

Aetiological investigations in early developmental impairment: are they worth it?

Anthony Richard Hart,¹ Ruchi Sharma,² Mark Atherton,³ Samer Alabed,⁴ Sally Simpson,³ Stuart Barfield,³ Judith Cohen,⁵ Nicholas McGlashan,⁴ Asha Ravi,⁶ Michael James Parker,⁷ Daniel JA Connolly⁴

► Additional material is published online only. To view please visit the journal online (<http://dx.doi.org/10.1136/archdischild-2017-312843>).

For numbered affiliations see end of article.

Correspondence to

Dr Anthony Richard Hart, Department of Paediatric and Neonatal Neurology, Sheffield Children's Hospital NHS Foundation Trust, Ryegate Children's Centre, Tapton Crescent Road, Sheffield S10 5DD, UK; anthony.hart@sch.nhs.uk

Received 6 February 2017

Revised 16 May 2017

Accepted 25 May 2017

Published Online First

22 July 2017

ABSTRACT

Objective To study the frequency a diagnosis is made in children with early developmental impairment (EDI), and the contribution made to diagnosis by specific investigations.

Design Retrospective case note review.

Setting Community, neurodisability and neurology department at a UK tertiary centre.

Participants Children referred to determine the aetiology of EDI where a cause was not evident on history and examination. Participants were divided into two groups: EDI and no additional features (EDI-) and EDI with additional features (EDI+).

Main outcome measures The frequency a cause was found for the child's EDI and which tests contributed to a diagnosis.

Results 699 participants, 68.8% boys, median age at investigation 2 years 8 months (range 3 months to 11 years 5 months). 61 (8.7%) of participants had no investigations, and children with EDI- were less likely to be investigated ($\chi^2=12.5$, $p<0.05$). A diagnosis was made in 166 children (23.7%) and was more frequent in EDI+ (EDI- 9.9%, EDI+ 27.3%, $\chi^2=19.0$; $p<0.05$). Full blood count, zinc protoporphyrin, renal or liver function, bone profile, biotinidase, creatine kinase or lead level revealed no diagnoses. The following investigations found causes for EDI: MRI (23.1%), microarray (11.5%), Fragile X (0.9%), plasma amino acids (1.2%), urine organic acids (0.9%) and thyroid function tests (0.5%).

Conclusions The majority of 'screening' investigations for EDI do not contribute to a diagnosis, highlighting an area of cost saving for the NHS and reduced burden for patients and families. We propose a streamlined guideline for the investigation of EDI based on our data.

INTRODUCTION

Early developmental impairment (EDI) occurs when a child's developmental skills fall 2 SD or more below the population mean in two or more developmental domains.^{1,2} Ten per cent to 12% of children have a developmental impairment,^{3,4} and 1%–3% of children have EDI.^{1,3} The causes of EDI include genetic, metabolic, antenatal, endocrine and infective conditions, among others. The number of recommended investigations has increased over the last 20 years,^{1,3,5–16} although opinion varies on whether children with EDI are investigated appropriately, and the usefulness of specific investigations.¹⁷ This work aims to determine the frequency an aetiology was found for EDI, and the contribution of individual investigations.

What is already known on this topic?

- Early developmental impairment (EDI) is a common reason for referral to paediatricians and has many different aetiologies.
- Few of the aetiologies of EDI are treatable, but discovering the cause is important to families and helps predict recurrence risk.
- Recent recommendations suggest a battery of tests to determine the aetiology of EDI, but little data exist on the effectiveness of these tests.

What this study adds?

- The majority of recommended aetiological investigations do not reveal a cause for EDI.
- First-line investigations should include microarray and thyroid function. Creatine kinase could help identify Duchenne muscular dystrophy in boys.
- Other investigations should be reserved as second line, unless additional features exist or a strong suspicion of a specific condition exists.

METHODS

Patient identification

Children with EDI referred to Community Paediatrics, Neurodisability or Neurology services in Sheffield between January 2010 and December 2015 were identified by searching clinic letter databases. Children were included if the referral was to assess the level of EDI or determine cause. Children were not included if they were referred for another reason, such as epilepsy management, or if the cause was obvious on initial assessment. Although the term 'early developmental impairment' typically refers to children under 5 years of age, with intellectual or learning disability reserved for older children, we have included older children in our study because a small number of our referrals were children who had developmental impairment from a young age but did not have access to medical services or investigations until they moved to the UK.

Clinical phenotype and investigation results

Data were extracted from clinical notes including basic patient characteristics, investigations performed and investigation results. Clinical



- <http://dx.doi.org/10.1136/archdischild-2017-312738>
- <http://dx.doi.org/10.1136/archdischild-2017-312739>
- <http://dx.doi.org/10.1136/archdischild-2016-311271>
- <http://dx.doi.org/10.1136/archdischild-2017-313415>
- <http://dx.doi.org/10.1136/archdischild-2017-313532>



CrossMark

To cite: Hart AR, Sharma R, Atherton M, et al. *Arch Dis Child* 2017;**102**:1004–1013.

features were noted and two groups established: EDI without additional features (EDI-) and EDI with additional features (EDI+). Subgroups of additional features were formed if the feature was thought clinically relevant and affected at least five participants.

Four paediatricians reviewed participants' investigation results to determine the cause of their EDI. Results were categorised as:

- ▶ normal
- ▶ abnormal, but non-diagnostic (for example just outside the reference range)
- ▶ diagnostic.

Where doubt existed about the categorisation of the investigation, the first author was the final arbiter.

Two neuroradiologists reviewed participants' neuroimaging and categorised them as:

- ▶ 'normal'
- ▶ 'specific abnormalities explaining the participant's EDI or pointing towards a diagnosis'
- ▶ 'specific or non-specific abnormalities that did not suggest an aetiology'.¹⁸

A consultant paediatric neuroradiologist was the final arbiter on categorisation.

An investigation was considered to contribute to the diagnosis if it explained the cause of a child's EDI or allowed parents to comprehend the problem and the child to access educational support, as per Makela *et al.*¹⁹ Non-aetiological findings were considered diagnostic, such as 'cortical brain malformation', as were tests pointing towards a diagnosis proven on another investigation.

Statistical analysis

We report the frequency participants received the investigations recommended in our hospital guideline (online supplementary table 1), the frequency each test was diagnostic and differences in the frequency of diagnostic investigations between groups using a χ^2 test calculated with an online χ^2 generator. A p value <0.05 was assumed statistically significant.

Governance

The Clinical Governance department approved this work as a service evaluation. Data were collected in an anonymised format. No ethical approval was required.

RESULTS

Participant characteristics

Seven hundred and fifteen participants were referred over our 6-year period. Sixteen participants were excluded because either a diagnosis was obvious or insufficient clinical details were available. 481/699 (68.8%) participants were boys, 218 (31.2%) girls. Median age at time of investigation was 2 years 8 months (range 3 months to 11 years 5 months). A cause was found in 166 (23.7%) participants (table 1).

Investigation and diagnosis rates

61/699 (8.7%) participants had no investigations: 23/142 (16.2%) participants with EDI- and 38/557 (4.1%) with EDI+ ($\chi^2=12.5$, $p<0.05$). The local guideline was followed in entirety in 45 (6.4%) participants. A diagnosis was made in 14/142 (9.9%) children with EDI- and 152 (27.3%) with EDI+ ($\chi^2=19.0$; $p<0.05$). The additional features are shown in online supplementary figure 1a and the proportion of diagnoses for each additional feature in online supplementary figure 1b.

Results of specific investigations

The proportion of participants receiving each investigation on our guideline is shown in table 1. None of the following contributed to a diagnosis: full blood count, zinc protoporphyrin/haematinics, renal or liver function, bone profile, biotinidase, creatine kinase, lead level or urine GAGs.

During our study period, karyotype was replaced by Comparative genomic hybridization (CGH) microarray. 430/669 (64.3%) participants had a microarray only, 84 (12.0%) karyotype analysis only and 12 (1.7%) had microarray and karyotype analysis. Of the 442 participants who had microarray testing, 51 (11.5%) were diagnostic and 49 (11.1%) were a non-diagnostic abnormal result, such as polymorphisms or results of uncertain significance (online supplementary table 2). Participants with EDI+ were more likely to receive microarray testing (65.2% vs 55.6%, $\chi^2=4.4$; $p<0.05$), but diagnostic rates were similar between groups (EDI- 11.4%, EDI+ 11.6% ($\chi^2=0.0$; $p=0.96$)). Table 2 and online supplementary figure 1c show the proportion of diagnostic microarrays for each additional feature. Participants with dysmorphia had similar diagnostic rates to participants with EDI+ but no dysmorphia (11.8% vs 11.5%, $\chi^2=0.2$; $p=0.66$).

Fragile X was requested in 113/699 (16.2%) participants, 32 of whom had a family history of learning/developmental difficulties. Fragile X was abnormal in one case: a 2-year-old boy with no family history or additional features. Genetic diagnoses were made following review by a clinical geneticist or specific gene analysis (online supplementary table 2).

Thyroid function was diagnostic in three participants, all of whom had additional features (table 1), and were autoimmune in aetiology. Plasma amino acid screens were diagnostic in five participants and urine organic acids in three, two of whom had abnormal plasma amino acids. Clinicians ordered a variety of other investigations (table 3) and revealed a diagnosis in five participants.

Neuroimaging

MR imaging was performed in 368/699 (52.6%) participants. Children with EDI+ were more likely to receive neuroimaging compared with EDI- (additional features 60.7% vs 21.1% for no additional features, $\chi^2=71.0$; $p<0.05$). MRI was diagnostic in 85 (23.1%) participants: 81/338 (24.0%) participants with EDI+ and 4/30 (13.3%) EDI- ($\chi^2=1.8$; $p=0.18$). Table 4 shows the distribution of abnormalities found on MR imaging and online supplementary figure 1d shows the frequency of abnormalities for additional feature subgroups.

DISCUSSION

Why investigate for the cause of EDI?

The reasons why families wish to know the aetiology include¹⁹:

- ▶ validation, that is, proof of a credible problem and to explain the problem to others;
- ▶ to provide prognostic information and help set realistic expectations/plans;
- ▶ for potential treatments;
- ▶ to allow access to educational support;
- ▶ to allow for early intervention;
- ▶ to provide support opportunities;
- ▶ the 'need to know';
- ▶ to end the 'diagnostic odyssey';
- ▶ for prenatal testing and recurrence risk.

Not all diagnoses are equally effective at enabling access to educational support despite children having similar levels of

Table 1 The frequency a diagnosis was achieved in participants, and the frequency investigations on our guidelines were normal, abnormal non-diagnostic and diagnostic

		Investigations (%)																
Participants	Diagnosis reached?	Result	FBC	ZPP	U&E	LFT	Bone profile	TFT	Biotinidase	CK	Lead level	Plasma amino acids	Urine organic acids	Urine GAGs	Karyotype	Microarray	Fragile X	MRI brain
All participants (n=699)	166 (23.7%)	Number receiving test	526/699 (75.2%)	278/699 (39.8%)	512/699 (73.2%)	491/699 (70.2%)	429/699 (61.4%)	469/699 (67.1%)	227/699 (32.5%)	388/699 (55.5%)	40/699 (5.7%)	413/699 (59.1%)	318/699 (45.4%)	217/699 (31.0%)	96/699 (13.7%)	442/699 (63.2%)	113/699 (16.2%)	368/699 (52.6%)
		Normal	444/526 (84.4%)	205/278 (73.7%)	477/512 (93.2%)	434/491 (88.4%)	412/429 (96.0%)	431/469 (91.9%)	216/227 (95.2%)	340/388 (87.6%)	40/40 (100%)	392/413 (94.9%)	286/318 (90.0%)	200/217 (92.2%)	94/96 (97.9%)	342/442 (77.4%)	112/113 (99.1%)	208/368 (56.5%)
		Abnormal: non-diagnostic	82/526 (15.6%)	73/278 (26.3%)	35/512 (6.8%)	57/491 (11.6%)	17/429 (4.0%)	35/469 (7.5%)	11/227 (4.8%)	11/227 (4.8%)	48/388 (12.4%)	0/40 (0%)	16/413 (3.9%)	29/318 (9.1%)	17/217 (7.8%)	0/96 (0%)	49/442 (11.1%)	0/113 (0%)
EDI without additional features (n=142)	14 (9.9%)	Number receiving test	99/142 (69.7%)	66/142 (46.5%)	91/142 (64.1%)	87/142 (61.3%)	79/142 (55.6%)	91/142 (64.1%)	44/142 (31.0%)	80/142 (56.3%)	5/142 (3.5%)	75/142 (52.8%)	53/142 (37.3%)	38/142 (26.8%)	9/142 (6.3%)	79/142 (55.6%)	24/142 (16.9%)	30/142 (21.1%)
		Normal	86/99 (86.9%)	52/66 (78.8%)	90/91 (98.9%)	81/87 (93.1%)	75/79 (94.9%)	86/91 (94.5%)	44/44 (100%)	73/80 (91.3%)	5/5 (100%)	71/75 (94.7%)	50/53 (94.3%)	31/38 (81.6%)	9/9 (100%)	60/79 (75.9%)	23/24 (95.8%)	20/30 (66.7%)
		Abnormal: non-diagnostic	13/99 (13.1%)	14/66 (21.2%)	1/91 (1.1%)	6/87 (6.9%)	4/79 (5.1%)	5/91 (5.5%)	0/44 (0%)	0/44 (0%)	7/80 (8.7%)	0/5 (0%)	4/75 (5.3%)	3/53 (5.7%)	7/38 (18.4%)	0/9 (0%)	10/79 (12.7%)	0/24 (0%)
All participants with EDI and additional features (n=557)	152 (27.3%)	Number receiving test	427/557 (76.7%)	212/557 (38.1%)	421/557 (75.6%)	404/557 (72.5%)	350/557 (62.8%)	378/557 (67.9%)	183/557 (32.9%)	308/557 (55.3%)	35/557 (6.3%)	338/557 (60.7%)	265/557 (47.6%)	179/557 (32.1%)	86/557 (15.4%)	363/557 (65.2%)	89/557 (16.0%)	338/557 (60.7%)
		Normal	358/428 (83.6%)	153/212 (72.2%)	387/421 (91.9%)	353/404 (87.4%)	337/350 (96.3%)	345/378 (91.3%)	172/183 (94.0%)	267/308 (86.7%)	0/35 (0%)	321/338 (94.9%)	236/265 (89.1%)	169/179 (94.4%)	84/86 (97.7%)	282/363 (77.7%)	89/89 (100%)	188/338 (55.6%)
		Abnormal: non-diagnostic	69/428 (16.1%)	59/212 (27.8%)	34/421 (8.1%)	51/404 (12.6%)	13/350 (3.7%)	30/378 (7.9%)	11/183 (6.0%)	11/183 (6.0%)	41/308 (13.3%)	35/35 (100%)	12/338 (3.6%)	26/265 (9.8%)	10/179 (5.6%)	0/86 (0%)	39/363 (10.7%)	0/89 (0%)
Additional features	Abnormal: diagnostic	Number receiving test	0/428 (0%)	0/212 (0%)	0/421 (0%)	0/404 (0%)	0/350 (0%)	0/378 (0%)	0/183 (0%)	0/308 (0%)	0/35 (0%)	5/338 (1.5%)	3/265 (1.1%)	0/179 (0%)	2/86 (2.3%)	42/363 (11.6%)	0/89 (0%)	81/338 (24.0%)
		Normal	0/428 (0%)	0/212 (0%)	0/421 (0%)	0/404 (0%)	0/350 (0%)	0/378 (0%)	0/183 (0%)	0/308 (0%)	0/35 (0%)	0/338 (0%)	0/265 (0%)	0/179 (0%)	0/86 (0%)	0/363 (0%)	0/89 (0%)	0/338 (0%)
		Abnormal: diagnostic	0/428 (0%)	0/212 (0%)	0/421 (0%)	0/404 (0%)	0/350 (0%)	0/378 (0%)	0/183 (0%)	0/308 (0%)	0/35 (0%)	0/338 (0%)	0/265 (0%)	0/179 (0%)	0/86 (0%)	0/363 (0%)	0/89 (0%)	0/338 (0%)

*Thyroid function: (a) boy presented at 1 year 10 months as a placid baby with EDI most obvious in motor skills and with hypotonia. Autoimmune hypothyroidism diagnosed and he improved with treatment although subsequently developed ASD; (b) ex-preterm (29 weeks) girl investigated at 4 years 7 months to a mother with learning difficulties, had general developmental impairment, especially in speech skills who improved on treatment but had attention and behavioural difficulties; (c) girl investigated at 5 years 6 months for EDI, nystagmus with a history of maternal illicit drug use in pregnancy—showed improvement on treatment, but not to normal abilities. Dual diagnosis is being investigated.
 †Plasma amino acids: (a) boy investigated at 4 years 11 months with EDI and seizures whose brother had similar phenotype diagnosed with homocystinuria; (b) 4-month-old boy with EDI, microcephaly, hypertonia and poor visual function with low serum and CSF serine, diagnosed with serine deficiency; (c) girl investigated at 2 years 11 months with EDI, brittle hair, eczema and dysmorphism diagnosed with ethylmalonic aciduria; (d) boy investigated at 4 years 7 months with EDI and faltering growth diagnosed with lysinuric protein intolerance; (e) boy investigated at 10 months with EDI, visual impairment and hypotonia noted to have low serum and CSF serine, diagnosed as serine deficiency.
 ‡Urine organic acids: ex-premature girl investigated at 8 months for EDI, acquired microcephaly and faltering growth with raised lactate and glutamate, subsequently diagnosed with pyruvate dehydrogenase deficiency, see cases (c) and (d) in the plasma amino acid results.
 ASD, Autistic spectrum disorder; CSF, cerebrospinal fluid; CK, creatine kinase; EDI, early developmental impairment; FBC, full blood count; GAG, glycosaminoglycans; LFT, liver function test; TFT, thyroid function test; U&E, urea and electrolytes; ZPP, zinc protoporphyrin;

Table 2 The frequency thyroid function, plasma amino acids, urine organic acids, microarray, karyotype and MR imaging were positive subgroups of additional features

Participants	No of participants in whom a cause was found (%)	Investigations						
		TFT	Plasma amino acids	Urine organic acids	Karyotype	Microarray	Fragile X	MRI brain
All participants with EDI and additional features (n=557)	152 (27.3%)	378 (67.9%) 3/378 (0.8%)	338 (60.7%) 5/338 (1.5%)	265 (47.6%) 3/265 (1.1%)	86 (15.4%) 2/86 (2.3%)	363 (65.2) 42/363 (11.6%)	89 (16.0%) 0/89 (0%)	338 (60.7%) 81 (24.0%)
Additional features and a family history of EDI/ID or neurological disease like seizures (n=115)	26 (22.6%)	82 (71.3%) 1/82 (1.2%)	75 (65.2%) 1/75 (1.3%)	50 (43.5%) 0/50 (0%)	13 (11.3%) 0/13 (0%)	81 (70.4%) 10/81 (12.3%)	19 (16.5%) 0/19 (0%)	67 (58.3%) 10/67 (14.9%)
Consanguinity admitted by family (n=26)	7 (26.9%)	20 (76.9%) 0/20 (0%)	17 (65.4%) 0/17 (0%)	15 (57.7%) 0/15 (0%)	4 (15.4%) 0/4 (0%)	16 (61.5%) 2/16 (12.5%)	2 (7.7%) 0/2 (0%)	16 (61.5%) 5 (31.3%)
Dysmorphia (n=132)	38 (28.8%)	95 (72.0%) 0/95 (0%)	89 (67.4%) 2/89 (2.2%)	68 (51.5%) 1/68 (1.5%)	14 (10.6%) 0/14 (0%)	110 (83.3%) 13/110 (11.8%)	20 (15.2%) 0/20 (0%)	91 (68.9%) 19/91 (20.9%)
Macrocephaly (n=33)	8 (24.2%)	22 (66.7%) 0/22 (0%)	20 (60.6%) 0/20 (0%)	15 (45.5%) 0/15 (0%)	3 (9.1%) 0/3 (0%)	23 (69.7%) 4/23 (17.4%)	5 (15.2%) 0/5 (0%)	25 (75.8%) 4/25 (16.0%)
Microcephaly (n=162)	46 (28.4%)	110 (67.9%) 1/110 (0.9%)	106 (65.4%) 2/106 (1.9%)	85 (52.5%) 1/85 (1.2%)	27 (16.7%) 1/27 (3.7%)	112 (69.1%) 15/112 (13.4%)	27 (16.7%) 0 (0%)	100 (61.7%) 24 (24.0%)
Hypertonia/upper motor neuron findings (spasticity, dystonia, brisk reflexes) (n=82)	40 (48.8%)	56 (68.3%) 0/56 (0%)	57 (69.5%) 1/57 (1.8%)	51 (62.2%) 0/51 (0%)	10 (12.2%) 0/10 (0%)	60 (73.2%) 4/60 (6.7%)	3 (3.7%) 0/3 (0%)	79 (96.3%) 31 (39.2%)
Hypotonia (n=84)	27 (32.1%)	57 (67.9%) 1/57 (1.8%)	54 (64.3%) 1/54 (1.9%)	47 (56.0%) 0/47 (0%)	10 (11.9%) 0/10 (0%)	56 (66.7%) 5/56 (8.9%)	8 (9.5%) 0/8 (0%)	65 (77.4%) 18/65 (27.7%)
Gait abnormalities, tremor, cerebellar signs or other movement disorders (n=49)	17 (34.7%)	37 (75.5%) 0/37 (0%)	28 (57.1%) 0/28 (0%)	24 (49.0%) 0/24 (0%)	11 (22.4%) 0/11 (0%)	26 (53.1%) 3/26 (11.5%)	4 (8.2%) 0/4 (0%)	39 (79.6%) 11/39 (28.2%)
Epilepsy (n=29)	17 (58.6%)	17 (58.6%) 0/17 (0%)	22 (75.9%) 1/22 (4.5%)	16 (55.2%) 0/16 (0%)	4 (13.8%) 0/4 (0%)	17 (58.6%) 2/17 (11.8%)	1 (3.4%) 0/1 (0%)	27 (93.1%) 10/27 (37.0%)
Ophthalmological signs (n=96)	43 (44.8%)	64 (66.7%) 1/64 (1.6%)	59 (61.5%) 2/59 (3.4%)	46 (47.9%) 0/46 (0%)	15 (15.6%) 0/15 (0%)	64 (66.7%) 7/64 (10.9%)	9 (9.4%) 0/9 (0%)	78 (81.3%) 28/78 (35.9%)
Hearing difficulties (n=27)	7 (25.9%)	17 (63.0%) 0/17 (0%)	18 (66.7%) 0/18 (0%)	15 (55.6%) 0/15 (0%)	2 (7.4%) 0/2 (0%)	18 (66.7%) 2/18 (11.1%)	2 (7.4%) 0/2 (0%)	22 (81.5%) 4/22 (18.2%)
Cardiac disorder, including congenital cardiac lesions (n=24)	8 (33.3%)	14 (58.3%) 0/14 (0%)	13 (54.2%) 0/13 (0%)	11 (45.8%) 0/11 (0%)	2 (8.3%) 0/2 (0%)	20 (83.3%) 6/20 (30.0%)	1 (4.2%) 0/1 (0%)	13 (54.2%) 1/13 (7.7%)
Airway problem/tracheostomy/sleep apnoea (n=14)	6 (42.9%)	7 (50.0%) 0/7 (0%)	11 (78.6%) 0/11 (0%)	9 (64.3%) 0/9 (0%)	3 (21.4%) 1/3 (33.3%)	9 (64.3%) 2/9 (22.2%)	1 (7.1%) 0/1 (0%)	11 (78.6%) 3/11 (27.3%)
Feeding/swallowing difficulties (n=28)	9 (32.1%)	18 (64.3%) 0/18 (0%)	17 (60.7%) 0/17 (0%)	16 (57.1%) 0/16 (0%)	1 (3.6%) 0/1 (0%)	20 (71.4%) 2/10 (20.0%)	2 (7.1%) 0/2 (0%)	23 (82.1%) 7/23 (30.4%)
Hepatomegaly, abnormal liver function tests, splenomegaly or other features suggestive of inborn error of metabolism (n=12)	3 (25.0%)	9 (75.0%) 0/9 (0%)	12 (100%) 0/12 (0%)	10 (83.3%) 0/10 (0%)	4 (25.0%) 0/4 (0%)	7 (58.3%) 1/7 (14.3%)	2 (15.4%) 0/2 (0%)	12 (100%) 0/12 (0%)
Kidney, urinary tract abnormalities (n=17)	6 (35.3%)	9 (52.9%) 0/9 (0%)	12 (70.6%) 0/12 (0%)	9 (52.9%) 0/9 (0%)	5 (29.4%) 0/5 (0%)	11 (64.7%) 2/11 (18.2%)	1 (5.9%) 0/1 (0%)	12 (70.6%) 3/12 (25.0%)
Poor growth/failure to thrive/short stature (n=61)	22 (36.1%)	46 (75.4%) 0/46 (0%)	39 (63.9%) 1/39 (2.6%)	31 (50.8%) 2/31 (6.5%)	12 (19.7%) 0/12 (0%)	44 (72.1%) 5/44 (11.4%)	10 (16.4%) 0/10 (0%)	39 (63.9%) 14/39 (35.9%)
Obesity (n=17)	2 (11.8%)	12 (70.6%) 0/12 (0%)	9 (52.9%) 0/9 (0%)	7 (41.2%) 0/7 (0%)	1 (5.9%) 0/1 (0%)	9 (52.9%) 1/9 (11.1%)	1 (5.9%) 0/1 (0%)	7 (41.2%) 0/7 (0%)
Skeletal/rheumatological disorder (n=16)	9 (56.3%)	9 (56.3%) 0/9 (0%)	9 (56.3%) 0/9 (0%)	6 (37.5%) 0/6 (0%)	3 (18.8%) 0/3 (0%)	12 (75.0%) 3/12 (25.0%)	2 (12.5%) 0/2 (0%)	14 (87.5%) 6/14 (42.9%)

Continued

Table 2 Continued

Participants	No of participants in whom a cause was found (%)	Investigations							
		TFT	Plasma amino acids	Urine organic acids	Karyotype	Microarray	Fragile X	MRI brain	
Features suggestive of ASD (n=99)	13 (13.1%)	62 (62.6%) 0/62 (0%)	54 (54.5%) 0/54 (0%)	44 (44.4%) 0/44 (0%)	19 (19.2%) 1/19 (5.3%)	55 (55.6%) 4/55 (7.3%)	26 (26.3%) 0/26 (0%)	42 (42.4%) 4/42 (9.5%)	
Concerns about attention (n=14)	2 (14.3%)	9 (64.3%) 0/9 (0%)	5 (35.7%) 0/5 (0%)	2 (14.3%) 0/2 (0%)	3 (21.4%) 0/3 (0%)	8 (57.1%) 1/8 (12.5%)	3 (21.4%) 0/3 (0%)	8 (57.1%) 1/8 (12.5%)	
Antenatal concerns for example, hydrops, alcohol or drug use (n=24)	10 (41.7%)	15 (62.5%) 1/15 (6.7%)	12 (50.0%) 0/12 (0%)	8 (33.3%) 0/8 (0%)	4 (16.7%) 0/4 (0%)	17 (70.8%) 2/17 (11.8%)	4 (16.7%) 0/4 (0%)	16 (66.7%) 5/16 (31.3%)	
Ex-preterm (but not felt to explain EDI) (n=59)	13 (22.0%)	39 (66.1%) 1/39 (2.6%)	34 (57.6%) 0/34 (0%)	22 (37.3%) 1/22 (4.5%)	11 (18.6%) 0/11 (0%)	33 (55.9%) 2/33 (6.1%)	5 (8.5%) 0/5 (0%)	28 (47.5%) 5/28 (17.9%)	

ASD, autistic spectrum disorder; EDI, early developmental impairment; LD, learning disabilities; TFT, thyroid function test.

disability. Descriptive diagnoses could be equally, if not more, effective than aetiological diagnoses.

Investigating the cause of EDI

The initial steps to investigate children with EDI are history and examination, and an assessment of hearing should also be considered early. The effect of the child's environment, including poverty, parental educational levels, employment status and family status, needs to be taken into account and addressed. In some situations, these may need to be tackled before investigations are undertaken.

In cases where no clear cause for the child's EDI is evident on initial assessment, there is little agreement on the best investigations to perform, with wide variation in opinion existing between UK paediatricians.¹⁷ Over the years, the number of recommended investigations for EDI has increased,^{1 3 5-16} although the evidence base for this is lacking. The low adherence to our clinical guideline reflects clinician uncertainty about the optimal way to investigate children with EDI.

A cause was found in 23.7% of our participants. This may have been a result of a single test or a contribution of many. Many of the first-line investigations in our guideline were of no value in determining an aetiology.

Biotinidase level is recommended in some guidelines.^{3 12} The features of biotinidase deficiency include seizures, hypotonia, laryngeal stridor, tachypnoea, apnoea, alopecia, skin rash, hearing loss, optic atrophy or conjunctivitis, ataxia, fungal infection, recurrent myelopathy, hyperammonaemia or specific organic acid abnormalities.²⁰⁻²⁴ It is not clear that biotinidase deficiency presents with EDI.²⁵ The incidence of biotinidase deficiency on newborn screening suggests is low.²⁰ Recommendations that routine testing is cost-effective²⁶ are not evidence based. Our view is that serum biotinidase should only be performed second or third line in children with suggestive features.

High blood lead levels are associated with learning and mental health issues that can persist into adulthood.^{27 28} Low levels of blood lead (5-10 µg/dL) are associated with reading and writing difficulties.²⁹ However, it is unlikely that a modern child would have moderate or severe EDI because of extremely high lead levels alone, and the proportion of children with high lead levels has fallen to 0.8% in the USA.^{30 31} Therefore, routine lead levels appear to be unnecessary.

Creatine kinase (CK) is recommended in boys with EDI to facilitate early diagnosis of Duchenne muscular dystrophy (DMD),^{3 12} ensure treatment with steroids and/or newer drugs, and for entry into research studies. These children may present with EDI or social communication difficulties before muscle weakness is appreciated, so clinical phenotype cannot be relied on to trigger testing. Although no child in our study had DMD, it is our opinion that DMD is common and a CK cheap enough for it to be performed routinely in boys with EDI.

Investigations likely to determine the cause of EDI

The investigations that contributed to a diagnosis were:

- ▶ thyroid function
- ▶ plasma amino acids
- ▶ urine organic acids
- ▶ microarray/karyotype
- ▶ Fragile X
- ▶ MR imaging.

Congenital hypothyroidism is usually diagnosed on newborn screening in the UK, unless it arises from pituitary failure. Auto-immune hypothyroidism will also be missed. The cost of each

Table 3 Number of participants receiving additional investigations

Investigation	No of participants (% of total)	Comments
Blood investigations		
Clotting	5	Abnormal in one patient, but already known to be abnormal prior to referral for investigation of EDI
Folate	8	Not diagnostic in any participant
C reactive protein	1	Not diagnostic in any participant
Erythrocyte sedimentation rate	4	Not diagnostic in any participant
Glucose	58	No abnormalities
Lactate	178	Persistently abnormal in one boy investigated at 1.4 years of age with dysmorphism, pulmonary stenosis, squint and hypertonia; MRS showed lactate peak although respiratory chain enzymes normal and no genetic diagnosis of mitochondrial disease made
Acylcarnitine screen	100	Not diagnostic in any participant
Magnesium	3	Not diagnostic in any participant
Ammonia	67	Not diagnostic in any participant
Homocysteine	11	Not diagnostic in any participant
Uric acid	103	Not diagnostic in any participant
Lipid/cholesterols	40	Not diagnostic in any participant
White cell enzymes	45	Not diagnostic in any participant
Very long chain fatty acids	60	Not diagnostic in any participant
Transferrin isoelectric focusing	46	Not diagnostic in any participant
Galactosaemia screen	15	Not diagnostic in any participant
Copper	21	Not diagnostic in any participant
Caeruloplasmin	20	Not diagnostic in any participant
Free T3	9	Not diagnostic in any participant
Cortisol	12	Not diagnostic in any participant
Vitamin A	7	Not diagnostic in any participant
Vitamin B ₁₂	10	Not diagnostic in any participant
Vitamin D	193	Low levels were not felt to be the aetiological cause of EDI in any participant
Vitamin E	9	Not diagnostic in any participant
Coeliac screen	40	Not diagnostic in any participant
Amylase	5	Not diagnostic in any participant
Parathyroid hormone	48	Not diagnostic in any participant
Immunoglobulins	42	Not diagnostic in any participant
LDH	3	Not diagnostic in any participant
Alpha fetoprotein	5	Not diagnostic in any participant
ANA	4	Not diagnostic in any participant
Rheumatoid factor	2	Not diagnostic in any participant
Congenital infection screen	34	Not diagnostic in any participant
Urine investigations		
Urine amino acids	150	Not diagnostic in any participant
Urine creatinine	13	Not diagnostic in any participant
Oligosaccharides	12	Not diagnostic in any participant
Purine/pyrimidine studies	30	Often done after a marginally low uric acid, normal in all participants
Cerebrospinal fluid tests		
CSF glucose lactate	58	Lactate raised in one boy investigated at 1.4 years of age with dysmorphism, pulmonary stenosis, squint and hypertonia; MRS showed lactate peak although respiratory chain enzymes normal and no genetic diagnosis of mitochondrial disease made Glucose low in two patients: a boy investigated at 1.7 years with EDI, hypotonia, seizures and paroxysmal upgaze when fasted; a girl investigated at 1.9 years with EDI, left hemiplegia and seizures. Both confirmed to have mutations in Glut 1 deficiency syndrome gene
CSF amino acids	19	Diagnostic in two cases: a boy, investigated at 4 months of age for developmental impairment, poor visual function, evolving dystonia and microcephaly diagnosed with serine deficiency; a boy investigated at 0.8 year for developmental impairment, poor visual function and microcephaly whose plasma amino acids revealed low serine, ultimately diagnosed with serine deficiency
CSF neurotransmitters	32	Not diagnostic in any participant

Continued

Table 3 Continued

Investigation	No of participants (% of total)	Comments
CSF viral studies	18	Not diagnostic in any participant
Other		
Skeletal survey	2	Not diagnostic in any participant
Muscle biopsy	9	Not diagnostic in any participant
Fibroblast culture	2	Sent for variety of tests—not diagnostic in any participant

ANA, antinuclear antibody; CSF, cerebrospinal fluid; EDI, early developmental impairment; LDH, lactate dehydrogenase; MRS, magnetic resonance proton spectroscopy.

test is low (£5.94). Given 469 participants were tested and three were positive for this diagnosis, two of whom showed improvement on treatment, the cost per diagnosis in our cohort was £929, which appears cost-effective given a treatment exists to improve outcome.

Plasma amino acids were diagnostic in 1.2% of our participants, all of whom had EDI+. Organic acids were abnormal in 0.9%, two of whom had abnormal plasma amino acids. Urine Glycosaminoglycans (GAGs) revealed no diagnoses. Other metabolic investigations were performed in our cohort, outside of the guideline, but were rarely diagnostic. Because our participants with a positive result had EDI+, we do not recommend metabolic tests in EDI-. In children with EDI+, metabolic tests should be first line where concern exists about an inborn error of metabolism.³ Where no such concern exists, they should be second line, or performed if the presentation changes or other investigations point towards these conditions.

Microarray found a diagnosis in over 10% of our cases, with similar frequencies between EDI- and EDI+. We advocate microarray testing in all children with EDI. We recommend Fragile X testing if the microarray is normal in boys with consistent syndromal features or a child of either sex with a family history consistent with Fragile X. Where no diagnosis is found

after initial investigations in children with EDI+ or a relevant family history, referral to a clinical geneticist should be considered. During the study period, the Clinical Genetics service recruited to the Deciphering Developmental Disorders (DDD) study, and we await the results of this study and the UK 100,000 genome project in investigating children with EDI.

The role of MRI in EDI is controversial, particularly as sedation or general anaesthetic is required for those who cannot lie still. Consensus statements in 1997 and 2003 recommended MRI only in children with EDI and additional symptoms based on a belief that children with EDI and no additional symptoms were unlikely to have structural brain abnormalities.^{1 5} There was a paucity of data to support this view. Our previous analysis data show varying rates of abnormality between EDI+ and EDI- groups.^{18 32} Our current data show that MR imaging has the highest diagnostic rates of any tests, with 13.3% in children without additional features and 24% in those with additional features. The lack of a statistical difference between groups is probably a type 2 statistical error.

Clinically, we advocate a common-sense approach. In a child with EDI-, MRI should not be a first-line test because of the anaesthetic risks and because of the yield of microarray and thyroid function. Where no diagnosis is found on first-line

Table 4 MRI abnormalities seen in our cohort

MR abnormalities	All participants	EDI and no additional features	EDI and additional features
Specific abnormalities likely to explain EDI or point towards a diagnosis			
Hypoxic ischaemic injury:	25	1	24
▶ Periventricular leukomalacia			
▶ Central (predominately basal ganglia)			
▶ Peripheral (predominately watershed areas)			
Toxic metabolic	17	0	17
Brain malformation	37	2	35
Phakomatoses/tumours	3	1	2
Previous infection	0	0	0
Hydrocephalus	3	0	3
Total	85	4	81
Specific or non-specific abnormalities not likely to explain EDI			
Posterior fossa abnormalities, such as Chiari malformation	6	1	5
Cavum septum pellucidum	2	0	2
Under-opercularisation of the Sylvian fissure	0	0	0
Reduced volume of white matter/grey matter	21	2	19
Delayed myelination	12	0	12
Non-specific white matter abnormality or focal gliosis	30	2	28
Arachnoid cyst	2	0	2
Optic nerve hypoplasia without evidence of septic optic dysplasia	0	0	0
Subdural collection/effusion	0	0	0
Non-specific MRS abnormalities	2	1	1
Total	75	6	69

EDI, early developmental impairment.

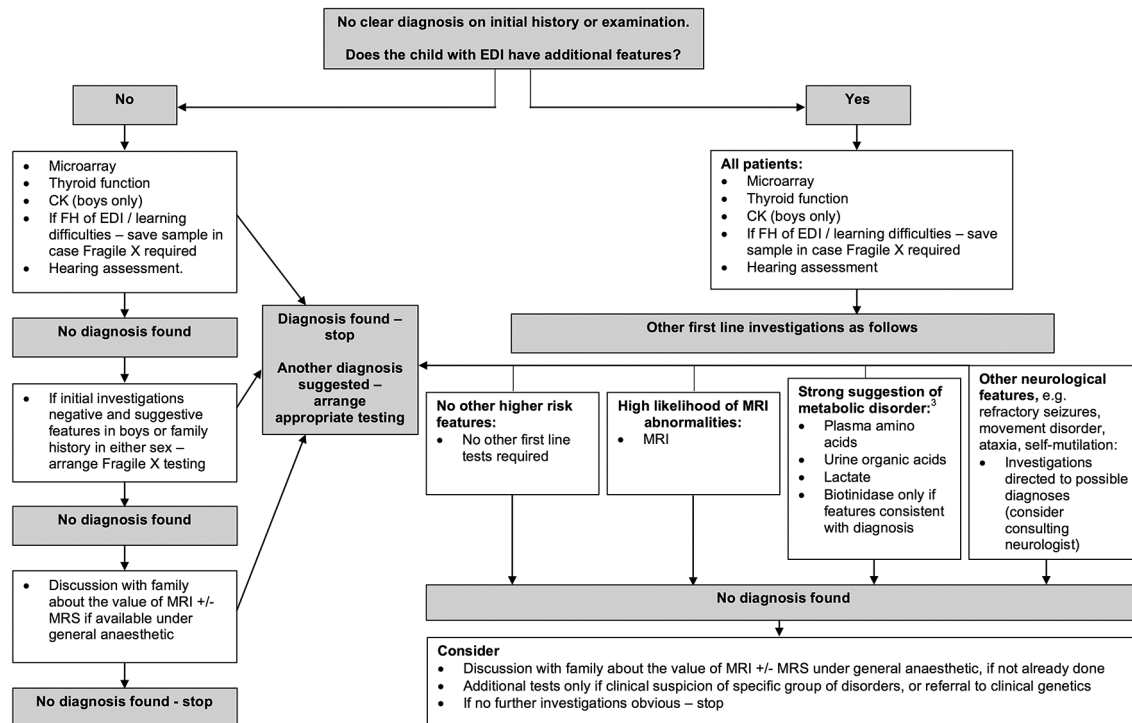


Figure 1 Guideline on how to investigate children with EDI. CK, creatine kinase; FH, family history; MRS, magnetic resonance proton spectroscopy. High likelihood of MRI abnormalities: skeletal dysplasia, signs of upper motor neuron involvement, refractory or focal seizures, ophthalmological signs, failure to thrive not explained by poor calorific input, antenatal concerns such as maternal drug use or hydrops, feeding/swallowing difficulties, gait or movement abnormalities, central mediated hypotonia, and airway problems like vocal cord palsy or central apnoea.

laboratory testing, MRI should be discussed with the family, including the likelihood that no specific treatment is available for any abnormality, and the risks of general anaesthesia.

In children with EDI+, MRI should be first line if a high risk of specific MRI abnormalities is likely: skeletal dysplasia, signs of upper motor neuron involvement, refractory or focal seizures, ophthalmological signs, failure to thrive not explained by poor calorific input, antenatal concerns such as maternal drug use or hydrops, feeding/swallowing difficulties, gait or movement abnormalities, central mediated hypotonia and airway problems. In other children, MRI could be second-line investigation if the parents wanted it or deferred. Where a genetic diagnosis is found, MRI is not necessary unless a specific indication exists that would change management.

Our unit performs MR spectroscopy in children with EDI to detect conditions like creatine deficiency.¹⁸ We found a case of creatine transporter deficiency using MRS, a child with large lactate peak suspected to have a defect in pyruvate metabolism, and a third participant with a non-specific reduction in choline levels. Because MRS adds only around 8 min to acquisition times, and creatine deficiencies are treatable, we recommend MRS for children with EDI when MRI is performed where it is available.

Where no diagnosis is found

If the initial screen of investigations prove negative, we would not recommend relentless investigations for increasingly rare conditions and the diagnostic voyage can cease. It may be beneficial for units to have a forum to discuss difficult cases, as the clinical experience of clinicians may vary: more experienced clinicians may recognise subtle clues to rare diagnoses, and geneticists may help identify dysmorphic features/diagnoses and facilitate access to single gene tests, gene exome studies

or research projects. When a diagnostic voyage does end, the clinician should remember to review periodically whether new features have arisen or new technologies developed to help identify a cause.

Limitations with our data

As with previous retrospective data on the investigation of children with EDI,³² our data have methodological flaws. We relied on the documentation in medical notes and clinical letters to phenotype our participants, and it is possible that features may not have been documented. Many recommended investigations were not performed, and we do not know why clinicians ordered specific additional tests. It is possible that missing data from investigations that were not performed may have contributed towards a diagnosis. Clinical notes are the result of review in busy clinics, so no formal developmental assessment is available to confirm the diagnosis, assess severity, or domains affected. Furthermore, while the term EDI is usually reserved for preschool children under 5 years of age, we included older children. Our rationale for this was that it was likely these children would have been diagnosed with EDI and investigated if they had been in the UK. It is also debatable whether the term 'early', and the division of under and over 5 years of age, is relevant to diagnostic rates of investigations.

It is also possible that many factors may combine to affect a child's development. For example, anaemia or vitamin D deficiency, low parental educational ability and/or poor stimulation/environment may each play an additive role. In these situations, medical treatment of any abnormalities found, such as mild to moderate anaemia or vitamin D deficiency, may lead to some improvement in development. Even though this may not return to the child's development level to 'normal', it may give a clinically significant improvement for the child, improving their sense

of well-being and quality of life. While a proportion of children in our study had non-diagnostic abnormalities in investigations like full blood count, renal, liver function or vitamin D, the clinical notes did not reveal any child in whom treatment led to a significant improvement in developmental abilities. This may reflect our methodology and the limitations of relying on medical notes. Until future work shows if and how frequently this happens, we retain a strict definition as to what a diagnostic test is.

There is also a temptation in paediatrics to do as many tests as possible 'in one go' to avoid repeated venesection and to include screening tests. We do not support this approach. Our data suggest none of these tests revealed a cause, and it is not appropriate to manage or reassure a clinician's curiosity by performing investigations. Although we did not measure this in our cohort, a proportion of these 'screening tests' were not processed because of technical factors, such as the bottle leaked. Often this test was not thought sufficiently important to repeat, which questions why it was requested in the first place. In addition, other investigations were mildly abnormal or the results were of uncertain significance, which lead the child to having further investigations that were ultimately negative. Such distress could have been avoided if the initial investigation was not performed at all. Cost savings from not performing screening tests could be redirected to other areas of care, such as therapy or psychology.

New recommendation on how to investigate EDI

We propose a diagnostic algorithm in figure 1 based on our data. Guidelines cannot cover every clinical eventuality and should not stop clinicians ordering additional investigations where a high degree of suspicion exists about a specific condition.

Future suggestions for research projects

To truly determine which investigations are useful in making a diagnosis, a large prospective study is required in which children are phenotyped in detail and investigations are applied uniformly across the cohort. Secondary analysis could include whether the age at time of parent's first concern about their child's developmental level, the degree of developmental impairment and the pattern of domains affected affect which investigations are likely to be diagnostic. The role of newer technologies like gene exome studies and cost-efficiency analysis of investigation pathways could also be assessed.

CONCLUSIONS

Many recommended first-line investigations for EDI are rarely diagnostic and can be safely deferred without missing diagnoses. This has cost-saving implications to the NHS. We recommend a streamlined guideline based on our results, but a prospective study is required to provide robust evidence of the use of investigations according to additional features, level of developmental impairment and domains affected, which would enable the generation of an improved evidence-based guideline.

Author affiliations

¹Department of Paediatric Neurology, Sheffield Children's Hospital NHS Foundation Trust, Ryegate Children's Centre, Sheffield, UK

²Department of Paediatric Neurodisability, Sheffield Children's Hospital NHS Foundation Trust, Ryegate Children's Centre, Sheffield, UK

³Department of Paediatrics, Sheffield Children's Hospital NHS Foundation Trust, Western Bank, Sheffield, UK

⁴Department of Paediatric Neuroradiology, Sheffield Children's Hospital NHS Foundation Trust, Sheffield, UK

⁵Clinical Trials Research Unit, School of Health and Related Research, University of Sheffield, Sheffield, UK

⁶Department of Community Paediatrics, Sheffield Children's Hospital NHS Foundation Trust, Sheffield, UK

⁷Department of Clinical Genetics, Sheffield Children's Hospital NHS Foundation Trust, Sheffield, UK

Contributors ARH designed/conceived the project, collated data from medical notes and investigation software, analysed data and drafted the paper. RS, MA, SA, SS, SB, NM, AR, MJP and DJAC significantly contributed to revising project design, identified participants, reviewed notes and investigation software to determine phenotype and the contribution made to diagnosis by investigations, reviewed and commented on data analysis, reviewed drafts of the paper and agreed on the final version. JC significantly contributed to reviewing study design and recommended changes to methodology, reviewed and assisted with data analysis and interpretation, assisted in drafting the paper and agreed on the final version submitted. All authors are accountable for the integrity and accuracy of the data submitted.

Competing interests None declared.

Provenance and peer review Not commissioned; externally peer reviewed.

© Article author(s) or their employer(s) unless otherwise stated in the text of the article) 2017. All rights reserved. No commercial use is permitted unless otherwise expressly granted.

REFERENCES

- Shevell M, Ashwal S, Donley D, *et al*. Practice parameter: evaluation of the child with global developmental delay: report of the Quality Standards Subcommittee of the American Academy of Neurology and The Practice Committee of the Child Neurology Society. *Neurology* 2003;60:367–80.
- Majnemer A, Shevell MI. Diagnostic yield of the neurologic assessment of the developmentally delayed child. *J Pediatr* 1995;127:193–9.
- Horrige KA. Assessment and investigation of the child with disordered development. *Arch Dis Child Educ Pract Ed* 2011;96:9–20.
- Boyle CA, Boulet S, Schieve LA, *et al*. Trends in the prevalence of developmental disabilities in US children, 1997–2008. *Pediatrics* 2011;127:1034–42.
- Curry CJ, Stevenson RE, Aughton D, *et al*. Evaluation of mental retardation: recommendations of a Consensus Conference: American College of Medical Genetics. *Am J Med Genet* 1997;72:468–77.
- García-Cazorla A, Wolf NI, Serrano M, *et al*. Mental retardation and inborn errors of metabolism. *J Inher Metab Dis* 2009;32:597–608.
- Gillberg C. Practitioner review: physical investigations in mental retardation. *J Child Psychol Psychiatry* 1997;38:889–97.
- Battaglia A, Bianchini E, Carey JC, *et al*. Diagnostic yield of the comprehensive assessment of developmental delay/mental retardation in an institute of child neuropsychiatry. *Am J Med Genet* 1999;82:60–6.
- Gillberg C, Soderstrom H. Learning disability. *Lancet* 2003;362:811–21.
- Jain S, Chowdhury V, Juneja M, *et al*. Intellectual disability in Indian children: experience with a stratified approach for etiological diagnosis. *Indian Pediatr* 2013;50:1125–30.
- Jauhari P, Boggula R, Bhawe A, *et al*. Aetiology of intellectual disability in paediatric outpatients in Northern India. *Dev Med Child Neurol* 2011;53:167–72.
- McDonald L, Rennie A, Tolmie J, *et al*. Investigation of global developmental delay. *Arch Dis Child* 2006;91:701–5.
- Poplawski NK. Investigating intellectual disability: a genetic perspective. *J Paediatr Child Health* 2003;39:492–506.
- Shevell MI, Majnemer A, Rosenbaum P, *et al*. Profile of referrals for early childhood developmental delay to ambulatory subspecialty clinics. *J Child Neurol* 2001;16:645–50.
- van Karnebeek CD, Scheper FY, Abeling NG, *et al*. Etiology of mental retardation in children referred to a tertiary care center: a prospective study. *Am J Ment Retard* 2005;110:253–67.
- Wilksa ML, Kaski MK. Why and how to assess the aetiological diagnosis of children with intellectual disability/mental retardation and other neurodevelopmental disorders: description of the Finnish approach. *Eur J Paediatr Neurol* 2001;5:7–13.
- Hughes R, Sharma R, Connolly DJ, *et al*. Survey of UK attitudes and protocols for the investigation of early developmental impairment. *J Paediatr Child Health* 2017;53:318–20.
- Griffiths PD, Batty R, Warren D, *et al*. The use of MR imaging and spectroscopy of the brain in children investigated for developmental delay: what is the most appropriate imaging strategy? *Eur Radiol* 2011;21:1820–30.
- Makela NL, Birch PH, Friedman JM, *et al*. Parental perceived value of a diagnosis for intellectual disability (ID): a qualitative comparison of families with and without a diagnosis for their child's ID. *Am J Med Genet A* 2009;149A:2393–402.
- Wolf B. Biotinidase deficiency. In: Pagon RA, Adam MP, Ardinger HH, eds. *GeneReviews*. Seattle: University of Washington, 2000. www.ncbi.nlm.nih.gov/books/NBK1322
- Asgari A, Rouhi Dehnehab S, Zargari M, *et al*. Clinical, biochemical and genetic analysis of biotinidase deficiency in Iranian population. *Arch Iran Med* 2016;19:774–8.

- 22 Mukhopadhyay D, Das MK, Dhar S, *et al*. Multiple carboxylase deficiency (late onset) due to deficiency of biotinidase. *Indian J Dermatol* 2014;59:502–4.
- 23 Wolf B. Biotinidase deficiency should be considered in individuals exhibiting myelopathy with or without and vision loss. *Mol Genet Metab* 2015;116:113–8.
- 24 Raha S, Udani V. Biotinidase deficiency presenting as recurrent myelopathy in a 7-year-old boy and a review of the literature. *Pediatr Neurol* 2011;45:261–4.
- 25 Karimzadeh P, Ahmadabadi F, Jafari N, *et al*. Biotinidase deficiency: a reversible neurometabolic disorder (an Iranian pediatric case series). *Iran J Child Neurol* 2013;7:47–52.
- 26 Ogundele MO. Question 2. What is the incidence of biotin deficiency in preschool children with global developmental delay? *Arch Dis Child* 2011;96:895–7.
- 27 Searle AK, Baghurst PA, van Hooff M, *et al*. Tracing the long-term legacy of childhood lead exposure: a review of three decades of the port Pirie cohort study. *Neurotoxicology* 2014;43:46–56.
- 28 Hou S, Yuan L, Jin P, *et al*. A clinical study of the effects of lead poisoning on the intelligence and neurobehavioral abilities of children. *Theor Biol Med Model* 2013;10:13.
- 29 Chandramouli K, Steer CD, Ellis M, *et al*. Effects of early childhood lead exposure on academic performance and behaviour of school age children. *Arch Dis Child* 2009;94:844–8.
- 30 Pirkle JL, Brody DJ, Gunter EW, *et al*. The decline in blood lead levels in the United States. The National Health and Nutrition Examination Surveys (NHANES). *JAMA* 1994;272:284–91.
- 31 Centers for Disease Control and Prevention (CDC). Blood lead levels in children aged 1–5 years—United States, 1999–2010. *MMWR* 2013;61:245–8.
- 32 Hart AR, Batty R, Widjaja E, *et al*. MRI in children with global developmental delay—a retrospective case note review. *J Paediatr Neurology* 2011;9:15–21.