

ORIGINAL ARTICLE

High prevalence of food sensitisation in young children with liver disease: a clue to food allergy pathogenesis?

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Abstract

Background: The pathogenesis of food allergy is not completely understood – animal models suggest hepatic mechanisms may be important for immune tolerance to orally ingested antigens, but there is little direct evidence for this in humans.

Objectives: We investigated whether there is an association between liver dysfunction or transplantation in young children and IgE sensitisation to food.

Methods: We evaluated paired pre- and post- liver transplant sera from children aged 0–36 months treated at a single centre during 2001–2008. Sera were assayed for total IgE and cow's milk, egg and peanut-specific IgE. We quantified hepatic dysfunction pre-transplant using the Paediatric End-stage Liver Disease (PELD) score. We also assessed 70 children after renal transplant to establish whether any association between liver transplant and food sensitisation was organ specific.

Results: Paired sera were available from 50 of 94 children who had a liver transplant during the study period. 35 of 50 (70%) had IgE sensitisation (≥ 0.35 kUa/l) to ≥ 1 food pre-transplant and 18 (36%) post-transplant ($p = 0.001$). Ten (20%) children had food-specific IgE levels that carry high probability of challenge-confirmed food allergy pre-transplant. Food sensitisation pre-transplant was associated with severity of liver dysfunction [mean (s.d.) pre-transplant PELD score 1.52 (0.13) in food sensitised, 0.77 (0.22) in non-sensitised children $p = 0.004$]. Total IgE level was raised in 34/42 (81%) pre-transplant and fell significantly post-transplant. Interview assessment of the parents of 40 children revealed that 13 (33%) had a history consistent with food allergy. These findings were not replicated in the renal transplant group.

Conclusions: Young children with severe liver dysfunction appear to have a high prevalence of food sensitisation. Hepatic mechanisms may therefore be important for establishing immune tolerance to dietary antigens in humans.

Food allergy affects 4–5% of children and is the commonest single cause of anaphylaxis (1, 2). Food allergy appears to be increasing in prevalence for unknown reasons and can have serious adverse effects on quality of life and nutritional status (3–5). Our understanding of food allergy pathogenesis in humans is limited, with the most widely used paradigms derived from animal models of oral tolerance. Although these models have demonstrated for the past 40 yrs that hepatic mechanisms are important for immune tolerance to orally ingested antigens (6–8), a role for the liver in human food allergy pathogenesis has not been widely explored. Food allergy has, however, been described in a number of case series of young children undergoing liver transplantation, with 6–57% developing *de novo* food allergy post-transplant (9–13). It

is remarkable that this phenomenon is almost exclusive to young children (age <3 yrs), when the majority of liver transplants are undertaken in adults. Contrary to the reports of food allergy transfer from allergic organ donors to adult liver transplant recipients (14), the emergence of food allergy in children appears to occur in the absence of donor food allergy. This suggests that either liver dysfunction or liver transplant during a critical early life period prevents the establishment of immune tolerance to orally ingested food antigens. However, it remains unclear whether food sensitisation or allergy is truly over-represented in children undergoing liver transplantation, and if so whether this is a new phenomenon following transplantation or related to the pre-existing liver dysfunction. We used an established serum bank from children undergoing

liver transplant, to investigate the nature of any association between early liver transplantation and food sensitisation.

Methods

Study period and population

We analysed serum stored prospectively from a cohort of young children undergoing liver transplantation, and where possible we interviewed their parent(s) regarding allergy symptoms and risk factors for allergy. The study was designed to identify the prevalence of food sensitisation pre- and post-transplant in young children and to determine risk factors for this association. Children who underwent orthotopic liver transplantation prior to their 3rd birthday at King's College Hospital, London (KCH), between January 2001 and July 2008 were studied. The study was approved by King's College Hospital Research Ethics Committee (Reference 09/H0808/37), and all parents gave informed consent. Sera taken prior to and subsequent to transplantation were stored from all children undergoing liver transplantation during the study period. Children were excluded from the study if paired pre- and post-transplant serum samples were not available – pre-transplant taken within 90 days prior to transplantation; post-transplant taken 365+/-90 days after transplantation. The age at transplant, underlying diagnosis, immunosuppressive regimen, episodes of allograft rejection, International Normalised Ratio (INR), serum albumin, bilirubin levels and peripheral blood eosinophil counts pre-transplant were recorded for each child. Liver dysfunction was quantified using a modified Paediatric End-stage Liver Disease (PELD) score, calculated as $0.480 (\log_e \text{bilirubin mg/dl}) + 1.857 (\log_e \text{INR}) - 0.687 (\log_e \text{albumin g/dl})$ (15).

To substantiate an association between liver transplantation or liver disease and food allergy, we also analysed serum samples from a comparator group of children who had undergone renal transplantation. Patients were recruited from the Renal Transplant Clinic at Great Ormond Street Hospital for Children NHS Trust between January and April 2011. Serological analysis of blood samples taken on the day of recruitment was performed for the same food allergens tested in the liver cohort, and an interview-based assessment of allergic status was performed, as detailed below. Any child who had also undergone liver transplantation was excluded. The age at transplant and assessment, primary diagnosis, immunosuppression regimen, transplant type, creatinine and peripheral blood eosinophil counts post-transplant were recorded, and renal dysfunction was quantified using estimated GFR (eGFR), calculated as $\text{height (cm)} \times 33/\text{plasma creatinine}$.

Serological assessment of food sensitisation

Sera were evaluated for total IgE and specific IgE to cow's milk, egg and peanut because approximately 90% of reported cases of food allergy following liver transplant involve at least one of these three foods. Serum samples were frozen at -80°C until batched analysis (lower limit of detection 0.01 kUa/l; Immunocap, Phadia, Sweden). Where sample volume was

limited, specific IgE measurements were prioritised over total IgE. Food sensitisation was defined as food-specific IgE ≥ 0.35 kUa/l and high-level food sensitisation as food-specific IgE level greater than published age-specific levels with 95% positive predictive value for challenge-confirmed food allergy, namely for cow's milk 15 kUa/l (5 kUa/l at <1 yr), egg 7 kUa/l (2 kUa/l at <2 yrs) and peanut 14 kUa/l (16).

Interview-based assessment of allergic status

The parents of all children with paired sera available for analysis were invited to an interview to determine prevalence of food allergic reactions, other allergic disease and known risk factors for allergic disease. A structured questionnaire was administered to these parents and their children between March and June 2010. The food allergy questionnaire was modified from those used in the EuroPrevall birth cohort studies with kind permission from the investigators and piloted in a paediatric allergy clinic to confirm that a complete food allergy history was comprehensively recorded (17). Determination of other allergic diseases eczema, rhinitis and asthma was made using questionnaires modified from the ISAAC study (18). Data regarding infant diet, medications and known risk factors for allergic disease were acquired at interview, and hospital records held at KCH were reviewed. All clinical histories were presented to a Paediatric Allergist (CG) blinded to the purpose of the study, who used the history and IgE results to evaluate whether a food allergic reaction had occurred in accordance with international guidelines (19).

Statistical analysis

Proportional differences were tested with chi-square tests (Pearson for unpaired or McNemar Bowker test for paired data) or Fisher's exact test. Wilcoxon test was used for non-parametric paired data, and Mann-Whitney *U*-test for non-parametric unpaired comparisons. Parametric data were analysed using either paired or independent sample *t*-test. Total and food-specific IgE levels were $\log_{10}$ -transformed; normality was tested with Kolmogorov-Smirnov test. Correlations between parametric variables were determined using Pearson's correlation coefficient. All reported *p* values are two-tailed. Analyses were performed with PASW software, v18.

Results

Ninety-four children aged <3 yrs underwent liver transplant at KCH during the study period. Forty-four (47%) patients were excluded because they did not have paired serum samples of sufficient volume stored from within the study timeframe. Paired sera were analysed for 50 children, and the parents of 40 of these consented to an interview. Median (Interquartile Range, IQR) age at transplant was 12.1 (7.9, 20.6) months. The indications for transplant were biliary atresia (31; 62%), metabolic liver disease (7; 14%), cryptogenic neonatal hepatitis (5; 10%), acute liver failure ($n = 3$; 6%), Alagille syndrome ($n = 2$; 4%), neonatal liver failure ($n = 1$; 2%) and hepatoblastoma ($n = 1$; 2%). The metabolic liver diseases were

alpha-1-anti-trypsin deficiency ($n = 3$), progressive familial intrahepatic cholestasis, bile acid synthesis disorder, glycogen storage disease type IV and carbamoyl phosphate synthetase deficiency. There was a similar age distribution and pattern of primary diagnosis in children with sufficient sera available, those whose parents consented to interview and those without serum available (Table S1).

Prevalence of food sensitisation in young children with severe liver dysfunction

Thirty-seven (74%) of the 50 study children had IgE sensitisation >0.35 kUa/l to ≥ 1 food of cow's milk, egg or peanut either pre- or post-liver transplant. Fourteen (38%) of these 37 had food-specific IgE levels greater than published age-specific levels with high predictive value for challenge-confirmed food allergy (13). Thirty-five of 50 (70%) had IgE sensitisation (≥ 0.35 kUa/l) to ≥ 1 food pre-transplant and 18 (36%) post-transplant ($p = 0.001$). Ten (20%) children had food-specific IgE levels that carry high probability of challenge-confirmed food allergy pre-transplant.

Relationship between food sensitisation and liver transplant

Thirty-five (95%) of the 37 children who were sensitised at either time point were sensitised prior to transplantation, with only two patients showing evidence of new onset food sensitisation post-transplantation. Total IgE levels were raised (>10 kUa/l) prior to transplant in 34 (81%) of the 42 children with sufficient serum available to analyse this. Both milk-specific and total IgE levels fell significantly post-transplant, but egg and peanut IgE were unchanged (Table 1). All patients had peripheral blood eosinophil counts within normal limits prior to transplant, with no significant change post-transplant (Table 1).

Relationship between severity of liver dysfunction and food sensitisation

Children with pre-transplant food sensitisation had more severe liver disease than those without – mean (s.d.) modified PELD score 1.52 (0.13) food sensitised vs. 0.77 (0.22) not sensitised, $p = 0.004$ (Fig. 1a). There was also a significant positive correlation between total IgE level and modified PELD score [$r = 0.42$, (95% CI, 0.14, 0.64), $p = 0.01$; Fig. 1b].

Risk factors for food sensitisation

Genetic predisposition to allergy was assessed by recording family history of allergic disease and other allergy risk factors for 40 of the 50 children with paired sera, whose parent agreed to undergo interview. We did not identify any specific risk factor for food sensitisation in these patients, and overall the rates of positive family history for allergic disease were no higher than the general population, but breast feeding rates were low with early introduction of cow's milk formula in most children. (Table S2). The small number of patients in each group meant that there was insufficient statistical power to

Table 1 Total and specific IgE pre- and post-transplant

	Pre-transplant	Post-transplant	p value
Total IgE median (IQR; $n = 42$)	97.5 (22.3, 432.0)	15.0 (4.0, 105.5)	0.001
Eosinophil count $\times 10^9/l$	0.22 (0.10, 0.46)	0.23 (0.07, 0.52)	0.92
Milk IgE median (IQR)	0.83 (0.24, 2.74)	0.12 (0.01, 0.45)	<0.001
Milk sensitised – no. (%)	32 (64)	17 (34)	0.001
High-level milk sensitisation – no. (%)	3 (6)	1 (2)	0.63
Egg IgE median (IQR)	0.01 (0.01, 0.31)	0.01 (0.01, 0.24)	0.41
Egg sensitised – no. (%)	11 (22)	10 (20)	1.0
High-level egg sensitisation – no. (%)	7 (14)	7 (14)	1.0
Peanut IgE median (IQR)	0.01 (0.01, 0.13)	0.01 (0.01, 0.02)	0.10
Peanut sensitised – no. (%)	8 (16)	7 (14)	1.0
High-level peanut sensitisation – no. (%)	0 (0)	1 (2)	–

Sensitised, food-specific IgE ≥ 0.35 kUa/l to milk, egg or peanut; High-level food sensitisation, food-specific IgE level greater than published age-specific levels with 95% positive predictive value for challenge-confirmed food allergy (13).

McNemar Bowker test was used for categorical data, Wilcoxon for continuous data. Total/specific IgE values are kUa/l.

provide a conclusive analysis of risk factors for food sensitisation in this population.

Clinical history of allergic disease

Interview assessments of allergy history were obtained for 40 of the 50 children with paired sera available, undertaken a mean of 4.88 (s.d. 2.48) yrs after transplant. Twenty (50%) had a history of eczema, 9 (23%) of wheezing and 9 (23%) of allergic rhinitis. Twenty-eight (70%) of the 40 children whose parents were interviewed had raised food-specific IgE level to ≥ 1 of egg, peanut or cow's milk prior to transplantation, two with 'high-level sensitisation'. Thirteen (33%) of the 40 were reported to have a clinical history of food allergy. Twelve of these 13 had onset of symptoms post-transplant, 10 of whom had not knowingly ingested the relevant food pre-transplant. Clinical and laboratory features of the children with a history of food allergic reaction are shown in Table 2. A clinical history of food allergy was not associated with tacrolimus immunosuppression, nor with different EBV or CMV infection rate compared with those without food allergy (data not shown).

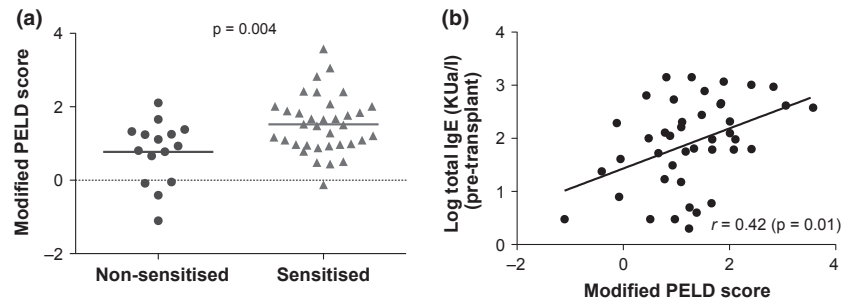


Figure 1 Relationship between liver dysfunction and IgE sensitisation. Panel a shows modified Paediatric End-stage Liver Disease (PELD) scores in patients with and without specific IgE ≥ 0.35 kUa/l to ≥ 1 of egg, cow's milk or peanut pre-transplant. Bars represent mean values. Panel b shows relationship between pre-transplant Total IgE levels and modified PELD score.

IgE sensitisation and clinical food allergy in renal transplant recipients

Seventy renal transplant recipients were recruited from Great Ormond Street Hospital for Sick Children, London, to establish whether food sensitisation or allergy was over-represented in this group. Median age at assessment was 161 months, and children were a median 37 months post-transplant at the time of assessment (Table S3). Eleven (16%) of the 70 children had IgE sensitisation >0.35 kUa/l to ≥ 1 food of cow's milk, egg or peanut, at the time of assessment, and 4 (6%) had 'high-level sensitisation' (16). Food sensitisation was significantly less common after renal transplant than after liver transplant – 11/70 (16%) renal, 18/50 (36%) liver, $p = 0.01$. A clinical history suggestive of food allergy was significantly less common in renal transplant than in liver transplant recipients – 8/70 (11%) renal, 13/40 (33%) liver, $p = 0.007$. In six of the eight food allergic children, food allergy symptoms first occurred prior to renal transplant. The prevalence of raised total IgE (>10 kUa/l) post-transplantation was lower in renal transplant than liver transplant recipients – 15/70 (21%) renal, 29/45 (64%) liver, $p < 0.001$. Severity of renal dysfunction was not significantly different in sensitised compared with that in non-sensitised children – median (IQR) eGFR 65.3 (46.3, 95.3) ml/min/1.73 m² sensitised; 61.9 (48.8, 79.3) non-sensitised; $p = 0.65$. There was no significant correlation between eGFR and total IgE levels; $r = 0.04$, $p = 0.75$. Further details of the children in receipt of a renal transplant are shown in Table S3.

Discussion

The occurrence of food allergy in young children undergoing liver transplantation is a poorly understood phenomenon. In this study, we confirmed a high prevalence of food sensitisation in this setting and found significantly raised total IgE levels that correlate with severity of liver dysfunction. Most importantly, we found that although food allergic reactions typically first occur after transplant, IgE sensitisation to foods and raised total IgE are commonly present before transplant. Indeed, both total and specific IgE fell significantly 1 yr after transplant. Together, these data suggest that abnormal liver

function during early life may be a risk factor for food sensitisation and allergy.

IgE sensitisation to food was present in 74% of our population, which compares with 6–22% in contemporaneous European cohort studies and 16% in the children we studied who had undergone renal transplantation (20, 21). Known risk factors for food sensitisation were not over-represented in this group of patients at the time of transplant, other than a high prevalence of early cow's milk formula feeding – however, some investigators have found early cow's milk feeding to be protective for cow's milk allergy (22), and the high prevalence of food sensitisation was not restricted to cow's milk. Children with IgE sensitisation were younger than non-sensitised children at the time of transplant, although this difference was not statistically significant. This is of note as previous series describing food allergy in children post-liver transplant have also found the highest rates in infancy (23, 24).

It is well established in animal models that delivery of antigen via the hepatic portal vein results in immune tolerance to that antigen (7). The importance of hepatic mechanisms in immune tolerance has also been demonstrated by surgical experiments in which diversion of blood via a porto-caval shunt abrogates tolerance to oral antigens (6–8). However, the importance of hepatic mechanisms for oral tolerance in humans has been uncertain. The association in our study between liver dysfunction and food sensitisation may reflect a failure of normal oral tolerance mechanisms, either through increased porto-caval shunting or due to inflammatory changes in the diseased liver or related sites (25, 26).

An IgE-mediated immune response to food antigens requires IL-4 secreting Th2 cells (27). Intra-hepatic mast cells are a histopathological feature of a number of chronic liver diseases including biliary atresia (28). Antigen presentation by hepatic antigen presenting cells in the context of mast cell-derived IL-4 might drive Th2 differentiation in young children with liver disease. Elevated serum IgE has been reported in other forms of chronic liver disease such as active type A and B hepatitis in adults (29, 30); however, food-specific IgE and food allergy have not been reported as increased in those diseases. We found increased serum IgE in early onset liver dysfunction correlated with the degree of liver dysfunction. The fall in total IgE post-transplant may be related to recovery of liver function

Table 2 Clinical and laboratory features of those children reported to have clinical food allergy

Food (s)	Time of first reaction	Liver disorder	IgE Pre-transplant	IgE Post-transplant	Eczema	Age at transplant (month)	Symptoms
Egg	Post-transplant	Neonatal liver failure	Total 126 Milk 2.68; Egg 0.01 Peanut 0.01	Total 2263 Milk 0.42; Egg 9.7 Peanut 1.06	Yes	2.6	Angioedema 5 min after eating a mouthful of scrambled egg aged 1 yr
Egg	Post-transplant	Acute liver failure	Total 62 Milk 4.55; Egg 0.01 Peanut 0.01	Total 2197 Milk 9.24; Egg 84.2 Peanut 3.07	Yes	2.7	Several episodes of acute vomiting within 5 min of ingesting egg-containing foods aged 1 yr
Fish, Peanut, Cashew, Walnut, Mango	Post-transplant	EHBA	Total 438 Milk 1.18; Egg 0.16 Peanut 0.03	Total 0 Milk 0.01; Egg 0.01 Peanut 0.01	Yes	6.4	Eczema flare on many occasions after eating fish, peanut, cashew nut, walnut, mango aged 3 yrs
Egg, Fish, Peanut, Cashew	Post-transplant	EHBA	Total 204 Milk 0.28; Egg 0.01 Peanut 0.01	Total 6930 Milk 28.1; Egg 8.78 Peanut 76.9	Yes	6.6	Angioedema/pruritis on several occasions after eating fish, peanut, cashew nut and egg from aged 2 yrs
Cow's milk	Post-transplant	EHBA	Total 197 Milk 12.3; Egg 0.01 Peanut 0.01	Total 12 Milk 0.59; Egg 0.01 Peanut 0.01	Yes	6.8	Angioedema, erythema within 5 min of ingesting one mouthful of cow's milk aged 2 yrs
Egg, Soya, Hazelnut	Post-transplant	EHBA	Total N/A Milk 3.3; Egg 0.01 Peanut 0.01	Total 19 Milk 0.61; Egg 2.75 Peanut 0.01	Yes	8.6	Immediate type hypersensitivity reaction to egg aged 4 yrs. Others diagnosed on IgE tests
Egg	Post-transplant	EHBA	Total 1170 Milk 1.92; Egg 0.33 Peanut 0.12	Total 74 Milk 0.14; Egg 0.01 Peanut 0.01	Yes	10.4	Urticaria, angioedema, 10 min after eating scrambled egg aged 1 yr
Sesame	Post-transplant	EHBA	Total 6 Milk 0.2; Egg 0.01 Peanut 0.01	Total 2 Milk 0.08; Egg 0.02 Peanut 0.01	Yes	11.1	Angioedema within 5 min of eating hummus aged 2 yrs
Peanut	Post-transplant	EHBA	Total 940 Milk 2.28; Egg 0.01 Peanut 1.21	Total 173 Milk 0.01; Egg 0.01 Peanut 0.01	No	11.4	Vomited within 5 min of eating a spoon of peanut butter aged 2 yrs
Soya, Egg, Cow's milk	Post-transplant	Acute liver failure	Total 52 Milk 0.04; Egg 0.01 Peanut 0.01	Total 33 Milk 0.01; Egg 0.01 Peanut 0.01	Yes	12.0	Enteropathy with eosinophilic colitis diagnosed aged 2 yrs. Symptoms improved after exclusion of milk
Egg, Prawn	Post-transplant	EHBA	Total 1427 Milk 10.7; Egg 14.1 Peanut 4.23	Total 384 Milk 8.72; Egg 42.8 Peanut 5.27	No	14.5	Vomited within 5 min of eating egg on three occasions; urticaria

Table 2 (Continued)

Food (s)	Time of first reaction	Liver disorder	IgE Pre-transplant	IgE Post-transplant	Eczema	Age at transplant (month)	Symptoms
Banana	Pre-transplant	EHBA	Total 56 Milk 0.70; Egg 0.38 Peanut 0.03	Total 12 Milk 0.09; Egg 0.04 Peanut 0.01	No	23.0	twice after eating prawns aged 2 yrs Abdominal pain and diarrhoea 30 min after eating banana, on several occasions from age 5 months
Peanut	Post-transplant	Hepato-blastoma	Total 3 Milk 0.44; Egg 0.04 Peanut 0.01	Total 4 Milk 0.03; Egg 0.01 Peanut 0.01	Yes	25.4	Vomited within 10 min of eating peanut aged 3 yrs

EHBA, extrahepatic biliary atresia.

Specific IgE levels (kUa/l) were available for cow's milk, egg and peanut from pre- and 1 yr post-transplant, but not for other food allergens.

or due to post-transplant immunosuppression. The fall in cow's milk-specific IgE may be part of the same phenomenon or alternatively may reflect the natural history of cow's milk allergy where a fall in specific IgE and clinical reactivity is seen over time. The same fall was not seen in egg- and peanut-specific IgE, however, which may reflect the lower frequency with which these food allergies spontaneously resolve compared with cow's milk allergy, or perhaps the lower frequency of exposure and sensitisation to egg and peanut pre-transplant. It is unlikely that the fall in total or cow's milk-specific IgE levels relate to tailored dietary management of food allergies in these patients, because there was not a specific management strategy in this group, which differed from other food allergic populations.

In our study, liver transplantation was not associated with increased levels of IgE sensitisation to foods, but it was associated with onset of clinical reactivity to foods. A number of case series of liver transplant-associated food allergy have reported elevations in total and specific IgE and clinical food allergic reactions post-transplantation (9–11, 13, 24); however, only one of these series investigated pre-transplant levels of specific IgE. In this study (24), pre-transplant total IgE levels were elevated in two of six children who developed food allergy with milk-specific IgE detected pre-transplant in one of the six patients. In 5 of the 22 non-food allergic patients, total IgE levels were also elevated pre-transplant; however, specific IgE levels in this group were not described. Our findings suggest that total and food-specific IgE rise with early onset liver disease and, in the majority of children, decrease after transplantation. Clinical reactivity to foods usually first occurs after transplantation, which is in keeping with previous case series. The delay in clinical manifestations of food allergy until the post-transplant period remains unexplained. This may be due to restricted intake of relevant food allergens pre-transplant, or to suppression of clinical allergic disease in those with severe liver dysfunction. Impaired immune response in chronic liver disease is a well-recognised phenomenon (31, 32). Previous reports of food allergy associated with liver

transplant in young children have suggested that tacrolimus effects (immune or intestinal) may increase the risk of food allergy. Our data suggest that the process of food sensitisation in these patients may be more closely related to liver dysfunction than to transplantation or to a specific immunosuppressant.

An important limitation of our study is that IgE food sensitisation does not always reflect clinical reactivity to the food, and the gold standard for diagnosis of food allergy is a double blind placebo-controlled food challenge. Although both serological and clinical data sets suggested that these patients have a high prevalence of food allergy, results for individual patients were inconsistent (Table 2). In some cases, specific IgE was not detected at a significant level despite a history of food allergic reaction to the relevant food, and in some children with a clinical history of food allergy, there was a significant post-transplant rise in specific IgE to the relevant food – suggesting that food sensitisation may develop or worsen after transplant in selected cases. Only 2 of the 10 patients with high-level food sensitisation pre-transplant attended for interview, which may account for some of the inconsistency between serological and clinical data sets. For ethical reasons, it was not possible to reconcile these differences by undertaking blinded food challenges in this population of 50 sick young children.

A further limitation of the study is the absence of an age-matched control group without liver dysfunction; however, analysis of paediatric renal transplant recipients demonstrated the levels of IgE sensitisation and clinical allergy in keeping with the normal population. Although the liver transplant and renal transplant populations were not matched for age, the prevalence of raised IgE, food sensitisation and clinical food allergy was significantly higher in children after liver transplant than in children after renal transplant. Some of the renal transplant recipients had elevated eGFR and thus chronic kidney disease; however, we found no association between renal impairment and food sensitisation or elevated IgE level. This suggests that transplantation of other solid organs and other chronic diseases are unlikely to be risk factors for food

allergy development in the same way as early onset liver disease.

Finally, this study must be considered preliminary due to the relatively small numbers of subjects included and the significant number of children without serum available to analyse. We found no evidence that those without serum available differed from the included subjects, but cannot exclude the possibility that those excluded from the study had less food allergy than those with serum available for analysis.

In summary, we observed a strikingly high prevalence of IgE sensitisation to foods in a subset of children with early onset liver dysfunction requiring transplantation. Liver transplant did not increase the risk of food sensitisation in our study, but clinical reactions were almost always delayed until after transplant – for unknown reasons. These results suggest a potentially important link between severe liver disease and a failure to establish tolerance to dietary antigens, and this may be significant for our understanding of food allergy. Raised levels of hepatic enzymes and hepatic steatosis are relatively common in early infancy (33, 34) – the possibility that subclinical hepatic dysfunction in infancy is a risk factor for food sensitisation needs to be explored in future studies.

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Contributors

CB contributed to the design of this study, prepared and checked clinical coding, undertook most of the data analysis, wrote the first and contributed to subsequent drafts. NH

developed the structured questionnaire, undertook interview assessments and reviewed KCH records for participants. CD undertook interview assessments of renal transplant recipients and contributed to the data analysis. CG evaluated questionnaire responses. GM-V established the serum bank and contributed to study design. DV contributed to study design. MH managed the serum bank and identified samples for analysis. JW contributed to study design. SM contributed to the study design and data analysis regarding renal transplant recipients. RB developed the idea, contributed to the study design and data analysis and wrote the final manuscript. All authors approved the final version.

Conflicts of interest

CB, CD, NH, CB, CG, MH, GM-V, DV and SM declare that they have no conflicts of interest. JW and RB have received grant support from Danone, Airsonett and Allergy Therapeutics, and RB has received grant support from Lincoln Medical. JW is a member of scientific advisory boards and has given paid lectures for Danone, Airsonett, Merck, Novartis, UCB pharma, Mead Johnson, Allergy Therapeutics.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Table S1. Baseline characteristics of patients at time of liver transplant.

Table S2. Risk factors for food sensitisation in children with early onset liver disease.

Table S3. Characteristics of renal transplant patients.

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Table S1. Baseline characteristics of patients at time of liver transplant

Patient Characteristic	Serological study	*Clinical assessment	†Excluded
	n=50	n=40	n=44
Age at Transplant (months) median (IQR)	12.1 (7.9, 20.6)	11.1 (7.9, 17.3)	13.2 (9.1, 18.9)
Primary Diagnosis			
Biliary Atresia – no. (%)	31 (62)	25 (63)	22 (59)
‡Metabolic Liver Disease – no. (%)	7 (14)	4 (10)	6 (16)
Other – no. (%)	12 (24)	11 (28)	9 (25)

*Clinical assessment was successfully undertaken on 40 of 50 children with serum available.

†Excluded patients are children with insufficient serum available for the serological study; primary diagnosis was available for 37 of these

‡alpha-1-anti-trypsin deficiency, progressive familial intrahepatic cholestasis, bile acid synthesis disorder, glycogen storage disease type IV, carbamoyl phosphate synthetase deficiency are included within this group.

Table S2. Risk factors for food sensitisation in children with early onset liver disease

	Food Sensitised	Not Food Sensitised	p value
	(n=28)	(n=12)	
Age at transplant (months) – median (IQR)	9.4 (6.8, 17.2)	16.4 (6.6, 20.9)	0.12
Parent with asthma – no. (%)	5 (18)	3 (25)	0.61
Parent with eczema – no. (%)	2 (7)	1 (8)	0.90
Parental allergic rhinitis – no. (%)	7 (25)	2 (17)	0.56
Parental food allergy – no. (%)	1 (4)	1 (8)	0.52
Sibling asthma – no. (%)	4 (14)	1 (8)	0.60
Sibling eczema – no. (%)	5 (18)	2 (17)	0.93
Sibling allergic rhinitis – no. (%)	2 (7)	2 (17)	0.36
Sibling food allergy – no. (%)	1 (4)	2 (17)	0.15

Gestation in weeks median (IQR)	40.0 (40.0, 40.0)	39.5 (39.0, 40.8)	0.29
Caesarean delivery no. (%)	7 (25)	3 (25)	1.00
Birthweight in kg median (IQR)	3.46 (2.66, 3.70)	2.83 (2.68, 3.39)	0.26
Breast fed – no. (%)	20 (71)	8 (67)	0.76
Breastfed duration in weeks median (IQR)	2.3 (0.0, 10.5)	1.0 (0.0, 3.6)	0.10
Age at solid introduction in months median (IQR)	6.0 (4.5, 8.0)	5.5 (4.1, 8.1)	0.94
No. older siblings median (IQR)	1.0 (0.0, 2.0)	0.5 (0.0, 1.5)	0.43
Pet in home – no. (%)	8 (28.6)	3 (25.0)	0.82

Data are from the subset of 40 children whose parents consented to undergo interview.

Mann-Whitney U test was used for continuous variables, and χ^2 for categorical variables

Table S3. Characteristics of renal transplant patients

Patient Characteristic	
Age at Transplant (months) – median (IQR)	90.5 (51.8, 144.3)
Age at Assessment (months) – median (IQR)	161.0 (110.3, 183.8)
Months post-transplant at assessment –median (IQR)	37.0 (14.0, 71.0)
Primary Diagnosis	
Nephrotic Syndrome – n (%)	14 (20)
CAKUT – n (%)	32 (46)
*Other – n (%)	24 (34)
Type of Transplant	
Living Donor – n (%)	31 (44)
Deceased Donor – n (%)	39 (56)
Total IgE – median (IQR)	16.5 (5.0, 56.8)

Milk IgE – median (IQR)	0.01 (0.01, 0.01)
Milk sensitised – n (%)	6 (9)
High level milk sensitisation – n (%)	0 (0)
Egg IgE – median (IQR)	0.01 (0.01, 0.04)
Egg sensitised – n (%)	9 (13)
High level egg sensitisation – n(%)	4 (6)
Peanut IgE – median (IQR)	0.01 (0.01, 0.02)
Peanut sensitised – n (%)	7 (10)
High level peanut sensitised – n(%)	1 (1)
Eosinophil count x 10 ⁹ /L - median (IQR)	0.13 (0.07, 0.26)
Immunosuppression – n (%)	
Tacrolimus	60 (86)
Ciclosporin	1 (1)
Mycophenolate Mofetil	33 (47)

Prednisolone	59 (84)
Azathioprine	15 (21)

CAKUT, Congenital anomalies of the kidney and urinary tract. IQR, interquartile range.

* Other diagnoses were: nephronophthisis (7), neonatal renal failure (3), renal venous thrombosis, cystinosis (2), Joubert syndrome, polycystic kidney disease, Wilms tumour, haemolytic uremic syndrome, cortical necrosis, tubulointerstitial disease, renal failure of unknown aetiology (5).