ORIGINAL ARTICLE

Morbidity in Alagille Syndrome in 6 Malaysian Children

C B Lim, FRCPCH, Y S Choy, MRCP

Institute of Paediatrics, Kuala Lumpur Hospital, Jalan Pahang, 50586 Kuala Lumpur

Summary

We retrospectively studied the records of 6 Malaysian children who were diagnosed with Alagille Syndrome (AGS) according to this criteria from January 1999 to January 2001, at the Institute of Paediatrics, Kuala Lumpur Hospital. Four patients (66%) had a positive family history. Thirteen individuals (6 patients and 7 relatives) were diagnosed with AGS in these 5 families. Only 6/13 (46%) of them presented with liver involvement. All 6 patients presented with typical facies and cholestasis (100%). Three (50%) presented with portal hypertension (PHT) with synthetic liver dysfunction (1 died), 1/6 (17%) have PHT and normal synthetic liver function. Two have cleared their jaundice but have biochemical evidence of hepatitis and hepatomegaly, four have congenital heart disease, 5/6 posterior embryotoxon, 2/6 butterfly vertebrae, 4/6 hyperlipidaemia and 4/6 failure to thrive. One patient has a Jagged-1 gene disruption at the translocation breakpoint locus 20p12.3 2n=46,XX,t(12,20) (q22, p12.3). 5/6 (83%) are still alive. Two-thirds of our patients developed chronic liver disease by 3 years of age. Two-thirds of the index patients have a family history. Only 46% of individuals in these families have clinical evidence of liver involvement. Mortality depends on cardiac/renal disease, end-stage liver failure and intercurrent infection.

Key Words: Alagille, Children, Malaysian

Introduction

Alagille syndrome (AGS) or arteriohepatic dysplasia is an autosomal dominant disorder with a variable penetrance and expressitivity. It is caused by the mutation of the Jagged-1 gene encoding a ligand for Notch 1 protein at chromosome 20p[1]. The classic syndrome is characterised by five major features[2]; chronic cholestasis, congenital heart disease (usually peripheral pulmonary stenosis), skeletal defects (e.g. butterfly vertebrae), eye findings (e.g. posterior embryotoxon) and a typical facies with frontal bossing, deep set eyes, hypertelorism, straight nose and a small pointed chin. Other features include renal anomalies, failure to thrive, delayed puberty, mental retardation, neurological disease, abnormal high-pitched cry/voice, vascular anomalies, recurrent chest infections and otitis media. However, not all patients display all the classic features, making clinical diagnosis difficult. A revised list of diagnostic criteria has recently been proposed by Picolli DA (Table I)[3]. In this revised criteria, a proband with the absence of family history requires presence of 3-4 major features for diagnosis unless the gene defect is detected when only 1 or more features are required. With the presence of family history, 1 or more features will make the diagnosis, unless the gene defect is detected when no or any (minor) features are required.

AGS has not been previously described in Malaysia. We are presenting our retrospective findings in 6 Malaysian children diagnosed with AGS according to this revised diagnostic criteria.

This article was accepted: 7 April 2003

Corresponding Author: Lim Chooi Bee, Consultant Paediatrician (Gastro/Hepatology), Institute of Paediatrics, Hospital Kuala Lumpur, Jalan Pahang, 50586 Kuala Lumpur.

Materials and Methods

We retrospectively studied the records of 6 Malaysian children who were diagnosed with AGS according to this criteria (Table I) from January 1999 to January 2001, at the Institute of Paediatrics, Kuala Lumpur Hospital. In this revised criteria, a proband with the absence of family history requires presence of 3-4 major features for diagnosis unless the gene defect is detected when only 1 or more features are required. With the presence of family history, 1 or more features will make the diagnosis, unless the gene defect is detected when no or any (minor) features are required.

Chromosome analysis was performed on metaphase cells derived from peripheral blood cultures. The cells were cultured with phytohaemagglutinin (PHA) for 72h. For synchronised culture, the thymidine was added on the second day and deoxycytidine (DOC) was then introduced after another 18-20h incubation. The cultures were then harvested[3]. Metaphase cells were Q-banded and karyotyes were described according to the 1995 International System for Human Cytogenetic Nomenclature[4].

Results

The clinical findings for the 6 patients are summarised in Table II and III. A total of 6 patients were identified as AGS. There were 2 males and 4 females; the male to female ratio is 1:2. All were Malays except for one Indian girl (patient 4). Ages ranged from 2 months to 5 years at presentation. 4/6 patients (66%) had a positive family history (with a pair of brother-sister i.e. patient 2 and 3). In fact, there were 13 individuals (6 patients and 7 relatives) diagnosed with AGS in these 5 families. Only 6/13 (46%) of them presented with liver involvement.

All 6 patients presented with typical AGS facies and cholestasis (100%). Three (patients 1,2 and 6) presented with portal hypertension (PHT) and synthetic liver dysfunction. PHT was described in these patients with evidence of spider naevi (patient 2), clubbing (patient 2), wasting (patient 2 and 6), hepatosplenomegaly (patient 1, 2 and 6), ascites (patient 2 and 6) or caput medusae(patient 1)[2]. However, none of the patients developed gastrointestinal bleeding and hence no endoscopy was performed. Synthetic liver dysfunction was described in these patients with significant hypoalbuminaemia with serum albumin below 35g/l (patient 2 and 6) and significant prolonged prothrombin time (PT) of more than 15s) (patient 1, 2 and 6)[2]. One of them has died (patient 2). One patient (patient 3) have PHT and normal synthetic liver function. Two patients (patients 4 and 5) have cleared their jaundice but have biochemical evidence of hepatitis and hepatomegaly.

Four patients have evidence of congenital structural cardiac abnormality. These included an atrial septal defect (ASD) with mild pulmonary stenosis (PS), a patent ductus arteriosus (PDA)/ventricular septal defect (VSD), an ASD/VSD/overriding aorta and non-confluent pulmonary artery (PA) and a pulmonary artery stenosis (PAS). 5/6 have posterior embyotoxon, 2/6 butterfly vertebrae, 4/6 hyperlipidaemia and 4/6 failure to thrive. Two patients have hypertriglyceridaemia (patients 3 and 5) and 2 patients have hypercholesterolaemia (patients 4 and 6). None of them have renal anomalies. One patient (patient 5) has a karyotype showing Jagged-1 gene disruption at the translocation breakpoint locus 20p12.3 (2n=46,XX, t(12,20) (q22,p12.3) (Figures 1 and 2). Five patients are still alive.

Discussion

Liver disease in AGS may present during the neonatal period and vary considerably, sometimes mimicking biliary atresia which needs to be excluded. Bile duct paucity may not be evident histologically in early disease[6]; only 60% of infants younger than 6 months have the abnormality. Two-thirds of our patients developed clinical evidence of chronic liver disease by 3 years of age. In our cohort of patients ascertained through liver involvement, two-thirds of the index patients have a positive family history. Only 46% of individuals in these families have clinical evidence of liver involvement.

Supportive treatment is important to minimise complications of liver disease. These include fat soluble vitamin supplements and continuous or nocturnal nasogastric/ gastrostomy tube feeding with medium-chain triglyceride and branched-chain amino acid milk formulae to meet increased calorie requirements for growth. Diuretics like spironolactone and albumin 20% infusions are used to control ascites and oedema. Control of pruritus with bile salt binding resins like cholestyramine and ursodeoxycholic acid have been tried. Rare reports of hepatocellular

carcinoma justifies long term evaluation of stable cirrhotic patients[7]. Cardiac anomalies may require corrective surgery. Overall morbidity and mortality depends on cardiac/renal disease, end-stage liver failure and intercurrent infection[7].

Orthotopic liver transplantation (OLT) is rarely required in 21-31% of patients when medical modalities fail [6-9]. Indications include progressive end-stage liver failure with synthetic dysfunction and intractable PHT (ascites, bleeding varices), bone fractures, intractable pruritus, disfiguring xanthomas and severe growth failure. It is estimated that 50% of patients diagnosed in infancy with early onset severe liver disease require OLT by 19y of age[7, 8]. Overall mortality for OLT is 46% with the highest in patients with unnecessary procedures like portoenterostomies for misdiagnosed biliary atresia[10]. It is therefore important to establish a precise diagnosis. Survival post-OLT is lower than average because of associated cardiopulmonary disease[11]. Full cardiac evaluation including ejection fraction measurement is advised pre-OLT. Renal anomalies can cause significant pre-OLT renal impairment. Careful renal function assessment is essential as renal failure post-OLT is known to occur[12].

The identification of the Jagged-1 gene mutation will increase the precision of the diagnosis of probands and increase identification of relatives who are minimally affected. Prenatal diagnosis can be offered if the mutation has previously identified in the proband.

Conclusion

AGS exists in Malaysian children. Only half of the patients have chronic liver disease or present with classical biliary hypoplasia. Family history is important. Morbidity and mortality depends on hepatic and extrahepatic disease. Genetic testing will aid accurate diagnosis of probands and their families, and it will play an important role in prenatal diagnosis.

Acknowledgements

We thank the Director General of Health for permission to publish this paper. We also thank Dr. Azizon Othman, Consultant Haematologist and Aminah Mekesat, Scientific Officer at the Unit of Haematology, Pathology Department of Kuala Lumpur Hospital for their contribution.

Table I: Revised diagnostic criteria for diagnosis of Alagille Syndrome (Picolli DA) [3]

	AGS Family History	Paucity Bile Ducts	Jagged 1 defect	Number of criteria needed
1.	None (proband)	+	-	3 or more features
2.	None (proband)	-	-	4 or more features
3.	None (proband)	-	+	1 or more features
4.	Present	+	-	1 or more features
5.	Present	unknown	-	1 or more features
6.	Present	-	identified	Any or no features

Major criteria include cardiac, renal, ocular disease, butterfly vertebrae or characteristic AGS facies.

Table II : Characteristics of Alagille Syndrome patients studied

Patient	Age at Diagnosis	Present Age	Sex/ Race	Family History	Facies	Liver	Cardiac	Eye *	Bone **	FTT	Lipid	Others	Alive/ Dead
1.	2 mo	13mo	M Malay	+ mother	+	PHT, synthetic dysfunction	ASD, PS	+	-	+	N	No bile duct paucity	Alive
2.	5y	5y 9mo	F Malay	+ brother, sister, father	+	PHT, synthetic dysfunction	-	+	-	+	N	-	Dead
3.	3.5y	5y	M Malay	+ 2 sisters, father	+	PHT, synthetic function N	-	+	-	+	raised TG	-	Alive
4.	4 mo	16mo	F Indian	+ 2 sisters, mother, grand-mother	+	hepatitis, hepatomegaly	PDA, VSD	+	-	-	raised choles	-	Alive
5.	birth	1y	F Malay	-	+	hepatitis, hepatomegaly	ASD, VSD, overriding aorta, PAS	-	+	-	raised TG	Jagged-1 gene disrupted by t(12,20) (q22,p12.3)	Alive
6.	2 mo	3y 7mo	F Malay	-	+	PHT, synthetic dysfunction	PAS	+	+	+	raised choles	xanthoma, rickets	Alive

Abbreviations : years (y), months(mo), male (M), female(F), failure to thrive (FTT), portal hypertension(PHT), atrial septal defect(ASD), pulmonary stenosis(PS), patent ductus arteriosus(PDA), ventricular septal defect(VSD), pulmonary artery stenosis(PAS), triglyceride(TG), cholesterol(Choles), normal (N). * Eye findings: posterior embryotoxon in all; **Bone findings were butterfly vertebrae in both. None had renal anomalies.

Table III: Frequency of major and minor features in the AGS patients studied

Major Features	Number/Total	%
Cholestasis	6/6	100
PHT/ synthetic dysfunction	3/6	50
	(2/6 alive)	(33)
PHT/ normal synthetic function	1/6	17
Hepatomegaly	2/6	33
AGS facies	6/6	100
Cardiac anomaly	4/6	66
	(ASD with mild PS; PDA/VSD; ASD/VSD/PAS/ overriding aorta; and PAS)	
Posterior embryotoxon	5/6	83
Butterfly vertebrae	2/6	33
Renal anomaly	0/6	0
Minor Features		
Hyperlipidaemia	4/6	66
	2 hypertriglyceridaemia, 2 hypercholesterolaemia	
Failure to thrive	4/6	66

Abbreviations : Atrial septal defect (ASD), pulmonary stenosis (PS), patent ductus arteriosus (PDA), ventricular septal defect (VSD), pulmonary artery stenosis (PAS).

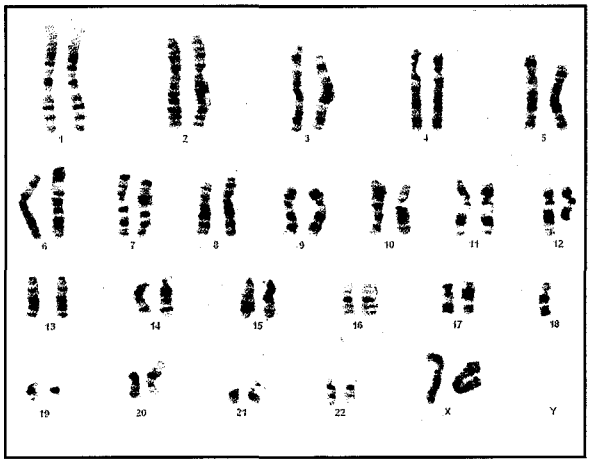

Fig. 1 : Karyotype of patient 5 showing Jagged-1 gene disruption at translocation breakpoint locus 20p12.3
Karyotype 2n = 46, XX, t(12,20)(q22,p12.3)
(Reproduced with permission from Dr. O. Azizon and Aminah Mekesat, Unit of Haematology, Pathology Department, Kuala Lumpur Hospital)

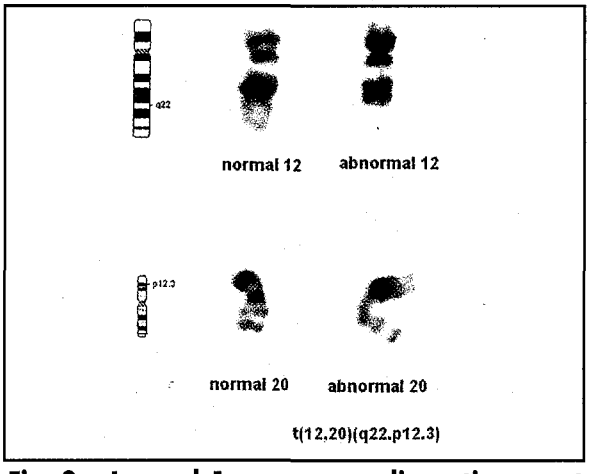

Fig. 2 : Jagged-1 gene disruption at translocation breakpoint locus 20p12.3
(Reproduced with permission from Dr. O. Azizon and Aminah Mekesat, Unit of Haematology, Pathology Department, Kuala Lumpur Hospital)

References

1. Li L, Krantz ID, Deng Y, et al. Alagille Syndrome is caused by mutations in human Jagged 1, which encodes a ligand for Notch 1. Nat Genetics 1997; 16: 243-51.
2. Eva A Roberts. The Jaundiced Baby. In : Deirdre A Kelly (ed). Diseases of the Liver and Biliary System in Children. London: Blackwell Science, 1999; 11-45.
3. Picolli DA. Alagille syndrome. In : Suchy FJ, Sokol RJ, Balistreri WF (eds). Liver Disease in Children. Philadelphia: Lippincot, Williams and Wilkins, 2001; 327-42.
4. Brigham and Women's Hospital, Boston. Protocols on Culture and Harvest of Peripheral Blood Lymphocytes. In : Barch MJ, Knutsen and Spurbeck JL. The AGT Cytogenetics Laboratory Manual. (3rd ed) Philadelphia: Lippincott-Raven, 1997; 91-3.
5. Brown MG, Lawce HJ. Peripheral Blood Cytogenetic Methods. In: Mitelman F (ed). An International System for Human Cytogenetic Nomenclature (ISCN). Karger, 1995; 88-172.
6. Emerick KM, Rand EB, Gold Muntz E, et al. Features of Alagille syndrome in 92 patients: requency and relation to prognosis. Hepatology 1999; 29: 822-9.
7. Quiros-Tejeira RE, Ament ME, Heyman AB, Martin MG, Rosenthal P, et al. Variable morbidity in Alagille syndrome : a review of 43 cases. J Pediatr Gastroenterol Nutr 1999; 29: 431-7.
8. Hoffenberg EJ, Narkewicz MR, Sondheimer JM, Smith DJ, Silverman A, Sokol RJ. Outcome of syndromic paucity of interlobular bile ducts (Alagille syndrome) with onset of cholestasis in infancy. J Pediatr 1995; 127: 220-4.
9. Tzakis AG, Reyes J, Tepetes K, et al. Liver transplantation for Alagille's syndrome. Arch Surg 1993; 128: 337-9.
10. Marino IR, ChapChap P, Esquievel CD, et al. Liver transplantation for arteriohepatic dysplasia (Alagille's syndrome) Transpl Int 1992; 5: 61-4.
11. Markowitz J, Daum F, Kahn E, et al. Arteriohepatic dysplasia : I. Pitfalls in diagnosis and management. Hepatology 1983; 3: 74-6.
12. Ellis D, Avner ED. Renal failure and dialysis therapy in children with hepatic failure in the postoperative period of orthotopic liver transplantation. Clin Nephrol 1986; 25: 295-6.