (0.59, 1.94)	0.83	581	
	Hyperactivity	7 yr	1.02 (0.84, 1.25)	0.84	3466	1.54 (0.91, 2.58)	0.11	582	
	Emotional	7 yr	1.21 (0.96, 1.53)	0.10	3464	1.42 (0.80, 2.50)	0.23	581	
	Conduct	7 yr	1.06 (0.89, 1.28)	0.52	3467	1.27 (0.82, 1.97)	0.29	581	
	Total Score	7 yr	1.24 (0.96, 1.60)	0.10	3463	1.61 (0.83, 3.11)	0.16	581	
Cognition	Verbal IQ	8 yr	0.99 (0.81, 1.22)	0.95	2917	0.97 (0.60, 1.58)	0.91	625	
	Performance IQ	8 yr	0.99 (0.81, 1.21)	0.93	2910	1.05 (0.66, 1.69)	0.83	625	
	Total IQ	8 yr	0.99 (0.80, 1.21)	0.90	2903	0.81 (0.50, 1.32)	0.40	625	
Reading ability	Words per min	9 yr	1.17 (0.95, 1.43)	0.15	2726	1.52 (0.96, 2.41)	0.07	597	
	Accuracy	9 yr	1.04 (0.84, 1.29)	0.73	2730	1.24 (0.75, 2.04)	0.41	597	
	Comprehension	9 yr	1.04 (0.84, 1.29)	0.73	2730	1.15 (0.69, 1.93)	0.59	597	
	Reading Score	9 yr	1.02 (0.84, 1.24)	0.81	2988	1.34 (0.85, 2.09)	0.21	604	

mo, month; OR, odds ratio; n, number of subjects; yr, years. Suboptimal outcome defined as scores in the bottom quartile for ALSPAC pre-school development assessments, cognition, and reading ability. Published cut-offs² were used for behaviour: Prosocial (≤5; 9.8%), Peer problems (≥3; 13.5%), hyperactivity (≥6; 18.7%), emotional symptoms (≥4; 12.2%), conduct problems (≥3; 24.3%), and total score (≥14; 10.5%). †Model 3 plus birthweight (<2500 or ≥2500 g) and preterm birth (<37 weeks or ≥37 weeks). ‡Model 3 plus suboptimal iodine-to-creatinine ratio in the first trimester [sufficient (≥150 µg/g) or deficient (<150 µg/g)]. As just 787 women also had measure of iodine, we used a simplified model to ensure that the model would converge; we excluded ethnicity and crowding. § Age of child at development test included in all models.

Supplemental Table 3: Odds of suboptimum outcomes in children according to maternal vitamin D status (< 25.0 vs ≥ 25.0 nmol/L), minimally adjusted and fully adjusted for potential confounders

		Model1†				Model2‡			Model3§			
		Age	OR (95% CI)	p value	n	OR (95% CI)	p value	n	OR (95% CI)	p value	n	
ALSPAC early development tests	Gross Motor Skills	6 mo ‖	1.22(0.91, 1.63)	0.18	6242	1.42 (1.00, 2.03)	0.05	4383	1.35 (0.94, 1.93)	0.10	4380	
		18 mo	1.04 (0.80, 1.36)	0.76	6269	1.25 (0.90, 1.74)	0.18	4385	1.32 (0.95, 1.84)	0.10	4383	
		30 mo	1.31 (0.99, 1.73)	0.06	5843	1.39 (0.99, 1.96)	0.06	4135	1.43 (1.01, 2.02)	0.05	4133	
		42 mo	1.02 (0.75, 1.39)	0.89	5695	1.16 (0.80, 1.68)	0.44	4073	1.20 (0.83, 1.75)	0.34	4070	
	Fine Motor Skills	6 mo ‖	0.97 (0.73, 1.30)	0.84	5880	1.24 (0.87, 1.79)	0.24	4141	1.22 (0.84, 1.76)	0.29	4139	
		18 mo	0.91 (0.69, 1.20)	0.50	6268	0.97 (0.69, 1.37)	0.86	4383	0.99 (0.70, 1.40)	0.94	4381	
		30 mo	1.13 (0.85, 1.50)	0.40	5854	1.21 (0.85, 1.73)	0.28	4138	1.19 (0.84, 1.71)	0.33	4136	
		42 mo	1.32 (0.99, 1.76)	0.06	5692	1.21 (0.83, 1.76)	0.31	4071	1.23 (0.84, 1.80)	0.28	4068	
	Social Development	6 mo ‖	1.01 (0.75, 1.37)	0.95	6010	1.06 (0.74, 1.54)	0.74	4209	1.02 (0.70, 1.49)	0.91	4207	
		18 mo	1.16 (0.88, 1.54)	0.29	6268	1.21 (0.85, 1.73)	0.30	4383	1.22 (0.85, 1.75)	0.29	4381	
		30 mo	0.98 (0.72, 1.32)	0.88	5843	0.93 (0.63, 1.38)	0.73	4129	0.88 (0.60, 1.30)	0.51	4127	
		42 mo	1.37 (1.03, 1.82)	0.03	5689	1.41 (0.98, 2.03)	0.07	4069	1.40 (0.97, 2.02)	0.08	4066	
	Communication	6 mo ‖	1.06 (0.75, 1.50)	0.73	6100	1.46 (0.98, 2.19)	0.06	4285	1.44 (0.96, 2.17)	0.08	4283	
		18 mo	1.11 (0.85, 1.48)	0.43	6279	1.29 (0.91, 1.82)	0.15	4390	1.26 (0.89, 1.79)	0.19	4388	
	Behaviour	Prosocial	7 yr	1.11 (0.69, 1.78)	0.68	4791	1.07 (0.58, 1.98)	0.83	3513	1.11 (0.68, 2.08)	0.73	3511
		Peer problems	7 yr	1.04 (0.68, 1.59)	0.84	4785	0.96 (0.56, 1.63)	0.88	3510	0.94 (0.55, 1.62)	0.84	3508
Hyperactivity		7 yr	0.70 (0.46, 1.06)	0.10	4780	0.62 (0.36, 1.05)	0.08	3513	0.61 (0.36, 1.04)	0.07	3511	
Emotional		7 yr	0.85 (0.53, 1.36)	0.50	4785	0.72 (0.39, 1.32)	0.29	3511	0.73 (0.39, 1.35)	0.31	3509	
Conduct		7 yr	0.93 (0.65, 1.31)	0.66	4790	0.77 (0.49, 1.21)	0.26	3514	0.76 (0.48, 1.21)	0.25	3512	
Total Score		7 yr	0.63 (0.36, 1.12)	0.12	4777	0.57 (0.28, 1.15)	0.12	3510	0.60 (0.30, 1.22)	0.16	3508	
Cognition	Verbal IQ	8 yr	1.51 (1.06, 2.14)	0.02	3997	1.20 (0.76, 1.91)	0.44	2952	1.07 (0.67, 1.71)	0.76	2950	
	Performance IQ	8 yr	1.32 (0.92, 1.89)	0.13	3990	1.43 (0.92, 2.21)	0.12	2945	1.42 (0.91, 2.21)	0.12	2943	
	Total IQ	8 yr	1.47 (1.03, 2.09)	0.03	3978	1.46 (0.93, 2.28)	0.10	2938	1.39 (0.89, 2.18)	0.15	2936	
Reading ability	Words per min	9 yr	1.03 (0.72, 1.49)	0.87	3794	1.05 (0.65, 1.68)	0.85	2763	1.04 (0.64, 1.67)	0.88	2761	
	Accuracy	9 yr	1.07 (0.74, 1.54)	0.74	3802	1.17 (0.73, 1.89)	0.52	2767	1.14 (0.70, 1.84)	0.61	2765	
	Comprehension	9 yr	1.05 (0.73, 1.52)	0.80	3802	0.98 (0.60, 1.60)	0.94	2767	0.98 (0.60, 1.62)	0.96	2765	
	Reading Score	9 yr	0.87 (0.60, 1.25)	0.45	4125	0.92 (0.58, 1.46)	0.74	3028	0.89 (0.56, 1.41)	0.61	3026	

mo, month; OR, odds ratio; n, number of subjects; yr, years. Suboptimal outcome defined as scores in the bottom quartile for ALSPAC pre-school development assessments, cognition, and reading ability. Published cut-offs² were used for behaviour: Prosocial (≤ 5 ; 9.8%), Peer problems (≥ 3 ; 13.5%), hyperactivity (≥ 6 ; 18.7%), emotional symptoms (≥ 4 ; 12.2%), conduct problems (≥ 3 ; 24.3%), and total score (≥ 14 ; 10.5%). Maternal vitamin D status >50 nmol/L was the reference group. †Model 1 adjusted for gestational week of vitamin D measurement; ‡Model 2: gestational week of vitamin D measurement plus additional 11 variables: maternal age, maternal BMI, maternal ethnic group, maternal education, maternal social class, parity, tobacco smoking in 1st trimester, home ownership status, crowding index, child gender, breastfeeding; §Model 3: additionally adjusted for oily fish and season of vitamin D measurement. ‖Age of child at development test included in all models.

Supplemental Table 4: Odds of suboptimum outcomes in children according to maternal vitamin D status (<75.0 vs ≥75.0nmol/L), minimally adjusted and fully adjusted for potential confounders

		Model1†				Model2‡				Model3§			
		Age	OR (95% CI)	p value	n	OR (95% CI)	p value	n	OR (95% CI)	p value	n		
ALSPAC early development tests	Gross Motor Skills	6 mo ‖	1.02 (0.90, 1.16)	0.76	6242	1.05 (0.90, 1.23)	0.54	4383	1.00 (0.85, 1.18)	1.00	4380		
		18 mo	0.96 (0.86, 1.06)	0.48	6269	1.05 (0.91, 1.20)	0.52	4385	1.11 (0.96, 1.29)	0.15	4383		
		30 mo	0.95 (0.84, 1.08)	0.45	5843	1.03 (0.89, 1.20)	0.67	4135	1.06 (0.91, 1.24)	0.47	4133		
		42 mo	0.98 (0.87, 1.12)	0.80	5695	1.00 (0.86, 1.17)	0.99	4073	1.05 (0.89, 1.23)	0.59	4070		
	Fine Motor Skills	6 mo ‖	0.86 (0.76, 0.98)	0.02	5880	0.94 (0.81, 1.10)	0.46	4141	0.92 (0.78, 1.08)	0.32	4139		
		18 mo	1.05 (0.94, 1.19)	0.39	6268	0.98 (0.85, 1.13)	0.79	4383	1.03 (0.89, 1.20)	0.69	4381		
		30 mo	0.95 (0.84, 1.07)	0.39	5854	0.95 (0.82, 1.10)	0.50	4138	0.95 (0.81, 1.11)	0.48	4136		
		42 mo	0.99 (0.87, 1.13)	0.88	5692	0.96 (0.82, 1.12)	0.62	4071	0.98 (0.83, 1.16)	0.85	4068		
	Social Development	6 mo ‖	0.94 (0.82, 1.07)	0.32	6010	0.91 (0.78, 1.06)	0.22	4209	0.87 (0.74, 1.03)	0.11	4207		
		18 mo	0.97 (0.86, 1.10)	0.64	6268	0.99 (0.85, 1.15)	0.91	4383	1.01 (0.86, 1.19)	0.88	4381		
		30 mo	0.93 (0.82, 1.05)	0.26	5843	1.00 (0.86, 1.17)	0.96	4129	0.95 (0.81, 1.12)	0.55	4127		
		42 mo	0.95 (0.83, 1.08)	0.40	5689	0.95 (0.82, 1.12)	0.56	4069	0.93 (0.79, 1.10)	0.39	4066		
Communication	6 mo ‖	0.97 (0.84, 1.13)	0.71	6100	0.96 (0.80, 1.15)	0.64	4285	0.95 (0.78, 1.15)	0.61	4283			
	18 mo	0.96 (0.85, 1.08)	0.51	6279	1.00 (0.86, 1.16)	0.98	4390	0.99 (0.84, 1.16)	0.88	4388			
Behaviour	Prosocial	7 yr	1.00 (0.83, 1.23)	0.97	4791	0.94 (0.74, 1.20)	0.61	3513	0.97 (0.75, 1.25)	0.79	3511		
	Peer problems	7 yr	1.16 (0.97, 1.39)	0.11	4785	1.06 (0.86, 1.31)	0.59	3510	1.09 (0.87, 1.37)	0.46	3508		
	Hyperactivity	7 yr	1.16 (0.99, 1.35)	0.07	4780	1.07 (0.89, 1.29)	0.48	3513	1.07 (0.87, 1.30)	0.52	3511		
	Emotional	7 yr	1.08 (0.90, 1.30)	0.42	4785	1.00 (0.81, 1.25)	0.97	3511	1.05 (0.83, 1.32)	0.71	3509		
	Conduct	7 yr	0.98 (0.85, 1.12)	0.75	4790	0.86 (0.73, 1.02)	0.09	3514	0.86 (0.72, 1.03)	0.10	3512		
	Total Score	7 yr	1.07 (0.88, 1.31)	0.49	4777	0.96 (0.75, 1.21)	0.71	3510	1.03 (0.80, 1.33)	0.81	3508		
Cognition	Verbal IQ	8 yr	1.23 (1.05, 1.43)	0.01	3997	1.16 (0.95, 1.41)	0.14	2952	1.09 (0.89, 1.35)	0.41	2950		
	Performance IQ	8 yr	1.07 (0.92, 1.25)	0.38	3990	0.96 (0.80, 1.16)	0.67	2945	0.98 (0.80, 1.19)	0.80	2943		
	Total IQ	8 yr	1.15 (0.99, 1.34)	0.07	3978	1.00 (0.83, 1.22)	0.96	2938	1.00 (0.82, 1.23)	0.99	2936		
Reading ability	Words per min	9 yr	1.13 (0.97, 1.33)	0.12	3794	1.16 (0.96, 1.42)	0.13	2763	1.18 (0.96, 1.46)	0.12	2761		
	Accuracy	9 yr	1.25 (1.06, 1.47)	0.007	3802	1.25 (1.02, 1.53)	0.04	2767	1.26 (1.01, 1.57)	0.04	2765		
	Comprehension	9 yr	1.19 (1.01, 1.39)	0.03	3802	1.07 (0.87, 1.31)	0.52	2767	1.10 (0.89, 1.37)	0.38	2765		
	Reading Score	9 yr	1.12 (0.97, 1.30)	0.12	4125	1.10 (0.92, 1.32)	0.29	3028	1.08 (0.90, 1.31)	0.41	3026		

mo, month; OR, odds ratio; n, number of subjects; yr, years. Suboptimal outcome defined as scores in the bottom quartile for ALSPAC pre-school development assessments, cognition, and reading ability. Published cut-offs² were used for behaviour: Prosocial (≤5; 9·8%), Peer problems (≥3; 13·5%), hyperactivity (≥6; 18·7%), emotional symptoms (≥4; 12·2%), conduct problems (≥3; 24·3%), and total score (≥14; 10·5%). Maternal vitamin D status >50nmol/L was the reference group. †Model 1 adjusted for gestational week of vitamin D measurement; ‡Model 2: gestational week of vitamin D measurement plus additional 11 variables: maternal age, maternal BMI, maternal ethnic group, maternal education, maternal social class, parity, tobacco smoking in 1st trimester, home ownership status, crowding index, child gender, breastfeeding; §Model 3: additionally adjusted for oily fish and season of vitamin D measurement. ‖Age of child at development test included in all models.

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