NEONATAL SCREENING FOR CONGENITAL HYPOTHYROIDISM IN DUHOK GOVERNORATE (IRAQ); A PRELIMINARY STUDY

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ABSTRACT

Background: Congenital hypothyroidism is one of the commonest treatable endocrine diseases and can affect he growth and mental development of neonates. The neonatal screening program for congenital hypothyroidism has become a routine procedure in all developed countries and considered a monitoring tool for the early detection and therapy of hypothyroidism. In this study we investigated the prevalence and associated factors of congenital hypothyroidism in Duhok governorate of Kurdistan Region (Iraq).

Methods: A cross-sectional study was conducted during the period from July 2014 to February2015. Capillary blood samples were obtained from 3000 newborn babies by heel prick and tested initially for the thyroid stimulating hormone (TSH) followed by assessing free thyroxine in those with TSH level >10 µIU/ml. Demographic and clinical data such as residency, ethnicity, mother age, newborn age, sex, maturity, birth weight, presence of congenital anomalies as well as related marriage and thyroid diseases of the mother were collected.

Results: The number of neonates having TSH level >10µIU/ml was 88 resulting in a recall rate of 2.9%. Of the88 recalled neonates, 4 were diagnosed with congenital hypothyroidism (4.5%; 1 in 750 live births) and 7 with subclinical hypothyroidism (7.9%, 1 in 428 live births). There was significant association between the occurrence of congenital hypothyroidism and prematurity (p=0.04), congenital anomalies (p=0.03) and thyroid disease of the mother (p=0.03).

Conclusions: The study sample showed a high prevalence of congenital hypothyroidism among neonates of Duhok governorate. Appropriate neonatal screening program and monitoring are essential to reduce the irreversible health consequences of this disorder.

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Keywords: congenital hypothyroidism, screening program, TSH, Kurdistan, Iraq.

Congenital hypothyroidism is one of the commonest treatable endocrine diseases and can affect the growth and mental development of neonates[1]. The clinical diagnosis of congenital hypothyroidism is difficult, and since most neonatal cases do not show specific symptoms and signs, the disease is detected in less than 5% of neonatal patients[2]. Congenital hypothyroidism prevalence has been found to differ among races and regions, with a higher prevalence among Asian neonates than among neonates of other regions. The incidence of congenital hypothyroidism in live births varies from 1:3000 to 1:4000 in different parts of the world, and the incidence and prevalence of the disease are influenced by multiple environmental, genetic, and autoimmune factors[3].

Early diagnosis and treatment of congenital hypothyroidism by using a screening method are essential; as without prompt treatment, irreversible mental

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retardation, growth failure, and a variety of neuropsychological deficits are inevitable.[4] Newborn screening for congenital hypothyroidism has become routine in all developed countries and most of Eastern Europe, and is under development in many developing countries[3,4]. In Iraq, nationwide screening for congenital hypothyroidism has still not been adopted and to our knowledge there are no published local data about the incidence of congenital hypothyroidism in Duhok governorate. This was the primary impetus for conducting this study to establish our local data which will help the health authorities in establishing local and then nationwide neonatal screening programs for congenital hypothyroidism in order to reduce the undesirable health consequences of this disorder.

METHODS

Setting and Selection of neonates: A total of 3000 newborn babies were screened over8 months using blood spot with diagnosis by TSH analysis. The screening included newborns babies brought by their mothers for vaccination units at maternity hospitals in five districts in Duhok governorate, namely Duhok city, Semil, Akre, Zakho and Amedy. Duhok governorate has an estimated catchment population of more than one million. Currently these vaccination units serve two important functions; administering the scheduled vaccines and providing the mother with the vaccination card which is a pre-requisite to have the birth certificate in Duhok governorate. Vaccination units provide vaccination for every newborn with daily volume load ranging from 10 babies in Amedy to 120 babies in Duhok city. A simple randomization procedure was used to include the neonates in the study, every third baby, aged more than 24 hour and less than 28 days. Exclusion criteria included mother's refusal, neonates who were apparently ill and neonates who had blood transfusion. The study protocol was approved by the Kurdistan Board for Medical Specialties and the General Directorate of Health in Duhok Governorate.

Data Collection and Information: Data were collected from mothers interviewed using a special questionnaire form. Informed mother consent was obtained before sample collection. Demographic and clinical data such as residency, ethnicity, mother age, newborn age, sex, maturity, birth weight, congenital anomalies, consanguinity, and thyroid disease of the mother were collected.

Specimen collection and laboratory testing: The specimen collection from the neonates was carried out using capillary blood obtained from the heel by lancing the heels of the neonates on the lateral edge of the planter surface avoiding the posterior aspect of the heel. Enough whole blood was spotted on a special filter paper card (What man protein saver 903 card) to fill and saturate the marked circle. Spotting blood over a previous blood spot or double spotting was avoided. The filter papers were left to dry at room temperature overnight away from heat, moisture, contaminating surfaces or substances. The dry filter papers were stored and transported in paper envelopes (not plastic envelops) to the public health laboratory in the preventive health directorate in Duhok city.

The blood TSH in the spotted on filter paper was measured by ELISA (Enzyme

Linked Immunosorbent Assay) technique using neonatal TSH kit (DIAGNOTIC AUTOMATION, INC. CA 91302, USA). This kit has a detection range of 0 - 250 µIU/ml, specificity of 97% and sensitivity of 1.0 µIU/ml. Neonates with TSH levels of 10µIU/mlor less were excluded from further evaluation. Parents of neonates with TSH level of more than 10µIU/ml in filter - paper blood sample were recalled by phone within 4 weeks for reassessment. A blood sample (5 ml) was collected by venipuncture into a gel tube. The blood sample was allowed to clot at room temperature for 1 hour, and was then centrifuged for 15 minutes at 3000 round per minute for measurement of TSH and free T4 levels. The analysis was done by Elecsys and Cobas-immunoassay analyzers (Roche-Germany) by using the "ECLIA" electrochemiluminescence immunoassay method. Abnormal results were confirmed twice. Primary congenital hypothyroidism was diagnosed if TSH was > 10 µIU/mL and the free T4 that was below the normal range for age as following:

- Between one and four days of life, the normal range for serum free T4 concentration is about 2 to 5 ng/dL (25 to 64 pmol/L).
- Between one and four weeks of life, the normal range for serum free T4concentration is 0.8 to 2.0 ng/dL (10 to 26 pmol/L). Neonates were diagnosed with subclinical hypothyroidism when serum TSH level >10 µIU/ml and the free T4 level normal[5].

Data analysis: Statistical analysis was performed using SPSS statistical software for Windows (SPSS 19.0, Chicago, IL, USA). For continuous variables, the mean and 95% confidence intervals or standard deviation (SD) were calculated. For interval and qualitative variables, frequencies and percentages were calculated and expressed in frequency tables. Statistical analysis has been conducted to calculate the P-value using chi square test (or Fisher's exact test if an expected number in any cell was less than 5); *P value* $\leq$ 0.05 was considered as statistically significant.

RESULTS

Over an eight-month period, 3000 neonates were screened. Of these, 1554 (51.8%) were males and 1446 (48.2%) were females with a male to female ratio of 1.1:1. The mean age of the neonates was 8.5 days (95% Confidence Interval (CI) was 8.1 - 8.8) with a range of 2 - 28 days. The mean age of the mothers was 27.9 years (95% CI was 27.6 - 28.2) and the range was 14 - 48 years. The most common age group was 20 to 30 years (N=1864, 62.1%). The major ethnical group was Kurds (N=2832, 94.4%) followed by Arabs (N=144, 4.8%) and Turkmen (N=24, 0.8%). About two thirds of the study population (N=1954, 65.1%) was residents of Duhok city (Table 1).

Table 1: Residency of the Study Population (N = 3000)

Place	Number	Percentage
Duhok city	1954	65.1
Semil	470	15.7
Akre	266	8.9
Zakho	196	6.5
Amedy	114	3.8
TOTAL	3000	100

The majority of the neonates had normal body weight at birth (N=2784, 92.8%) with 6.1% (N=184) being underweight and 1.1% (N=34) being overweight. Only 2.8% (N=84) of the neonates were

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premature. Congenital anomalies were detected in 118 neonates (3.9%). Marriages were consanguineous in 36% (N=1080). Thyroid disease of the mother was present in 1.2% (N=36), (Table 2).

Table 2: Clinical Data of the Study Population (N = 3000)

Clinical features		Number	Percentage
Baby weight at birth	Less than 1500 grams	184	6.1
	1500-4000 grams	2782	92.8
	More than 4000 grams	34	1.1
Maturity	Mature	2916	97.2
	Premature	84	2.8
Congenital anomalies	Yes	118	3.9
	No	2882	96.1
Related marriage	Yes	1080	36
	No	1920	64
Thyroid disease of the mother	Yes	36	1.2
	No	2964	98.8

The mean value of TSH level was 2.65 µIU/ml (95% CI = 2.4 - 2.9) and the standard deviation was 5.4 µIU/ml. The range was 0-85 µIU/ml. The number of neonates having initial blood TSH level > 10 µIU/ml was 88 resulting in a recall rate of 2.9%. The mean TSH level for the recalled neonates was 7.6 µIU/ml (95% CI = 4.4 - 10.8) with a standard deviation of 5.8 µIU/ml. The mean free T4 level for the recalled neonates was 31 pmol/l (95% CI=25-37) with a standard deviation of 10.4 pmol/l. Out of the 88 recalled neonates, 4 were diagnosed with congenital hypothyroidism (4.5%; 1 in 750 live births) and 7 with subclinical hypothyroidism (7.9%, 1 in 428 live births). The mean TSH for neonates with congenital hypothyroidism was 32.4 µIU/ml (95% CI = 8-72) while the mean free T4 was 5.1 pmol/l (95% CI = 2.3 - 7.9). For neonates with subclinical hypothyroidism, the mean TSH was 15 µIU/ml (95% CI = 13.1 - 17) with the mean freeT4 being 36 pmol/l (95% Cl = 29 – 43), (Table 3).

Table 3: Laboratory Data of the study Population (N = 3000)

Parameter	Value
Mean TSH of study population (95% CI)	2.65 µIU/ml (2.4 - 2.9)
Recall rate	2.9% (88/3000)
Mean TSH for recalled neonates (95% CI)	7.6 µIU/ml (4.4 - 10.8)
Mean free T4 for recalled neonates (95% CI)	31 pmol/l (25-37)
Incidence of congenital hypothyroidism (CH)	1 in 750 live births
Incidence of subclinical hypothyroidism (SCH)	1 in 428 live births
Mean TSH for neonates with CH (95% CI)	32.4 µIU/ml (8-72)
Mean free T4 for neonates with CH (95% CI)	5.1 pmol/l (2.3 - 7.9)
Mean TSH for neonates with SCH (95% CI)	15 µIU/ml (13.1 - 17)

Parameter	Value
Mean free T4 for neonates with SCH (95% CI)	36 pmol/l (29 - 43)

Abbreviations: CH= Congenital hypothyroidism, SCH = Subclinical hypothyroidism

There were significant association between congenital hypothyroidism and each of prematurity (p=0.04), congenital anomalies (p=0.03) and thyroid disease of the mother (p=0.03). There was no significant statistical association between congenital hypothyroidism and either of sex of the neonates, body weight at birth, age of the mother, residency, ethnicity and related marriage (Table 4).

Table 4: Factors Associated with Congenital Hypothyroidism in the Recalled Neonates

Parameters		Congenital hypothyroidism (N = 4)	No congenital hypothyroidism (N = 84)	P-value
Sex of neonates	Male sex	1	40	0.36
	Female sex	3	44	
Age of themothers	11-20 years	0	3	0.86
	21-30 years	2	53	0.48
	31-40 years	2	26	0.38
	41-50 years	0	2	0.91
Residency	Duhok city	2	47	0.60
	Semil	0	6	0.75
	Akre	1	14	0.53
	Zakho	0	12	0.55
Ethnicity	Amedy	1	5	0.24
	Kurds	4	81	0.86
	Arabs	0	3	
Baby weight at birth	Less than 1500 g.	1	7	0.32
	1500-4000 g.	3	74	0.41
	More than 4000 g.	0	3	0.86
Maturity	Premature	3	19	0.04
	Mature	1	65	
Congenitalanomalies	Yes	2	5	0.03
	No	2	79	
Relatedmarriage	Yes	2	36	0.58
	No	2	48	
Thyroid diseaseof the mother	Yes	2	6	0.03
	No	2	78	

DISCUSSION

Hypothyroidism is one of the most common and treatable endocrine disease in which there is insufficient amount of thyroid hormones[6]. The thyroid gland dysfunction causes mental and physical slowing, poor resistance to cold and in children, mental retardation and dwarfism with its personal and social sequelae[7].

Because the treatment is simple, effective, and inexpensive, nearly all of the

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developed world practices newborn screening to detect and treat congenital hypothyroidism in the first weeks of life[8]. The primary aim for this study was to establish local data in order to determine the size of problem so that we can help the local health authorities in establishing neonatal screening programs, we used the initial blood TSH assay with a cut-off point of 10µIU/mL. This cut-off point is the initial one that was used by the Quebec study[9]. The most striking findings of our study were a high prevalence of congenital hypothyroidism (1 in 750 live births), subclinical hypothyroidism (1 in 428 live births) and high patient recall rate (2.9%) after primary screening. The prevalence of congenital hypothyroidism among the neonates in this study was about 3-4 times higher than the figures reported earlier among newborns in other areas in the middle-East and in the developed countries. Studies conducted in the West Black Sea area in Turkey reported incidence of congenital hypothyroidism as 1 in 2326 and a recall rate 1.6% .4A study from Lebanon reported incidence of 1 in 1823 live births[10]. It is also higher than that reported in United States (1 in 2372)[11] and most communities of Europe (1 in 3500 to 1 in 4000)[12] although the incidence rates in Europe are variable ranging from 1: 800 in the Greek Cypriot population[13] to 1:10000 in France.[14] Moreover, the worldwide incidence of congenital hypothyroidism is reported to be 1 in 3000-4000 live births[15]. On the other hand, our results are in-line with those from studies conducted in many areas of Iran ranging from 1:500 to 1:1000[16,17]. The results suggest a high congenital hypothyroidism prevalence rate, despite that the obtained figures may not necessarily reflect the real prevalence of congenital hypothyroidism of the general population in the Duhok governorate. Targeting a small number size for such a screening study could be a factor behind the higher rate of congenital hypothyroidism obtained in the current study compared to other published data worldwide. In addition, the majority of newborns were from Kurd ethnicity and their families were habitants of Duhok governorate in Iraq, an endemic area with iodine deficiency[18]. Although we did not measure the neonatal and maternal urinary iodine levels, a study done in Duhok by Rasheed in 2013 showed that iodine deficiency is still a major public health problem in Duhok Governorate with more than half (53.4%) having median urinary iodine level less than the optimal level of "100µg/l"[19].

The prevalence of subclinical hypothyroidism in our study was 1 in 428 live births. These are comparable to those reported from Isfahan (1 in 349) and Zanjan (1 in 398) provinces of Iran3 but are higher than those reported in the Hispanic population of the United States (1 in 2000 live births) and in Europe (1 in 4000 live births) [11].

In this study, with a cut-off point of capillary heel-prick blood TSH>10 µIU/ml, the overall recall rate was in the higher range of recall rates reported by European countries. With cut-off points of capillary blood TSH values 25 µIU/ml or 30 µIU/mL, most of the European countries reported their recall rates between 0.16 and 2.7 %[17,20]. Whereas in the United States, with cut-off points of capillary blood TSH values >15 µIU/ml,

the recall rate after primary TSH screening is approximately 0.05%[1]. These varying recall rates are attributable to several factors, such as the use of T4 or TSH level or both for screening, differences in sample collection methods and analysis procedures in different laboratories, and differences in recall criteria, which are related to the cultural regional and social factors of a country[3]. It may also reflect the levels of iodine deficiency in different regions[21]. the factors associated with congenital hypothyroidism in this study were prematurity, thyroid disease of the mother and congenital anomalies. The association of congenital hypothyroidism with prematurity and family history of thyroid disease has been shown in some studies[4,11,16] but not in others[3]. We found no significant relationship between congenital hypothyroidism occurrence and sex of the neonates, body weight at birth, age of the mother, residency, ethnicity, and related marriage. Despite the lack of significant statistical association between congenital hypothyroidism and sex of neonate, which has also been shown in another study[3], congenital hypothyroidism incidence was higher in female neonates with a female to male ratio of 3:1. Recent studies suggest that nearly all-screening programs report a female preponderance, approaching a female: male ratio of 2:1. In our study, there was no association between mother's age and congenital hypothyroidism which could be attributed to the small number of mothers less than 20 years and more than 40 years who were included in the study. There were noticeable differences in the prevalence rates among different districts of Duhok governorate (1: 114) in Amedy, 1: 266 in Akre, 1:977 in Duhok city). This difference could reflect a status of iodine deficiency in Amedy and Akre which are mountainous areas and are more likely to be iodine deficient[22].In our study, there was no significant statistical difference between Arabs and Kurds which could be due to the small number of Arabs included in the study population. The body weight of the neonate at birth was not associated with congenital hypothyroidism which also is similar to that reported in earlier study[3].

Limitations of the study: The major limitation of this study is in the sample size which is small for a neonatal screening program. Using initial blood TSH assay carries important limitations like missing delayed TSH elevation in infants with thyroid-binding globulin (TBG) deficiency, central hypothyroidism, and hypothyroxinemia[23].The lack of a follow up period was another important limitation to differentiate cases of transient hypothyroidism from permanent hypo-thyroidism.

CONCLUSIONS

The study sample showed a high prevalence of congenital hypothyroidism among neonates of Duhok governorate compared to other areas in the middle-East and in the developed countries. Factors associated with congenital hypothyroidism were prematurity, congenital anomalies and thyroid disease of the mother. Appropriate neonatal screening program and monitoring are essential to reduce the irreversible health consequence of this disorder. Further larger scale randomized studies are advised to study the factors

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associated with congenital hypo-thyroidism.

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ثوختة

ظةراكرنا ساظايان بو نةخوشيا ثةريزادةيا سروشى ل ثاريَزطةها دهوكىَ

ئارماج: كيَم ثةريزادةيا سروشتى ئيَكة ذ مشةترين نةخوشييَن هرمونى كو ب ساناهى دئيَتة ضارةسةركرن بةلىَ نيشانيَن وىَ ل زوربةيا زاروكيَن بةرةبةر ديار دكةت و مةزن بونا وان لاواز دبيت و نةظاذييارى شيَتبن. ثروطرامىَ ظةراكرنا ساظايان بو نةخوشيا كيَم ثةريزادةيا سروشتى يىَ بوية ريَبازةكا ئاسايي ل هةمى وةلاتين هةظجةرخ و يىَ ريَزدار كرية وةك كةرةستةكىَ زيَرظاكرنىَ يا زوى بو سةحكرن و ضارةسةركرنا نةخوشيا كيَم ثةريزادةيىَ. ئارمانجا ظىَ ظةكولينىَ ذبو ظةراستكرنا هةلكةتنا ئيَشا كيَم ثةريزادةيا سروشى.

ريَكيَت ظةكولينىَ:

ئةف خويندنة هاتة رةفتاركرن لسةر 3000 زاروكيَن ساظا ل بنطةهىَ كوتاندنىَ ل نةخوشخانةييَن سةرةكى ييَن دابيكبونىَ ل ثاريَزطةها دهوكىَ دماوىَ 8 هةيظادا هةر ذ تيرمةها سالا 2014 تاكو شواتا سالا 2015. زاروكيَن ساظا ييَن ذيىَ وان ثتر ذ 24 دةمذميَرا و كيَمتر ذ 28 روذا هاتنة وةرطرتن. ريَكا سادة يا ريَزكرنىَ هاتة بكارئينان بو ذيَطرتنا زاروكيَن ساظا دناظ ظةكولينىَ دا (زاروكىَ سيىَ ذىَ طرت). خوينا دةمارىَ موى هاتةكيَشان ب كونكرنا ثشتثةحنيىَ و هاتةثيظان بةراهيكىَ ب ثشكنينا TSH و ديظدا ريَذا فرى تايروكسين دطةل وان ئةويَن TSH ثتر ذ 10 مايكرويونت – مل.

ئةنجام:

ذمارا زاروكيَن ساظا ئةويَن TSH وان ثتر ذ 10 مايكرويونت – مل 88 بون و دئةنجامدا ريَذا طازيكرنا وان بو جارا دووىَ بوية 2.9% ذظان 88 طازيكريا 4 هاتنة دةستنيشانكرن وةك نةخوشيَن كيَم ثةريزادةيا سروشى (4.5%، 1 ذ 750 دايكبوييَن ساخ) و 7 وةكذيَر كيَم ثةريزادةيا (7.9%, 1 ذ 428 داييك بوييَن ساخ). ثةيوةنديا هةذمارى ديار بو ودناظبةرا هةلكةتنا نةخوشيا كيَم ثةريزادةيا سروشى و كاظشكيتى، نةريَكييَن سروشتى و ديروكا خيَزانىَ بو نةخوشييَن ثةريزادىَ هةبن.

دةرئةنجام:

ثشكنينا ديار كر كو زارووييَن ساظا ل ثاريَزطةها دهوكىَ هةلكةتنا زيَدة يا هةى بو ئيَشا كيَم ثةريزادةيا سروشى 4.5%. تةرخانكرنا ثروطرامىَ ظةراكرنا ساظايان و زيَرةظانيكرنا وان يا ثيَدظية و طرنطة بو كيَمكرنا ئةنجاميَن نةساخلةمييَن ظىَ نةخوشيىَ.

الخلاصة

تحري قصور الغدة الدرقية الخلقي لدى حديثي الولادة في محافظة دهوك

الهدف: قصور الغدة الدرقية هو واحد من أشهر أمراض الغدد الصماء شيوعاً قابل للعلاج، ويمكن أن توثر على النمو والتطور العقلي للاطفال حديثي الولادة. أصبح برنامج فحص حديثي الولادة لقصور الغدة الدرقية الخلقي إجراء روتيني في جميع الدول المتقدمة وتعتبر أداة رصد للكشف المبكر والعلاج من قصور الغدة الدرقية. في العراق لم يتم بعد اعتماد هذا البرنامج الوطني. الهدف من هذه الدراسة التحقيق في حدوث حالات قصور الغدة الدرقية الخلقي ومعدل استدعاء المريض في محافظة دهوك.

طرق البحث:

أجريت هذه الدراسة المقطعية على 3000 اطفال حديثي الولادة في وحدات التطعيم داخل مستشفيات الولادة الرئيسية في محافظة دهوك خلال فترة ثمانية أشهر من يوليو 2014 الى فبراير 2015. تم إدراج حديثي الولادة الذين تتراوح أعمارهم بين أكثر من 24 ساعة وأقل من 28 يوماً في البحث. استخدم الانتقاء العشوائي البسيط ليشمل حديثي الولادة في الدراسة (كل ثالث طفل). تم اخذ الدم من الاوعية الدموية الشعرية من خلال وخز الكعب ومن ثم إجراء اختبار هرمون (TSH) أولاً ويضاف اليه هرمون الثيروكسين الحر للذين مستوى الـTSH أكثر من 10 ميكروبونت – مل.

النتائج:

بلغ عدد المواليد الذين لديهم مستوى TSH أكثر من 10 ميكروبونت – مل 88 وهذا أدى الى الاستدعاء بمعدل 0.2% من المستدعين الحديثي الولادة الثمانية والثمانون، تم تشخيص 4 بقصور الغدة الدرقية الخلقي (4.5%، 1 من 750 ولادة حية) و7 مع قصور الغدة الدرقية تحت الكلينيكي (7.9%، 1 من 428 ولادة حية). كان هناك ارتباط ذات دلالة إحصائية بين حالات قصور الغدة الدرقية الخلقي والخدج، التشوهات الخلقية والتاريخ العائلي لامراض الغدة الدرقية.

الاستنتاجات:

أظهرت فحوصات حديثي الولادة في محافظة دهوك وجود نسبة عالية من حالات قصور الغدة الدرقية الخلقي (4.5%). وجود برنامج مناسب لفحص الاطفال حديثي الولادة ورصدهم ضروري للحد من عواقب صحية لا رجعة فيها لهذا الاضطراب.

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