Thalassaemia genes in Baghdad, Iraq

Hassan I. Yahya,[1] Khalid J. Khalel,[2] Nasir A.S. Al-Allawi [3] and Ferial Helmi [3]

جينات الثلاسيمية في مدينة بغداد ـ العـــراق

حسان إبراهيم يحي وخالد جمعة خليل ونصير عبد السلام العلاوي وفريال عباس حلمي

خلاصـة : أجريت هذه الدراسة بهدف تقدير مدى انتشار جينات الثلاسيمية في مدينة بغداد ، واختير لها على نحو عشـوائي 502 من الحوامل اللاتي كن يترددن على إحدى العيادات الكبرى لرعاية الأمومة في المدينة . واسـتخدم متوسط حجم كريات مخفض يبلغ <80 فمتولتر كاختبار لتقصي الثلاسيمية لدى هؤلاء النسوة ، وتبع ذلك إجراء مجموعة من الاختبارات شملت تحليل الهيموغلوبين ودراسة مستوى الحديد للتأكد من صحة التشخيص . وتم على هذا النحو تشخيص 22 إصابة بخلّة الثلاسيمية البيتا و5 إصابات بخلّة الثلاسيمية الألفا وإصابة واحدة بخلّة الثلاسيمية الدلتا البيتا . واستناداً إلى هذه الأرقام تكون معدلات الانتشار المقدرة لخلّات الثلاسيمية البيتا والألفا والدلتا البيتا في بغداد ، 4,4% و1,0% و0,2% على التوالي . وتشمل الدراسة أيضا بحث هذه النتائج في سياق النتائج التي تم التوصل إليها في بلدان مجاورة .

ABSTRACT To estimate the prevalence of thalassaemia genes in Baghdad, a study was made of 502 randomly selected pregnant women attending a major maternity care clinic in the city. A reduced mean corpuscular volume (MCV) of < 80 fl was used as a screening test for thalassaemia in this population, followed by a battery of tests, including haemoglobin analysis and iron studies, to confirm the diagnosis. Using this approach, 22 cases were diagnosed of β-thalassaemia trait, five cases of α-thalassaemia trait and one case of δβ-thalassaemia trait. Based on these figures the estimated prevalence rates of β-, α- and δβ-thalassaemia traits in Baghdad, would be 4.4%, 1.0% and 0.2%, respectively. The study also includes a discussion of the above findings in the context of those reported in other neighbouring countries.

Les gènes de la thalassémie à Bagdad (Iraq)

RESUME Afin d'estimer la prévalence des gènes de la thalassémie à Bagdad, une étude a été réalisée chez 502 femmes enceintes qui ont été choisies au hasard parmi les femmes venant consulter dans une grande maternité de cette ville. Un volume globulaire moyen réduit (VGM) < 80 fl a été utilisé comme test de dépistage de la thalassémie dans cette population. Ce test a été suivi par un ensemble de tests comprenant l'analyse de l'hémoglobine et l'étude de la sidérémie afin de confirmer le diagnostic. En utilisant cette approche, 22 cas de trait β-thalassémique ont été diagnostiqués, ainsi que 5 cas de trait α-thalassémique et un cas de trait δβ-thalassémique. Sur la base de ces chiffres, les taux estimatifs de prévalence du trait de la β-, α- et δβ-thalassémie seraient de 4,4%, 1,0% et 0,2% respectivement. Cette étude comprend également une discussion relative à ces résultats en les comparant aux résultats rapportés dans d'autres pays voisins au Moyen-Orient.

[1] *Assistant Professor of Medicine;* [3] *Lecturer, Department of Pathology (Haematology); College of Medicine and Teaching Hospitals, University of Baghdad, Baghdad, Iraq.*
[2] *Senior Haematologist, Kurkuk Hospital, Kurkuk, Iraq*

Introduction

Thalassaemia syndromes are inherited defects in the rate of synthesis of one or more of the globin chains of haemoglobin. These disorders are widely distributed throughout the world, but occur in considerable frequencies in Mediterranean, Indian, south-east Asian and Middle Eastern populations [*1*]. Some studies have focused on the prevalence of thalassaemia genes in some Middle Eastern countries [*1–4*], but none were from Iraq, in spite of evidence suggesting that thalassaemias are not uncommon among Iraqis [*5*].

The aim in this study was to address this issue, by utilizing electronically measured mean corpuscular volume (MCV), coupled with a battery of confirmatory haemoglobin (Hb) and iron studies [*6*], to screen and confirm the diagnosis of thalassaemia in an obstetric population attending a major maternity care clinic in central Baghdad.

Baghdad, virtually at the heart of Iraq, has a population of about 4 million people (around 25% of the population of Iraq). It is the major business and cultural centre in Iraq and has always attracted people from all over the country. The bulk of these people have become permanently integrated over the decades, many through intermarriage, with the population of the city. Thus, unlike most other cities in Iraq, Baghdad's population is rather heterogeneous with most ethnic groups being represented to one extent or another.

Materials and methods

The study included 502 pregnant women (aged 15–45 years) in their second (306 women) and third trimesters of pregnancy (196 women). The subjects were randomly selected over a three-month period (1 May to 31 July 1990) from pregnant women attending a large maternity care clinic (Sheikh Omar Maternity Care Centre) in central Baghdad.

Full blood counts were performed on all subjects using an electronic Coulter S-plus counter. The Coulter counter was calibrated each day using a calibrant material provided by the manufacturers. Based on these blood counts, all subjects with microcytosis (as defined by an MCV < 80 fl) were investigated further. These further investigations included May–Grünwald and Giemsa-stained blood smears, reticulocyte counts, Hb H preparations, haemoglobin electrophoresis on cellulose acetate strips (pH = 8.6), Hb A_2 quantification (Marengo–Rowe method), Hb F quantification (Betke et al. method), serum ferritin by radio-immunoassay (ferritin kit—Amersham International UK) and serum iron and total iron binding capacity (atomic absorpion spectrometry) [*7*]. These investigations were performed by standard laboratory procedures as detailed by Dacie and Lewis [*8*], except where otherwise specified.

Results

Microcytosis was found in 98 of the 502 (19.5%) pregnant women investigated. β-thalassaemia heterozygosity (β-thalassaemia trait) was identified in 22 (22.4%) of these microcytic subjects, based on elevated Hb A_2 level (> 3.5%; normal range in our laboratory 1.7% to 3.5%). Hb A levels ranged in this category of patients between 3.8% and 6.1%, while Hb F levels ranged between 0.6% and 4.2%, with 35.4% having an increased F level (> 1%). Furthermore, when the predictive value of MCV was utilized to categorize patients into

Table 1 **Haematological findings in 98 subjects with microcytosis categorized into α, β thalassaemia traits and iron deficiency**

Parameter	Mean ± *s*		
	β thalassaemia	α thalassaemia	Iron deficiency
Haemoglobin (g/dl)	10.6 ± 1.3	10.8 ± 0.8	10.1 ± 1.0
Packed cell volume (%)	33.2 ± 4.3	33.9 ± 2.8	32.0 ± 2.9
Mean corpuscular volume (fl)	66.3 ± 3.3	69.5 ± 5.0	74.2 ± 4.0
Mean corpuscular haemoglobin (pg)	20.9 ± 1.1	22.1 ± 1.4	23.7 ± 1.3
Red cell count (x 10^{12}/l)	5.0 ± 0.7	4.9 ± 0.6	4.3 ± 0.3
Reticulocyte count (%)	1.9 ± 0.3	1.6 ± 0.2	1.2 ± 0.4
Haemoglobin A_2 level (%)	4.7 ± 0.6	2.3 ± 0.3	2.5 ± 0.4
Haemoglobin F level (%)	1.4 ± 1.0	0.8 ± 0.1	0.7 ± 0.1
Transferrin concentration (%)	26.4 ± 6.6	34.0 ± 12.0	7.8 ± 3.0
Serum ferritin (μg/l)	52.3 ± 38.1	57.6 ± 35.3	6.6 ± 2.1

those with β° or β^{+} mutations (MCV > 66.96 fl carries a β^{+} mutation as proposed by Rund and colleagues [9]), 45.5% of β-thalassaemia cases had the β° mutation, the rest β^{+}.

δβ-thalassaemia heterozygosity (trait) was identified in one subject based on the presence of microcytosis with increased Hb F (17%) and low Hb A (2%). Red-cell indices in this patient were a haemoglobin level of 10.49 g/dl, MCV of 71.9 fl, mean corpuscular haemoglobin (MCH) of 22.7 pg and a red cell count of 4.65×10^{12} per litre.

α-thalassaemia trait, on the other hand, was diagnosed in five subjects, based on the presence of microcytosis, with normal Hb A_2 and F and normal serum ferritin level (i.e. by exclusion of β- and δβ-thalassaemia, and iron deficiency as causes of microcytosis). No cases with haemoglobin H disease were identified in the studied cases. The remaining 70 cases with microcytosis had iron deficiency, based on reduced serum ferritin levels (< 10 μg/l). It is noteworthy that when a 15% transferrin concentration was used to identify iron-deficient cases, it managed to pick up all such cases as detected by serum ferritin, except one. This indicates the high level of reliability of the latter approach, and thus future screening studies in our country could utilize it, instead of the more expensive and less readily available serum ferritin assays.

Based on the above results the projected prevalence of β-thalassaemia trait in Baghdad is 4.4%, that of α-thalassaemia trait is 1.0% and that of δβ-thalassaemia trait is 0.2%. The full haematological profile of α- and β-thalassaemia, compared with cases of microcytosis due to iron deficiency are outlined in Table 1.

Discussion

Malaria was endemic throughout Iraq (including Baghdad) until the late 1950s (data from the records of the Centre for the Control of Communicable Diseases, Baghdad). It would not be unexpected to find thalas-

saemia genes prevalent in Baghdad, or in other parts of Iraq, in view of the theory of malaria selection [*1*] which has been offered as an explanation of the high prevalence rates observed in many parts of the world, including the Arabian Peninsula [*1–4,10*].

The prevalence rate of β-thalassaemia trait observed in this study is intermediate between those reported from Saudi Arabia and that from Turkey (Table 2). The findings of relatively equal numbers of β^+ and β° mutations in subjects in this study (based on the predictive value of the MCV) [*9*], although requiring confirmation by proper DNA studies, is quite interesting and appears to be different from preliminary reports from Saudi Arabia implying that the large majority of their mutations are β^+ [*4*].

α-thalassaemia is, however, quite common in the Arabian Peninsula [*2,4*], and although the prevalence rate in Baghdad, as found in this study, is much lower (Table 2), preliminary reports from the Basra area, in the southern part of Iraq, suggest a prevalence rate of about 20% (Dr D. Al-Shawi, personal communication, 1992). This latter figure is nearer to those from the Arabian Peninsula. α-thalassaemia in Iraq according to the experience of the authors is mostly caused by an α^+ defect, since haemoglobin Bart's hydrops fetalis has not been reported, while haemoglobin H disease is rare. Comprehensive studies from Saudi Arabia have also implicated α^+ defects to be almost solely responsible for their α-thalassaemia [*11*]. The latter point is important, and would imply that the low prevalence rate of α-thalassaemia obtained in this study may be an underestimation, since it is well known that a proportion of α^+ heterozygotes would be missed if the MCV is used as a screening test [*12*]. However, reliable conclusions on the molecular pathology of thalassaemia in Iraq will eventually require appropriate DNA studies.

Table 2 **Prevalence rates of α-and β-thalassaemia traits in some Middle Eastern countries**

Country	No. tested	Prevalence (%) β-thal.	α-thal.
Saudi Arabia	2643	10–20[a]	14–57[a]
United Arab Emirates	874	3.0	28
Oman	156	4.5	60
Turkey	900	1.66	0.1
Iraq (Baghdad)	502	4.4	1.0

[a]These ranges are due to variable prevalence rates in different areas of Saudi Arabia

While α-thalassaemia has never been a health problem in Iraq, β-thalassaemia in its homozygous state is [*5*]. It is the authors' opinion that the high prevalence rate of heterozygous β-thalassaemia, and that of consanguineous marriages in the community, in addition to high morbidity and mortality rates, and the heavy burden imposed on the health services in association with homozygous thalassaemia, should prompt the health authorities to establish an effective prenatal diagnostic programme. Such a programme could only be possible by initiating research programmes to determine the molecular pathology of this type of thalassaemia in Iraq. Moreover, further studies to estimate the prevalence of thalassaemia in other regions of the country, utilizing a similar approach as in the current study, is of prime importance in order to establish a comprehensive national programme to combat this important health problem.

References

1. Weatherall DJ. Clegg JB. *The thalassaemia syndromes*, 3rd ed. Oxford, Blackwell Scientific Publications, 1981.
2. White JM et al. Thalassaemia genes in peninsular Arabs. *British journal of haematology*, 1985, 60:269–78.
3. Cavdar AP, Arcasoy A. The Incidence of β thalassaemia and abnormal haemoglobin in Turkey. *Acta haematologica*, 1971, 45:312–8.
4. El-Hazmi MAF. Haemoglobin disorders: a pattern for thalassaemia and haemoglobinopathies in Arabia. *Acta haematologica*, 1982, 68(1):43–51.
5. Taj-Eldin S et al. Thalassaemia in Iraq. *Annals of tropical medicine and parasitology*, 1968, 62:147–53.
6. Pearson HA et al. Comprehensive testing for thalassaemia trait. *Annals of the New York Academy of Science,* 1974, 232:135–44.
7. Ramsay WNM. The determination of the total iron binding capacity of the serum. *Clinica chimica acta,* 1957, 2:221–6.
8. Dacie JV, Lewis SM. *Practical haematology*, 6th ed. Edinburgh, Churchill Livingstone, 1984.
9. Rund D et al. Mean corpuscular volume of heterozygotes for β-thalassaemia correlates with the severity of mutations. *Blood,* 1992, 79:238–43.
10. Niazi GA, Rowland HAK. Haemoglobinopathies. a review. *Saudi medical journal*, 1989, 10:340–51.
11. Weatherall DJ. Some aspects of the haemoglobinpathies of particular relevance to Saudi Arabia and other parts of the Middle East. *Saudi medical journal,* 1988, 9:107–15.
12. Higgs DR, Weatherall DJ. Alpha thalassaemia. *Current topics in hematology*, 1983, 4:37–97.