



Neural Tube Defects: Distribution and Associated Anomalies

Diagnosed by Prenatal Ultrasonography in Iranian Fetuses

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Abstract

Objectives: The current study aimed at providing detailed information about the distribution, associated anomalies, and syndromes in Iranian fetuses with neural tube defects (NTDs).

Methods: The current study was conducted in Yas Females' referral and teaching hospital in 18 months from 2014 to 2016. All fetuses with a prenatally detected neural tube defect were included in the study. Neural tube defect characterization, gestational age, maternal reproductive factors, maternal risk factors, and associated anomalies were recorded.

Results: Neural tube abnormalities were identified prenatally in 80 fetuses: 40 cases of ex/anencephaly, 22 cases of spina bifida, 13 cases of cephalocele and 5 cases of anencephaly/craniorachischisis. All the cases were detected before 21st week of gestation and 92.5% of the ex/anencephaly cases were diagnosed in the 1st trimester. Moreover, 40% of the open spina bifida cases in the current study were myelocele, while 75% of them referred only due to abnormal cranial findings. Incomplete consumption of folate was the most common associated risk factor (45%). Associated anomalies were recorded in 53 (66%) fetuses, with more prevalence in the fetuses with spina bifida (90%). Among the associated anomalies, central nervous system (CNS) anomalies were the most common type (26.26%). Chiari II was found in all the cases of open spina bifida and the ventriculomegaly rate was 30% in this group. Extremities anomalies and spine deformities were the 2nd and 3rd common associated findings, respectively. Limb-body-wall complex/amniotic band syndrome was the most common identified associated syndrome (6%).

Conclusions: Results of the current study confirmed the high prevalence of associated anomalies in neural tube defect cases and revealed the capability of detailed sonography to detect and define such abnormalities.

1. Background

Neural tube defect (NTD) is one of the most common congenital malformations caused by disturbance in neurulation at the week 3 - 4 of gestation, which could be associated with environmental, genetic, or both factors (1, 2). Despite the diagnostic role of ultrasound examination combined with the 2nd trimester maternal serum alpha-fetoprotein screening, NTDs cause considerable mortality and morbidity (1).

Based on Wilson et al. description of closure failure of neural tube in its rostral or caudal aspect, NTDs are classified into 4 types: 1. Ex/anencephaly, 2. Spina bifida, 3. Cephalocele, and 4. Anencephaly/craniorachischisis (3).

Extensive researches are performed on NTDs; however,

few studies exist on associated anomalies in prenatal ultrasound evaluation. The rate of associated malformation is reported 17% to 80% in different studies, most of which are performed on autopsy cases. The most common associated malformations are other central nervous system (CNS) or spinal defects, spinal deformities, club foot, and cleft lip/palate (1, 4-7). NTDs could be also a component of syndromic process such as amniotic band syndrome, the Meckel-Gruber syndrome, etc (4, 8-10).

The current study aimed at describing NTDs distribution as well as frequency and types of associated anomalies in prenatal ultrasound examinations in Iranian population and comparing the obtained results with those of previous studies. To the authors' best knowledge, it was the 1st detailed study designed in Iran to evaluate diagnosis

process, pattern of involvement, and concomitant anomalies and syndromes in fetuses with neural tube abnormalities.

2. Methods

The current descriptive cross-sectional study was conducted in 18 months from 2014 to 2016. It was conducted at Yas females hospital, a females referral and teaching hospital at Tehran University of Medical Sciences, which receives most NTD cases from Tehran and nearby cities. All referred fetuses with a prenatally detected NTD were included in the study. Only 1 of the cases was referred to the center for the 1st scan. Based on the study design, incidence rate could not be evaluated in the study. The institutional Ethics Board approved the protocol of the current prospective study.

All ultrasound examinations were performed transabdominally by a radiologist using a 2 - 6 MHz convex array transducer (Affiniti50G, Philips healthcare, 3000 minute-man road andover, MA 01810, USA)

NTD characterization (type, location, content), gestational age, maternal reproductive factors (mother's age, and number of previous deliveries and miscarriages), associated anomalies (hydrops, face/neck, genitourinary, gastrointestinal, spine, extremities, cord, heart, thoracic/abdominal wall), and karyotype were recorded. Furthermore, previous ultrasound examinations (often in private centers) were recorded and compared with the final diagnosis. Known maternal risk factors based on literature (obesity, diabetes mellitus, folate use, consanguineous marriage, drug, seizure, family history of NTD) were also investigated in mothers and recorded. Folic acid consumption in the study was 0.4 mg/day 1 month before the subjects attempted to conceive. Fetal karyotyping and postmortem autopsy were recommended to all of the couples. The data were analyzed with SPSS software, version 24 (IBM SPSS, Armonk, NY, USA).

3. Results

During the study period, NTD was detected prenatally in 80 fetuses, of which only 1 was referred to the center for the 1st scan. In those which had previous examinations, 1 case had normal ultrasound screening, but positive quad test. Moreover, 4 cases were referred for other abnormalities and the associated NTD was detected at the center (1 with achondrogenesis, 1 with the Meckel-Gruber syndrome, 1 with contracture in extremities, and 1 with amniotic band syndrome). The other cases were referred to the center with correct detection of NTD or report of cranial

signs suggesting NTD. All the cases were diagnosed before the 21st week of gestation.

Reproductive factors are summarized in [Table 1](#). NTDs were more common in mothers aged 20 - 29 years (49 cases: 61.3%). In 32 (40%) cases, mothers were primigravid. History of abortion was positive in 18 cases, among whom 6 cases had abortion due to NTD.

3.1. Risk Factors

Risk factors are summarized in [Table 2](#). About 41.25% of NTDs could not be attributed to a known risk factor. Insufficient consumption of folate was recorded in a total of 36 (45%) cases with dominance in the ex/anencephaly group (23 cases: 57.5%). The 2nd common associated risk factor was consanguineous marriage in 25 (31.2%). Only in few cases, obesity and history of diabetes mellitus were detected.

3.2. NTDs Distribution

Out of these cases, ex/anencephaly was the most common NTD (40 cases: 50%) and 37 cases were in the 11th to 14th week of gestation ([Table 3](#)).

Twenty-two cases (77.27%) were diagnosed with spina bifida with more prevalence in female fetuses. Only 2 cases were closed type; 1 with sacral meningocele and the other with anterior sacral meningocele ([Figure 1](#)). Among the open spina bifida cases, lumbosacral was the most common location. Eight (40%) cases were detected as myelocele and 12 (60%) cases as myelomeningocele. Among the myelocele cases, only 2 (25%) fetuses had true diagnosis in private centers and the others (75%) were referred due to cranial signs (lemon and banana signs). One referred fetus with open spina bifida (thoracolumbar myelomeningocele) had normal AFP in the quad test and the diagnosis was confirmed in the center.

Moreover, 13 cases were diagnosed with cephalocele. Among these, 2 cases had only a cystic structure protruded through a bony defect (meningocele), whereas in the others, brain tissue herniated through a bony defect (encephalocele). Occipital region was the most common location ([Table 4](#)).

Five cases were diagnosed with anencephaly/craniorachischisis. Furthermore, 3 cases had craniorachischisis and 2 cases had anencephaly.

3.3. Associated Anomalies

Associated anomalies were recorded in 53 (66%) fetuses, with higher prevalence in fetuses with spina bifida ([Table 5](#)). CNS anomalies were evaluated in the spina bifida and cephalocele groups.

Table 1. Reproductive Factors^a

	Ex/Anencephaly	Spina Bifida	Cephalocele	Anencephaly/Craniorachischisis	Total
Maternal age, y					
20 - 29	22 (55)	16 (72.72)	9 (69.23)	3 (60)	49 (61.3)
30 - 35	12 (30)	4 (18.18)	4 (30.76)	0	20 (25)
> 35	6 (15)	2 (9.09)	0	2 (40)	11 (13.8)
Previous Abortion					
Yes	8 (20)	6 (27.27)	4 (30.76)	0	18 (22.5)
No	32 (80)	16 (72.72)	9 (69.23)	5 (100)	62 (77.5)
Gravidity					
Primigravid	15 (37.5)	11 (50)	5 (38.46)	1 (20)	32 (40)

^aValues are expressed as No. (%).**Table 2.** Risk Factors

Risk Factor	Ex/Anencephaly	Spina Bifida	Cephalocele	Anencephaly/Craniorachischisis
Incomplete folate consumption	23	6	5	2
Diabetes mellitus	1	0	0	0
Obesity (BMI > 30 kg/m ²)	3	0	0	0
Drug	1	0	0	0
Consanguineous marriage	12	3	7	3

Table 3. NTD Distribution and Gestational Age^a

Type of NTD	Number of Cases	Time of Diagnosis, w	
		11 th - 14 th	15 th - 20 th
Ex/anencephaly	40 (50)	37 (92.5)	3 (7.5)
Spina bifida	22 (27.5)	4 (18.18)	18 (81.81)
Cephalocele	13 (16.25)	8 (61.53)	5 (38.46)
Anencephaly/craniorachis	5 (6.25)	3 (60)	2 (40)
Total	80	52 (65)	28 (35)

^aValues are expressed as No. (%).

3.4. Ex/Anencephaly

Associated anomalies were found in 22 (55%) fetuses with ex/anencephaly.

The most common type of associated anomaly in this group was extremity anomalies (8 fetuses). Club foot/lower limbs contracture was observed in 6 fetuses. The other types included: club hand in one side and cutting of the other side (1 fetus) and bilateral clenched hands (1 fetus). Thoracic/abdominal wall anomalies were found in 6 fetuses including: abdominoschisis (one fetus), thoracoabdominal schisis (2 fetuses), and omphalocele (3 fetuses).

Cardiac anomalies were found in 4 fetuses including: hypoplastic left heart syndrome (2 fetuses), atrioventricular septal defect (1 fetus), ventricular septal defect, and bradycardia (1 fetus). In addition, face/neck anomalies were found in 4 fetuses including: hypertelorism and extensive facial defect (1 fetus); hypotelorism (1 fetus); retrognathia and unilateral microphthalmia (1 fetus); and microphthalmia and cataract (1 fetus). Finally, spine anomalies including scoliosis and segmentation abnormalities were found in 4 fetuses. Moreover, 4 fetuses had single umbilical cord, 1 fetus had genitourinary anomaly (megacystitis), 1 fetus had gastrointestinal anomaly (distended stomach and echogenic bowel), and 1 fetus had hydrops fetalis.

3.5. Spina Bifida

Associated anomalies were found in 20 (90%) fetuses with spina bifida.

Associated CNS anomalies were detected in 19 fetuses; the Chiari II in all cases of open spina bifida, which 5 cases had also ventriculomegaly. Associated non-CNS anomalies were found in 13 fetuses, in which the spine anomalies (kyphosis or scoliosis) were the most common type (8 fetuses). Extremities anomalies were found in 6 fetuses including: bilateral club foot (2 fetuses), unilateral club

Table 4. Location of Defect in Fetuses with Open Spina Bifida and Cephalocele

	Open Spina Bifida			
	Cervical	Thoracolumbarsacral	Lumbosacral	Sacral
Myelocele	1	3	3	1
Myelomeningocele	0	3	9	0
	Cephalocele			
	Occipital	Parietooccipital	Parietal	
Meningocele	2	0	0	
Encephalocele	9	1	1	

Table 5. Associated Anomalies

Associated Anomaly	Ex/Anencephaly (N = 40)	Spina Bifida (N = 22)	Cephalocele (N = 13)	Anencephaly/Craniorachischisis (N = 5)
Hydrops fetalis	1	0	1	0
Gastrointestinal	1	1	1	0
Genitourinary	1	2	1	1
Spine	4	8	2	0
Extremities	8	6	4	1
Heart	4	0	2	0
Cord	4	2	0	1
Face/neck	4	0	5	0
Thoracic/abdominal wall	6	3	2	1
CNS	-	19	2	-

foot (2 fetuses), bilateral club hand (1 fetus), and bilateral clenched hand (1 fetus). Moreover, thoracic/abdominal wall anomalies were found in 3 fetuses; 2 fetuses with abdominoschisis and the other with omphalocele. Two fetuses had genitourinary anomaly (1 with probable cloacal extrophy and the other with non-visualized kidneys), 2 fetuses had cord anomaly (both with single umbilical cord), and 1 fetus had hydrops fetalis.

3.6. Cephalocele

Associated anomalies were found in 10 (76%) fetuses with cephalocele.

Face/neck anomalies were the most common type in this group; accordingly, cystic hygroma (1 fetus), cleft palate (1 fetus), retrognathia and absence of NB (1 fetus), microphthalmia and cataract (1 fetus), and hypertelorism (1 fetus). Extremities anomalies were found in 4 fetuses; bilateral radial ray deformity (1 fetus), bilateral club foot (1 fetus), bilateral hand polydactyly (1 fetus), and severe micromelia (1 fetus). CNS anomalies were found in 2 fetuses; one with cerebellar hypoplasia and hydrocephaly and the other with vermian agenesis. Thoracic/abdominal wall

anomalies were found in 2 fetuses; 1 with severe narrow thorax and the other with omphalocele. Spine anomalies were found in 2 fetuses; 1 with spine segmentation defect and the other with absence of ossification of sacrum. Heart anomalies were found in 2 fetuses; 1 with hypoplastic left heart syndrome and the other with right heart dominance and coarctation of aorta. Ultimately, 1 fetus had genitourinary anomaly (polycystic kidneys), 1 fetus gastrointestinal anomaly (echogenic bowel), and 1 fetus hydrops fetalis.

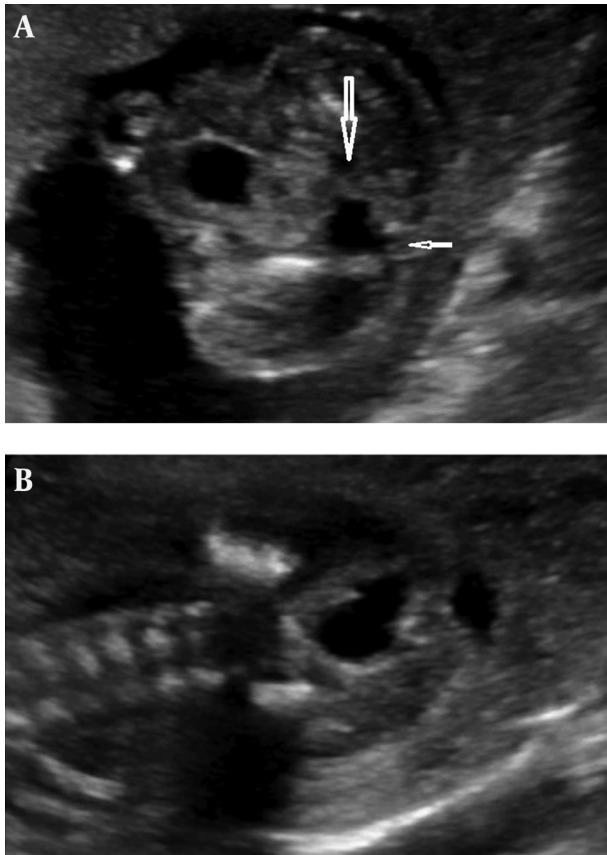
3.7. Iniencephaly/Craniorachischisis

Associated anomalies were found in 3 (60%) fetuses with anencephaly/craniorachischisis.

One fetus with single umbilical cord artery and omphalocele, 1 with club foot, and the last with bilateral pyelectasis were all diagnosed with craniorachischisis and none of the fetuses with anencephaly had associated anomaly.

3.8. Chromosome and Syndromes

In the current series, 9 fetuses were syndromic: 2 fetuses with amniotic band syndrome (1 with exencephaly,

Figure 1. Ultrasound Images of Anterior Sacral Meningocele at Mid-Gestation

Axial (A) and coronal view (B) are demonstrating a small cystic mass (large arrow) in presacral region communicating with spinal canal (small arrow).

the other with spina bifida) (Figure 2), 3 with limb-body-wall complex (1 exencephalic fetus and 2 spina bifida fetuses), 1 with the Meckel-Gruber syndrome (had occipital cephalocele), 1 with achondrogenesis type 2 (had occipital cephalocele), 1 with the Dandy-Walker malformation (had occipital cephalocele), and 1 with caudal regression syndrome (exencephalic fetus). Nearly all cases (except 1 case) of limb-body-wall complex/amniotic band syndrome were not truly defined by their previous examination and were referred due to multiple malformations.

Karyotype was evaluated only in 3 fetuses (2 fetuses with spina bifida and 1 with cephalocele), which were all reported as normal. All other cases referred in the 2nd trimester had low-risk 1st-trimester screening result. Autopsy was not performed on any fetuses.

4. Discussion

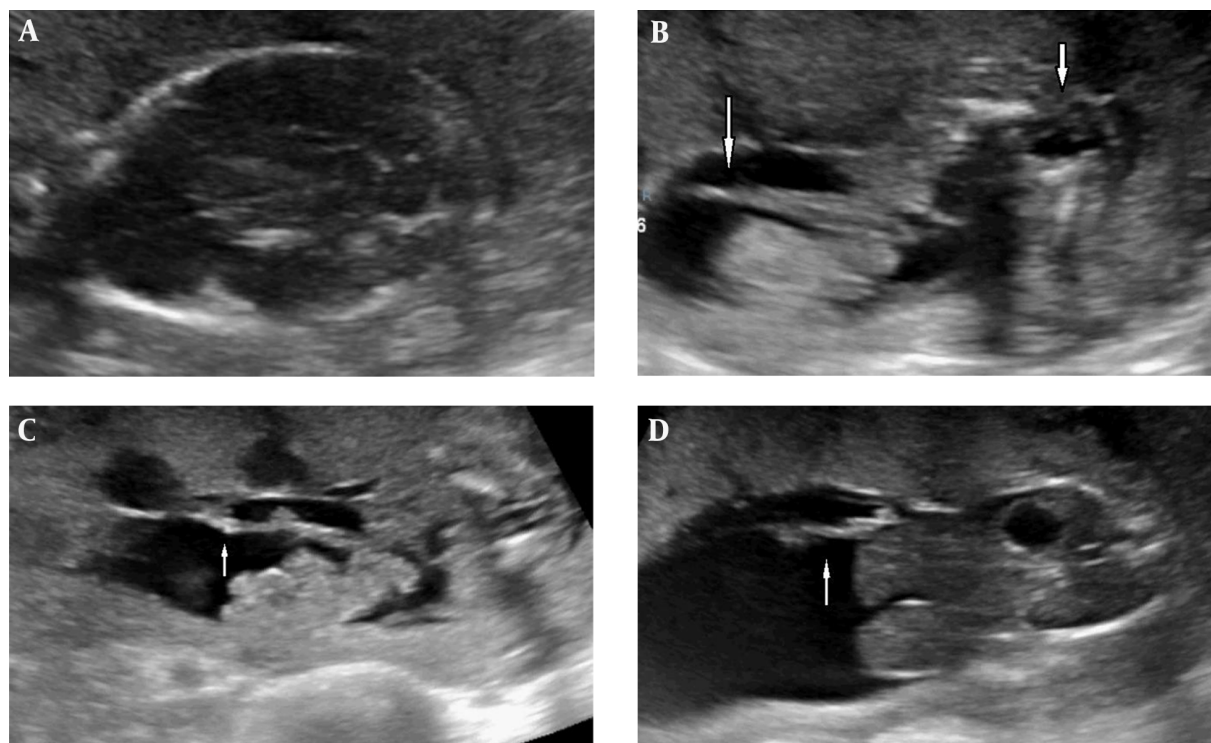
The current study provided detailed information on the risk factors, diagnostic process, distribution, associated anomalies, and syndromes in Iranian fetuses with NTDs.

Ultrasound screening is the non-invasive modality of choice for prenatal detection of NTD due to its safety, cost-efficiency, easy access of patients, and high detection rate. In addition to operator dependency, detection rate is significantly influenced by gestational age and type of NTD. For example, detection rate for spina bifida in 1st trimester ultrasound screening is 40%, while in the 2nd trimester screening it is 92% - 95% (11). On the other hand, the detection of ex/anencephaly is straightforward by demonstrating the absence of cranial vault; moreover, its detection rate in 1st trimester ultrasound is about 90% - 100% even in inexperienced operators (11, 12). Ultrasound specificity to diagnose open spina bifida is almost 100% by expert operators (13, 14). In the current study, 65% of the cases were detected in the 11th to 14th weeks of gestation, probably due to the routine NT evaluation. In Iran, the 1st trimester screening is recommended to all pregnant females. Early detection of NTD is helpful for obstetricians to make the proper plan and for parents to prepare themselves psychosocially and medically to have a disabled child or decide to terminate pregnancy.

The prevalence of NTD in Iranian population, based on previous studies in different parts of the country, is 28.7-50.1 per 10000 births with the most common types being spina bifida and anencephaly (15-17). In the current series, ex/anencephaly was detected as the most common type of NTD. Although the prevalence of anencephaly/craniorachischisis is reported as 1.5% of all NTDs (3), it was 6.25% in the current series. Moreover, 78 cases with abnormal findings in the 1st or 2nd trimester screening ultrasound were referred to the center for confirmation of the diagnosis and decision-making to terminate the pregnancy. Only one case was referred to the center due to positive quad test despite the normal ultrasound screening in a private center; moreover, 1 case was detected during routine sonography in the center.

In the current series, 61.3% of the cases were in the age group of 20 - 29 years and none of the cases had a maternal age under (18). In the ex/anencephalic group, 47.5% of the cases had maternal age of 30 years or more. History of previous abortion was positive in 22.5% and only 7.5% had previous aborted fetus with NTD.

In addition, 45% of mothers had incomplete folic acid consumption; more prevalence was recorded in ex/anencephalic fetuses and overall, most of them began folic acid consumption after pregnancy. This fact should alert

Figure 2. Ultrasound images of amniotic band syndrome and a lumbosacral myelomeningocele

A, Banana sign in head, (B) amniotic band (larger arrow) adherence to bowel loops and myelomeningocele (small arrow), (C and D) amniotic band adherence to bowel loops and extruded liver, respectively.

obstetricians and the health care system to provide an effective program for females of childbearing age.

The 2nd associated risk factor after incomplete folate consumption was the history of consanguineous marriage (31.2%). This finding was compatible with the results of studies conducted by Samadirad et al. in North-West Iran and Behrooz et al. in South-West Iran (15, 19).

In the current study, the proportion of cephalocele (16%) was similar to that of Nielsen et al. (18%), with the higher rate of anencephaly (50% vs 43%) and that of Pinar et al. in proportion of anencephaly cases (53%) (4, 18). Among the open spina bifida cases, lumbosacral (60%) was the most common location similar to those of Mennuti et al. and Nielsen et al. (4, 20). The rate of the current study myelomeningocele cases (40%) was higher in comparison with that of the previous study by Ghi et al. (3.7%) (21). Moreover, 75% of the cases referred to private centers only based on cranial signs without diagnosis of the location of myelomeningocele. This confirmed the importance of cranial signs to detect open spina bifida cases.

Based on ultrasound examination, all the anomalies previously reported in association with NTDs were

recorded in details (mainly based on autopsy findings). Anomalies assumed to be the consequences of the primary NTDs and various defects of syndromic cases were also included. This could be one of the reasons for the higher rates of associated anomalies (66.25%) in the current study in comparison with other studies (6-8, 10) and similarity to those of Nielsen et al. and Gale et al. (5). In 11.25% of these cases, the associated anomalies and NTDs were parts of a recognizable syndrome. Between the associated anomalies detected in the current series, CNS anomalies were the most common type (26.26%); compatible with the reports of Nielsen et al. (4), the Chiari II was found in all fetuses with open spina bifida, nearly similar to the findings of studies performed by Ozgun et al. and Ghi et al. (12, 21). The rate of ventriculomegaly in the current study (30%) was lower than that of previous studies by Ghi et al. (47%) and Nielsen et al. (64%) (4, 21), which could be due to lower gestational age of the current study cases. The 2nd common associated anomaly was extremity abnormalities (23.75%) with more prevalence in the spina bifida group (36.36%); club foot and lower extremity contracture were the most common anomalies. The 3rd common associated anomaly

was spine deformities (17.5%) with dominance of kyphoscoliosis.

Among the syndromic fetuses, more than 50% had limb-body-wall complex or amniotic band syndrome and its rate in all of the current cases was 6%. Only 1 of the cases was referred with true diagnosis of amniotic band syndrome, while in the remaining 4 cases, only a few defects were detected. These complex anomalies included defects in craniofacial, limb, and ventral wall. Between them, 3 cases had exencephaly and 2 cases had spina bifida. The most common associated syndromes were chromosomal abnormalities (15%) and the Meckel-Gruber syndrome (6%) in the study by Nielsen et al. while they were chromosomal abnormalities (3%) and the amniotic band syndrome (2.7%) in that of Stevenson et al. (4, 8). Among the other syndromic fetuses, only 1 (1.6%) was diagnosed with the Meckel-Gruber syndrome (presented with cephalocele, polycystic kidneys, polydactyly in both hands, and oligohydramnios as known parts of this syndrome), similar to that of Stevenson et al. (1.6%) (8). One fetus had the Dandy-Walker malformation in association with a small occipital cephalocele and ventriculomegaly; 1 fetus had caudal regression syndrome with lower limb contracture, echogenic bowel, and dilated stomach, and 1 fetus had achondrogenesis type 2 (confirmed by radiography of aborted fetus) with occipital cephalocele, cystic hygroma, severe micromelia, hydrops, delayed ossification of sacrum, and macrocrania.

Association of NTD with chromosomal abnormalities are reported as 9% to 16% in different studies (4, 18, 22, 23). Autopsy and karyotyping were recommended to all parents in the current study; however, only for 3 fetuses, karyotype was examined and no case underwent autopsy. It was the most important limitation of the current study and could be due to 3 factors in Iran, resulting in low recommendation by clinicians to do autopsy and karyotyping: 1) 1st trimester screening tests were performed in most of the current study cases with low risk result for chromosomal abnormalities; 2) The 2nd and more important reason was financial problems; 3) Limited awareness of parents and clinicians. These data showed that more efforts should be made to increase awareness in community and ask for help from government to overcome situations arising from financial limitations.

In conclusion, the current study reported the risk factors, diagnostic process, distribution, and associated anomalies in NTD cases. It showed the diagnostic capability of detailed ultrasonography to define syndromic processes and associated anomalies influencing prognosis, parent counselling, and planning. Limb-body-wall complex/amniotic band syndrome was the most common identified associated syndrome with higher incidence. The cur-

rent study results also emphasized the supplementation by folic acid as an important underlying risk factor.

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Footnote

Conflict of Interest: The authors declared no conflict of interest.

References

1. Kar A, Kar T, Kanungo S, Guru L, Rath J, Dehuri P. Risk factors, organ weight deviation and associated anomalies in neural tube defects: A prospective fetal and perinatal autopsy series. *Indian J Pathol Microbiol.* 2015;**58**(3):285–91. doi: [10.4103/0377-4929.162832](https://doi.org/10.4103/0377-4929.162832). [PubMed: [26275247](https://pubmed.ncbi.nlm.nih.gov/26275247/)].
2. Frey L, Hauser WA. Epidemiology of neural tube defects. *Epilepsia.* 2003;**44** Suppl 3:4–13. doi: [10.1046/j.1528-1157.44.s3.2.x](https://doi.org/10.1046/j.1528-1157.44.s3.2.x). [PubMed: [12790881](https://pubmed.ncbi.nlm.nih.gov/12790881/)].
3. Wilson RD, Sogc Genetics C, Special C. Prenatal screening, diagnosis, and pregnancy management of fetal neural tube defects. *J Obstet Gynaecol Can.* 2014;**36**(10):927–39. doi: [10.1016/S1701-2163\(15\)30444-8](https://doi.org/10.1016/S1701-2163(15)30444-8). [PubMed: [25375307](https://pubmed.ncbi.nlm.nih.gov/25375307/)].
4. Nielsen LA, Maroun LL, Broholm H, Laursen H, Graem N. Neural tube defects and associated anomalies in a fetal and perinatal autopsy series. *APMIS.* 2006;**114**(4):239–46. doi: [10.1111/j.1600-0463.2006.apm_325.x](https://doi.org/10.1111/j.1600-0463.2006.apm_325.x). [PubMed: [16689822](https://pubmed.ncbi.nlm.nih.gov/16689822/)].
5. Gole RA, Meshram PM, Hattangdi SS. Anencephaly and its associated malformations. *J Clin Diagn Res.* 2014;**8**(9):7–9. doi: [10.7860/JCDR/2014/10402.4885](https://doi.org/10.7860/JCDR/2014/10402.4885). [PubMed: [25386414](https://pubmed.ncbi.nlm.nih.gov/25386414/)].
6. Pelizzari E, Valdez CM, Picetti Jdos S, Cunha AC, Dietrich C, Fell PR, et al. Characteristics of fetuses evaluated due to suspected anencephaly: a population-based cohort study in southern Brazil. *Sao Paulo Med J.* 2015;**133**(2):101–8. doi: [10.1590/1516-3180.2013.8012608](https://doi.org/10.1590/1516-3180.2013.8012608). [PubMed: [25789781](https://pubmed.ncbi.nlm.nih.gov/25789781/)].
7. Sadovnick AD, Baird PA. Congenital malformations associated with anencephaly in liveborn and stillborn infants. *Teratology.* 1985;**32**(3):355–61. doi: [10.1002/tera.1420320305](https://doi.org/10.1002/tera.1420320305). [PubMed: [4082066](https://pubmed.ncbi.nlm.nih.gov/4082066/)].
8. Stevenson RE, Seaver LH, Collins JS, Dean JH. Neural tube defects and associated anomalies in South Carolina. *Birth Defects Res A Clin Mol Teratol.* 2004;**70**(9):554–8. doi: [10.1002/bdra.20062](https://doi.org/10.1002/bdra.20062). [PubMed: [15368552](https://pubmed.ncbi.nlm.nih.gov/15368552/)].
9. Chen CP. Chromosomal abnormalities associated with neural tube defects (1): full aneuploidy. *Taiwan J Obstet Gynecol.* 2007;**46**(4):325–35. doi: [10.1016/S1028-4559\(08\)60002-9](https://doi.org/10.1016/S1028-4559(08)60002-9). [PubMed: [18182338](https://pubmed.ncbi.nlm.nih.gov/18182338/)].
10. Kallen B, Robert E, Harris J. Associated malformations in infants and fetuses with upper or lower neural tube defects. *Teratology.* 1998;**57**(2):56–63. doi: [10.1002/\(sici\)1096-9926\(199802\)57:2lt;56::aid-tera3gt;3.0.co;2-4](https://doi.org/10.1002/(sici)1096-9926(199802)57:2lt;56::aid-tera3gt;3.0.co;2-4). [PubMed: [9562677](https://pubmed.ncbi.nlm.nih.gov/9562677/)].
11. Cameron M, Moran P. Prenatal screening and diagnosis of neural tube defects. *Prenat Diagn.* 2009;**29**(4):402–11. doi: [10.1002/pd.2250](https://doi.org/10.1002/pd.2250). [PubMed: [19301349](https://pubmed.ncbi.nlm.nih.gov/19301349/)].
12. Ozgun MT, Batukan C, Kaya D, Turkyilmaz, C, Basbug, M. Prenatal diagnosis of neural tube defects, evaluation of 112 cases. *Gynecol Obstet Reprod Med.* 2016;**15**(1).
13. Brand IR, Kaminopetros P, Cave M, Irving HC, Lilford RJ. Specificity of antenatal ultrasound in the Yorkshire Region: a prospective study of 2261 ultrasound detected anomalies. *Br J Obstet Gynaecol.* 1994;**101**(5):392–7. [PubMed: [8018609](https://pubmed.ncbi.nlm.nih.gov/8018609/)].

14. Antsaklis AJ. Debate about ultrasound screening policies. *Fetal Diagn Ther*. 1998;**13**(4):209-15. [PubMed: [9784640](#)].
15. Behrooz A, Gorjizadeh MH. Prevalence and Correlates of Neural Tube Defect in South West Iran: Retrospective analysis. *Sultan Qaboos Univ Med J*. 2007;**7**(1):31-4. [PubMed: [21654942](#)].
16. Talebian A, Soltani B, Sehat M, Zahedi A, Noorian A, Talebian M. Incidence and Risk Factors of Neural Tube Defects in Kashan, Central Iran. *Iran J Child Neurol*. 2015;**9**(3):50-6. [PubMed: [26401153](#)].
17. Gotalipour MJ, Mobasheri E, Vakili MA, Keshtkar AA. Epidemiology of neural tube defects in northern Iran, 1998-2003. *East Mediterr Health J*. 2007;**13**(3):560-6. [PubMed: [17687828](#)].
18. Pinar H, Tatevosyants N, Singer DB. Central nervous system malformations in a perinatal/neonatal autopsy series. *Pediatr Dev Pathol*. 1998;**1**(1):42-8. [PubMed: [10463270](#)].
19. Samadirad B, Khamnian Z, Hosseini MB, Dastgiri S. Congenital anomalies and termination of pregnancy in Iran. *J Pregnancy*. 2012;**2012**:1-4. doi: [10.1155/2012/574513](#).
20. Main DM, Mennuti MT. Neural tube defects: issues in prenatal diagnosis and counselling. *Obstet Gynecol*. 1986;**67**(1):1-16. [PubMed: [2415888](#)].
21. Ghi T, Pilu G, Falco P, Segata M, Carletti A, Cocchi G, et al. Prenatal diagnosis of open and closed spina bifida. *Ultrasound Obstet Gynecol*. 2006;**28**(7):899-903. doi: [10.1002/uog.3865](#). [PubMed: [17086581](#)].
22. Kennedy D, Chitayat D, Winsor EJ, Silver M, Toi A. Prenatally diagnosed neural tube defects: ultrasound, chromosome, and autopsy or postnatal findings in 212 cases. *Am J Med Genet*. 1998;**77**(4):317-21. [PubMed: [9600743](#)].
23. Sepulveda W, Corral E, Ayala C, Be C, Gutierrez J, Vasquez P. Chromosomal abnormalities in fetuses with open neural tube defects: prenatal identification with ultrasound. *Ultrasound Obstet Gynecol*. 2004;**23**(4):352-6. doi: [10.1002/uog.964](#). [PubMed: [15065184](#)].