ASSESSMENT OF PREVALENCE, TYPES ANDASSOCIATED RISK FACTORS OF NEURAL TUBE DEFECTS IN PREGNANT WOMEN VISITING HEALTH CENTERS IN ADDIS ABABA.

A thesis to be submitted to the research committee of the department of Radiology of college of health sciences, AAU as a requirement for partial fulfillment of post graduate program in Radiology.

By
Aga Legesse

Advisors:

Dr. Daniel Zewdineh, Associate Professor of Radiology, Department of Radiology, College of Health Sciences, AAU

Dr. YocabelGorfu, Assistant Professor of Radiology,Department of Radiology, College of Health Sciences, AAU

September 2019
Addis Ababa.

Acknowledgments

First I would like to forward my heart felt gratitude to my advisers who were there from the beginning of the research; for their continuous input and monitoring the progress of the research and constructive comments and suggestions during the carrying out of the research. It was privilege to be working with you. I would also like to forward my sincere gratitude to my fellow residents who diligently were responsible for performing anatomic scan during the data collection. The research would not have been realized without your active engagement. I would also like to thank the department of the radiology who allowed me to do this research and use various facilities the department provides. I'd also like to thank the research coordinator AfrahMohammedsani and those responsible for data collection and would like to appreciate the dedication and courage they showed during the undertaking of the research despite the various challenges we face on the way.

Contents

List of Graphs

List of Tables

Abbreviations

AChEAcetylcholinesterease

NTDNeural tube defects

OSB Open Spina Bifida

AFP Alpha Feto Protein

MS-AFP Maternal Serum Alpha Feto Protein

AF-AFP Amniotic fluid Alpha Feto Protein

BSD Brainstem diameter

BSOBD Brainstem occipital occipital bone diameter

CNS Central nervous system

CSF Cerebrospinal Fluid

WHO World Health Organization

MOM Multiple of median

TCD Trans cerebellar Diameter

IFA Iron with Folic Acid

FL Femoral Length

BPD Biparietal diameter

HC Head Circumference

AC Abdominal Circumference

IUGR Intrauterine Growth Restriction

Abstract

Introduction: Central nervous system (CNS) defects are the second most common congenital anomalies after congenital heart diseases. Neural tube defects (NTD's) are amongst the major CNS defects with significant public health burden and long term morbidity and functional impairment. Central nervous system (CNS) defects are the second most common congenital anomalies after congenital heart diseases. Neural tube defects (NTD's) are amongst the major CNS defects with significant public health burden and long term morbidity and functional impairment. Many literatures recommend offering screening for NTDs to all pregnant women. Amniotic fluid AFP (AF-AFP) and amniotic fluid acetylcholinesterease (AChE) are the primary biochemical tests performed on amniotic fluid for detection of open neural tube defects. The other modality used for screening of these defects is prenatal ultrasound scan.

Method: This hospital based cross sectional prospective study tried to show the utility of ultrasound and the prevalence and pattern of occurrence of these defects.About total of 956 pregnant mothers who came to the hospital were evaluated with ultrasound scan.

Results: We found a total of 6 open NTD's out of this group. There were 2 anencephaly, 1 Chiari II, 1encephalocele and 1 had both anencephaly and spinabifida. Only 55 % if the respondents took IFA supplement during pregnancy and of those who took the supplement 43% took it for a maximum of 1 week. Of the parameters we found TCD has a strong correlation with the calculated mean gestational age.

Conclusion and recommendation: The above findings reflect a high incidence of these defects. The low coverage of folic acid supplement as well as poorer socioeconomic factors all would contribute to the observed burden of the disease. Comprehensive screening and preventive strategies must be designed. Further studies and decisions policy maker level are necessary to improve the current figures.

Introduction

Central nervous system (CNS) defects are the second most common congenital anomalies after congenital heart diseases.Neural tube defects(NTD's) are amongst the major CNS defects with significant public health burden and longterm morbidity and functional impairment.Their prevalence in Americas ranges from 3.3 to 27.9 per 10,000 births.[1]Data from the low income countries is lacking. They are detected in about 300 000 neonates worldwide each year according to a WHO article[2]The risk of NTD rises to 20 to 30 per 1000 live birthsfor women with a previous infant with NTD. Three factors have played a significant role in the assessment and prevention of this disorder in developed countries:

- The widespread use of maternal screening programs to identify pregnancies at high risk
- Sonographic imaging combined with amniocentesis for diagnosis of affected fetuses
- Administration of folic acid supplements for prevention of the disorder.

NTDs can be classified based on embryological considerations and the presence or absence of exposed neural tissue, as open or closed types.Open NTDs also named Spina Bifida Apertafrequently involve the entire CNS (e.g., associated hydrocephalus, Chiari II malformation) and are due to failure of primary neurulation. Closed NTDs also termed spina bifida occulta are localized and confined to the spine (brain rarely affected) and result from a defect in secondary neurulation.

Encephalocele results from failure of the anterior neuropore to close during days 26-28 of gestation.Spinal dysraphisms constitute a heterogeneous group of congenital disorders of the spine and spinal cord due to aberrant formation of the midline mesenchymal, bony, and neural structures. They originate from abnormalities occurring during one of 3 embryonic periods. The first of these is gastrulation (at weeks 2-3), which involves the function of the intervening mesoderm in the initially bilaminarembryonic disk. The second is primary neurulation (at weeks 3-4) during which the neural ectoderm bends, and folds along the midline to form the neural tube. The third is secondary neurulation (during weeks 5-6) when an additional part of the neural tube is produced caudalto the posterior neuropore resulting in the formation of the tip of the conusmedullaris and the flumterminale.

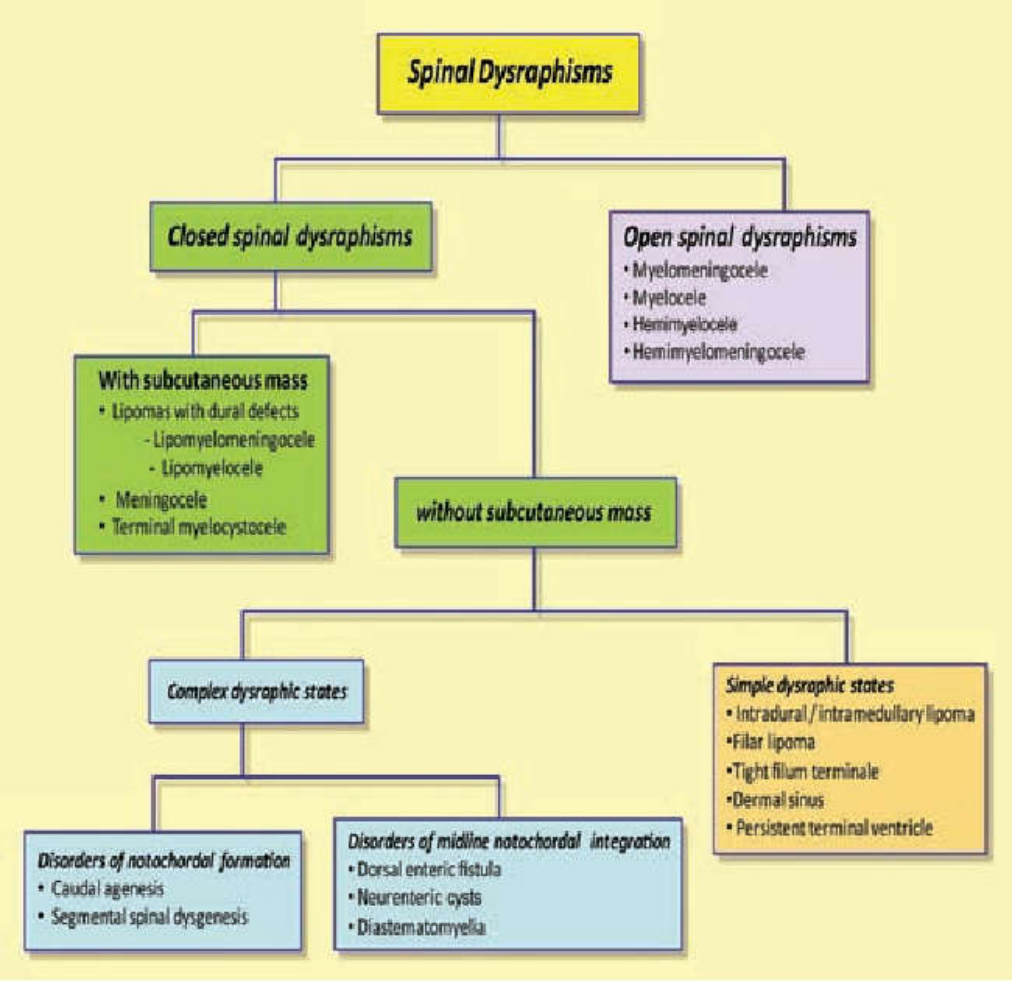


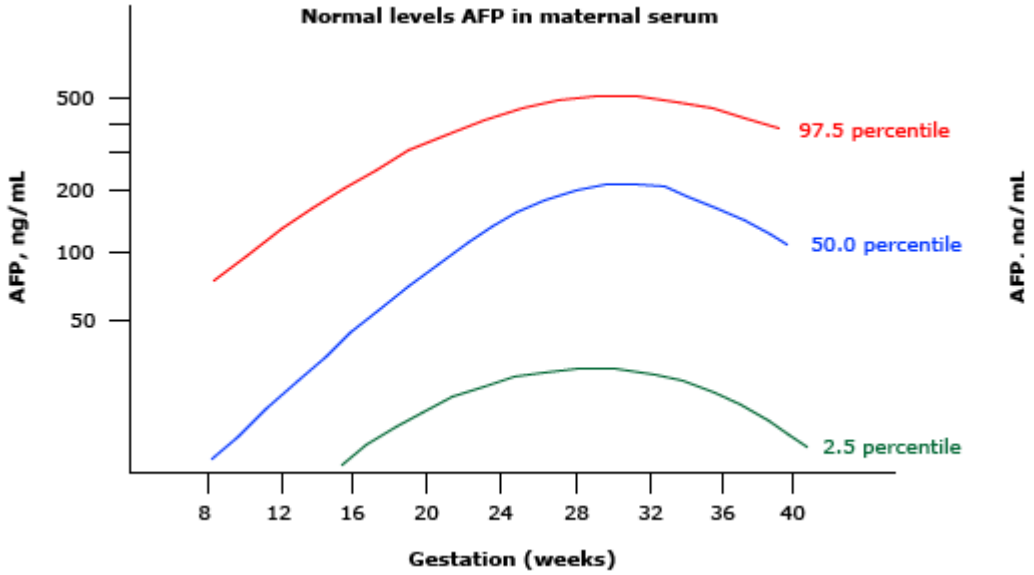
Figure 1 Classification of Spinal Dysraphisms[3]

Various factors have been implicated in their pathogenesis. Most notable are Maternal folate deficiency (folate sensitive NTD's), environmental factors which include drugs, excessive vitamin A intake, hyperthermia, maternal diabetes mellitus, obesity, and folate deficiency and genetic factors (high concordance rate in monozygotic twins, are more frequent among first degree relatives, and are more common in females than males). WHO recommends all women, from the moment they begin trying to conceive until 12 weeks of gestation, take a folic acid supplement (400 µg folic acid daily).Where as women who have had a fetus diagnosed as affected by a neural tube defect or have given birth to a baby with a neural tube defect should be offered high-dose supplementation (5 mg folic acid daily). A meta-analysis of randomized trials of folic acid for the prevention of recurrent NTDs demonstrated an 87% reduction in NTDs in women who took supplements before the start of pregnancy. Also based on observational studies

recurrence reduction of 85-100% were shown among those who took folic acid prior to subsequent pregnancies.[4]

Many literatures recommend offering screening for NTDs to all pregnant women. Amniotic fluid AFP (AF-AFP) and amniotic fluid acetylcholinesterease (AChE) are the primary biochemical tests performed on amniotic fluid for detection of open neural tube defects. AFP can be measured in maternal serum, amniotic fluid, and fetal plasma. According to Second report of the Collaborative Acetylcholinesterase Study which included 32,642 women with singleton pregnancies (including 428 with open spina bifida and 238 with anencephaly) who had an amniocentesis at 13-24 weeks' gestation an elevation of both AFP and AChE values suggests an open fetal NTD with 96 percent accuracy and a false positive rate of 0.14 percent.[5]

Some authors have suggested that the rate of detection of NTDs by ultrasound examination alone may preclude the need for amniocentesis. The American College of Medical Genetics recommends use of maternal serum alpha-fetoprotein and/or ultrasound for detection of neural tube defects between 15 and 20 weeks of gestation.ACOG recommends MSAFP screening at 15 to 20 weeks of gestation should be offered to all pregnant women as it is an effective method for detecting NTDs [13].AFP screening is primarily intended for the detection of open spina bifida and anencephaly, but can also uncover several non-neural fetal abnormalities



Graph 1- Graph- Approximate relationship between AFP values in maternal serum and gestational age.[6]

First elevated test may be repeated because as many as 30 percent of moderately elevated MSAFP results will be below the threshold level upon repeating the test and such findings are not associated with an increased frequency of false-negative NTD diagnoses.[7]If the elevation persists, then the next step is to obtain a specialized ultrasound examination to further assess whether a NTD, or other anomaly, is present.

Ultrasound examination is an effective modality for the prenatal diagnosis of these anomalies. An accurate fetal diagnosis depends upon a precise description of the sonographic appearance of the CNS and careful evaluation for associated malformations, which are often present. A thorough understanding of the normal sonographic appearance of the CNS at different gestational ages is crucial for accurate diagnosis because the presence or absence of a structure may be deemed normal or abnormal depending upon the age of the fetus.

In many places, traditional two-dimensional ultrasound has superseded MSAFP testing as a screening tool.Detection rates are influenced by gestational age and type of NTD. For example, first trimester studies generally have reported detection rates greater than 90 percent for anencephaly and 80 percent for encephalocele, but lower rates for spina bifida (44 percent). Second trimester ultrasound scanning increases the detection rate of spina bifida to approximately 92 to 95 percent.[8]

In a series of 366 patients identified as at risk for a fetal neural tube defect (NTD) before the 24th week of pregnancy, 64 had an abnormal fetus. The abnormalities included anencephaly (39), open spinal defect (17), closed spinal defect (2), encephalocele (1), and a miscellany of other abnormalities (5). An ultrasound examination prior to diagnostic amniocentesis positively identified all anencephalic fetuses, the fetus with the encephalocele, and 15 of the 19 fetuses with spina bifida. The spinal defects in 3 of the remaining 4 fetuses were demonstrated at a second examination.[9]

A retrospective population based study in southern Australia studied 243 births and terminations with NTD and for pregnancies with neural tube defects screened by any method (serum alpha-fetoprotein, ultrasound or amniocentesis), 86% sensitivity was achieved. Ultrasound screening

for anencephaly achieved 100% sensitivity even in low risk pregnancies, compared with 92% for serum alpha-fetoprotein. For spina bifida, the sensitivity of ultrasound screening increased with the level of risk in pregnancy: it was 60% in low risk pregnancies, which was equivalent to that of serum alpha-fetoprotein screening (64%); 89% in high risk pregnancies and 100% for women referred for confirmation of a suspected spina bifida by another ultrasonographer. [10]

Although the mainstay screening time for fetal defects is the routine 18-22 weeks GA ultrasound, helpful sonographic signs have been described for the diagnosis of these defects at much earlier gestational age; thus providing early option of treatment for parents and preventing possible complications.

A case based study demonstrated that 3-dimensional ultrasound allows for increased resolution of fetal surface anatomy, resulting in improved differentiation between fetal structures and early identification of developmental pathology. Using this technique a case of 9wks gravid patient with cystic lower pole fetal mass which made distinction using the conventional 2d gray difficult was subsequently diagnosed asmyelmeningocele. [11] However further studies (with appropriate sample size) need to be performed to uncover the true effectiveness of 3d technology in increasing early detection of these group of malformations.

R. Chaouiand K. H. Nicolaides described the intracranial lucency (IT) as an early marker;mainly used in 11-13+6 weeks; for the diagnosis of open spina bifida. They described in normal fetuses the fourth cerebral ventricle presents as an intracranial translucency (IT) parallel to the NT, while in fetuses with open spina bifida there may be absence of the IT. Themeasurement is taken as the antero –posteriordistanceof the maximum dimension of the 4th ventricle in the mid-sagittal plane.The measurement is preferably done with trans-abdominal scan. In the second trimester the manifestations of the Arnold-Chiari II; the cranial and cerebral signs like the lemon sign and banana signare the markers used to unmask OSB.[12]

The above described earlier signs may mark the future of sonographic screening, which might prove of great importance in early screening and diagnosis of affected fetuses thus improving the outcome of any therapeutic intervention or early patientmanagement decisions.

Ultrasound study in selected high risk pregnant women was shown to have higher diagnostic accuracy than routine ultrasound. In a survey that studied 2509 pregnant women who were

investigated between April 1, 1977, and March 31, 1980 (study 1), and April 1, 1980, and March 31, 1983 (study 2), to assess the effectiveness of diagnostic ultrasound in the diagnosis of neural tube defect (NTD) in women judged to be at high risk of NTD on the basis of raised serum α-fetoprotein or family history, or of other congenital malformation. The detection rate (sensitivity) for anencephaly was 100% in both studies. The detection rate for open spina bifida (OSB) improved from 33% in study 1 to 80% in study 2, specificity rose from 96% to 99%, the false-positive rate dropped from 57% to 9%, and the false-negative rate fell from 1% to 0·3%. Diagnostic ultrasound is likely to be conducted with greater vigilance and subjected to greater scrutiny than routine ultrasound because the patients have already been selected as being at high risk, the operators are more experienced, and their apparatus is likely to be more refined than that used for routine ultrasound.[13] Al these findings thus substantiate the need for pre evaluation risk stratification among pregnant women; thus maximizing the effectiveness/efficiency of the screening ultrasound.

There are also different sonographic signswhich are helpful in diagnosing open spina bifida at relatively later gestation. Careful search for these signs might sometimes be the only obvious clue suggesting an open defect.In a retrospective study which assessed the frequency of sonographic signs (cranial and cerebellar) in 70 fetuses with open spina bifida diagnosed by ultrasonography at 16-23 weeks' gestation found that biparietal diameter was below the 5th centile for gestation in 61% and head circumference measurements in 26%. The anterior horn of the lateral cerebral ventricle to hemisphere ratio was above the 95th centile in 77%, and ventriculomegaly of the posterior horn of the lateral ventricle was observed in 86%. In the 54 fetuses for which pictures were taken at the level of the biparietal diameter a scalloping of the frontal bones (the lemon sign) was seen. In 12 of 21 fetuses for which a sub-occipital bregmatic view of the cranium had been obtained, the cerebellar hemispheres were curved anteriorly with simultaneous obliteration of the cisterna magna (the banana sign); in a further 8 cases the cerebellum was not displayed. None of these changes was seen in 100 patients presenting consecutively for routine ultrasound examination in the second trimester. [14]

The sensitivity of sonographic diagnosis of other NTDS is high, but depends in part upon the size and location of the defect, the position of the fetus, the volume of amniotic fluid, maternal habitus, and the skill and equipment of the sonographer

Limitations including reverberation causing suboptimal visualization of the brain, maternal obesity, and oligohydramnios may hamper accurate sonographic diagnosis of fetal defects. In these case Fetal MRI have been shown to have important role in prenatal diagnosis. Further studies are still important to determine the role of MRI in uncertain US findings, but consideration of MRI is important for cases in which ultrasound imaging is not optimal or is uncertain.

Human birth defects remain a major public health burden.Neural tube defects are the second most common congenital defects as per previous epidemiologic studies. They are debilitating, even after surgical repair. Despite the fact that NTDs pose life-long challenges for patients, their families, and societies as a whole, research into their etiologies has lagged far behind that of other diseases.

This being said, despite the large magnitude of occurrence of these defects in our countrylittle has been done to uncover the true prevalence in our context and the possible factors contributing to the occurrence of the problem. Experience shows use of maternal screening programs to identify pregnancies at high risk, early diagnosis and administration of folic acid supplements has significant impact in reducing the magnitude of the problem and avoiding longterm disability in the affected newborns.

However there is little if any experience in systematic screening and Implementation of the preventionprograms of NTDs in our country. There are no previous data to evaluate the outcome of any intervention.

There were previous hospital based studies which try to investigate the incidence of neural tube defects among institutional births and description of the type of defects. No high quality studies have been done to study the role of ultrasound in screening and early diagnosis of these defects.

This researchhas tried to shade light on the prevalence of NTD in mothers visiting the teaching hospitals in Addiswith additional elucidation of patterns of occurrence. It also tried to answer the possible factors contributing to increased occurrence of NTD’s and give baseline insight forfurther investigation and planning future interventions as well as policy makers decision.

Research Methodology and Design

Study Setting and design:

The study was conducted at Black lion specialized hospital radiology department. The design of the study will be descriptive cross-sectional cohort study. Study period was between

Sampling method and sample size determination:

The sampling method will be non-probability sampling technique.

Sample size calculation was done with consideration of population prevalence of 1% and a margin of error of 5% and a 95% confidence level. Using the following single proportion formula; a sample size of 1250 was then formulated

$$n = \frac{z^2 p\,(1-P)}{d^2}$$

- n is the estimated sample size
- Z is the confidence level at a certain value of Significance
- P is the proportion of estimated incidence of neural tube defects from previous research
- d is the margin of error, expressed in proportion

Study population:

All pregnant mothers who come to the selected health centers from all sub cities for ANC follow up in and are within the gestational age specified (9-22 weeks) are included in the research. Pregnant mothers who are beyond the specified GA are excluded from the study.

Inclusion criteria:

All viable pregnancies within the specified Gestational age (9-22 weeks are included in the study).

Exclusion criteria:

Non-viable pregnancies.

Pregnancies with gestational ages beyond the specified range are excluded from the research.

Data collection, analysis and interpretation of Data

The data was collected through organized questionnaire which includes demographic, nutritional and radiologic parameters which were separate for the first trimester and the second trimester parameters. The questionnaire was administered to the mothers after gaining their consent and information regarding the socio-demographic, nutritional parameters as well as standard of living was entered by the trained data collector. Radiology residents were given training on how to perform targeted scans on screening for neural tube defects; based on parameters included in the questionnaire. They were responsible for handling the radiology part of the questionnaire and fill in the required measurements.Data was regularly checked for quality by giving training and supervision.Sonoscape SSI-8000was used with both low frequency as well as high frequency probes as per demand were usedto image patients.

The collected data was given specific Id number and data were coded, entered, cleaned and analyzed using SPSS version 25 software and descriptive analysis of the data done. Results are presented using tables and graphs.

Ethical considerations

The research was given approval by the Institutional Review Board of the College (IRB). Every pregnant mother who was enrolled on an individual basis, was provided with detailed information concerning the research objectives, benefits and the importance of participation and provided an informed consent before enrollment in the research. Confidentiality of the information was maintained. All information obtained from the research was kept strictly confidential and used solely for the purpose of the research. The mothers enrolled in the research got a formal standard obstetric evaluation and detailed anatomic survey if findings on the standard scan necessitated so. No incentives or benefits of any kind were used.

Results

Of the total number of patients enrolled for this study, about 956 fulfilled the inclusion criteria; with corresponding respondent rate of 76.5 % as calculated from the projected sample size rate.

Addisketema, Kirkos and AkakiKalitysub cities account for a little more than half of the pregnant women included in the study. Bole, Kolfe and Arada sub cities are the 3 sub-cities with lowest frequency of participants.

More than 3/4th of the women are between 20 and 30 years of age with minimum and maximum age of participants of 17 and 43 respectively with standard deviation of 4.48 years

Table 1: Sub city of study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Sub-city	Frequency	Percent
Addis ketema	304	33.63
AkakiKality	123	13.61
Arada	25	2.77
Bole	5	0.55
Gulele	104	11.50
Kirkios	127	14.05
Kolfe	11	1.22
Lideta	47	5.20
Nifas silk	62	6.86
Yeka	96	10.62
Total	905	100.0

Table 2: Age of study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Age	Frequency	Valid Percent	Cumulative Percent
1(0-15)	0	0	0
2.(16-20	70	7.7	7.7

3(21-25)	296	32.7	40.4
4 (26-30)	403	44.5	85.0
5 (31-35)	97	10.7	95.7
6 (36-40)	38	4.2	99.9
7 (41-45)	1	.1	100.0
Total	905	100.0	

Around 34% of participants have completed primary school while 30.6 % have completed secondary education. About 18.2 % have higher education and nearly 10% cannot read and write.Nearly half of the participants don't have means of income whereas about third of them are civil servants. Nearly 95% of the women in our study are married and living with their spouse where as a significant proportion (i.e. 4.2 % are single pregnant women).

About 95% of the participants are married; a figure correlating with better support for pregnancy. A significant proportion however are single mothers (4.2 %).

Table 3: Occupational and educational status of mothers which are included in the study at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019 and their husbands.

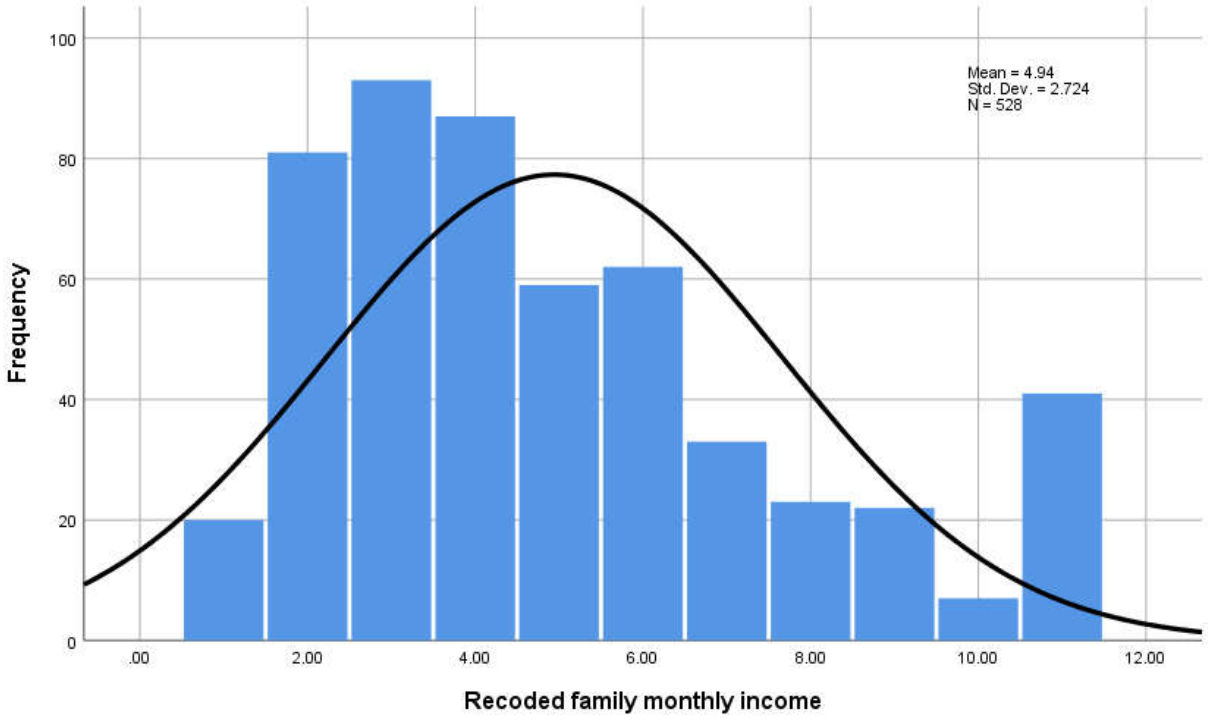
Mother educational status	**Frequency**	**Percent**
Can't read and write	89	9.83
Can read and write	67	7.40
Primary education completed	307	33.92
Secondary education completed	277	30.61
College/university	165	18.23
Total	905	100.0
Mother occupational status		
Unemployed	130	14.36
Student	9	0.99
Housewife	313	34.59
Daily labor	18	1.99
House maid	7	0.77
Merchant	25	2.76
Civil servant	259	28.62
Alcohol seller	6	0.66
Self employed	138	15.25
Total	905	100.0
Marital status		
Single	38	4.2
Divorced/separated	11	1.22
Widowed	0	0.00
Married/living together	856	94.59
Total	905	100.0
Husband educational Status		
Can't read and write	26	2.95
Can read and write	73	8.28
Primary education completed	243	27.55
Secondary education completed	311	35.26
College/university	229	25.96
Total	882	100.0
Husband occupational status		
Unemployed	12	1.36
Student	1	0.11

Daily labor	53	6.01
Merchant	88	9.98
Civil servant	407	46.15
Self employed	321	36.39
Total	882	100.0

In contrast to the women included in our study great majority the husbands of the participants have a personal means of income (>98%). Only about 1.5% of the husbands were unemployed or students. About half of the study participants gave an average monthly family income above 5,000 birr. About 3/4th of thestudy participants were orthodox Christians with Muslim participants coming second in frequency (14.5 %).

Table 4: family monthly income of study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Family monthly income			
Code and ranges of income	**Frequency**	**Valid Percent**	**Cumulative Percent**
1(0-999)	20	3.8	3.8
2(1000-1999	81	15.3	19.1
3(2000-2999)	93	17.6	36.7
4(3000-3999)	87	16.5	53.2
5(4000-4999)	59	11.2	64.4
6(5000-5999)	62	11.7	76.1
7(6000-6999)	33	6.3	82.4
8(7000-7999)	23	4.4	86.7
9(8000-8999)	22	4.2	90.9
10(9000-9999)	7	1.3	92.2
11(10 and above)	41	7.8	100.0
Total	528	100.0	



Graph 2: Family monthly income of study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019

Majority of the study participants i.e. 75% are orthodox Christians. Muslim and protestant Christians and second and third in order.

Table 5: Religion of study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Religion	Frequency	Valid Percent
Orthodox	687	75.91
Muslim	131	14.48
Protestant	86	9.5
Catholic	1	0.11
Total	905	100.0

About 38.3 percent the women gave a history of previous abortion. Of these 2/3rd had only one abortion while 24% had greater than 2 previous abortions.

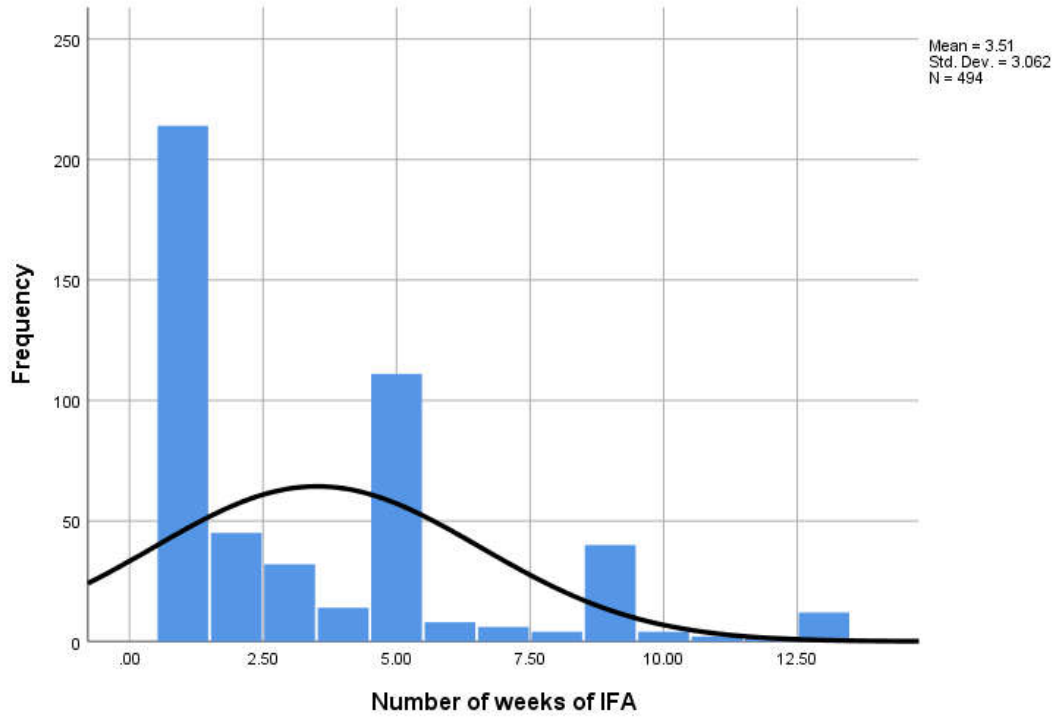
Table 6: Previous Abortion History of study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Did you have abortion			
Response	**Frequency**	**Valid Percent**	**Cumulative Percent**
Yes	252	38.3	38.3
No	406	61.7	100.0
Total	658	100.0	
Howmany abortions			
Number of Abortions	**Frequency**	**Valid Percent**	**Cumulative Percent**
1	163	67.6	67.6
2	44	18.3	85.9
3	18	7.5	93.4
4	6	2.5	95.9
5	3	1.2	97.1
6	1	.4	97.5
7	4	1.7	99.2
8	2	.8	100.0
Total	241	100.0	

Majority of the women (about 55 %) told that they took Iron and folate supplement during their current pregnancy. A substantial percent (43.3%) of the population took the supplement only for a maximum of 1 week. Of those who took the supplement about 48.3% of them said they took the supplement for more than 1 month.

Table 7: Status of intake of IFA supplementation during current pregnancy of study participants at TikurAnbessa specialized Hospital during Oct 2018 and Apr 2019.

Took IFA in Current pregnancy			
Response	**Frequency**	**Valid Percent**	**Cumulative Percent**
yes	497	55.0	55.0
No	406	45.0	100.0
Total	903	100.0	
Number of weeks of IFA			
Number of weeks	**Frequency**	**Valid Percent**	**Cumulative Percent**
1.00	214	43.3	43.3
2.00	45	9.1	52.4
3.00	32	6.5	58.9
4.00	14	2.8	61.7
5.00	111	22.5	84.2
6.00	8	1.6	85.8
7.00	6	1.2	87.0
8.00	4	.8	87.9
9.00	40	8.1	96.0
10.00	4	.8	96.8
11.00	2	.4	97.2
12.00	2	.4	97.6
85-100 days	12	2.4	100.0
Total	494	100.0	



Graph 3: Number of weeks of IFA study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

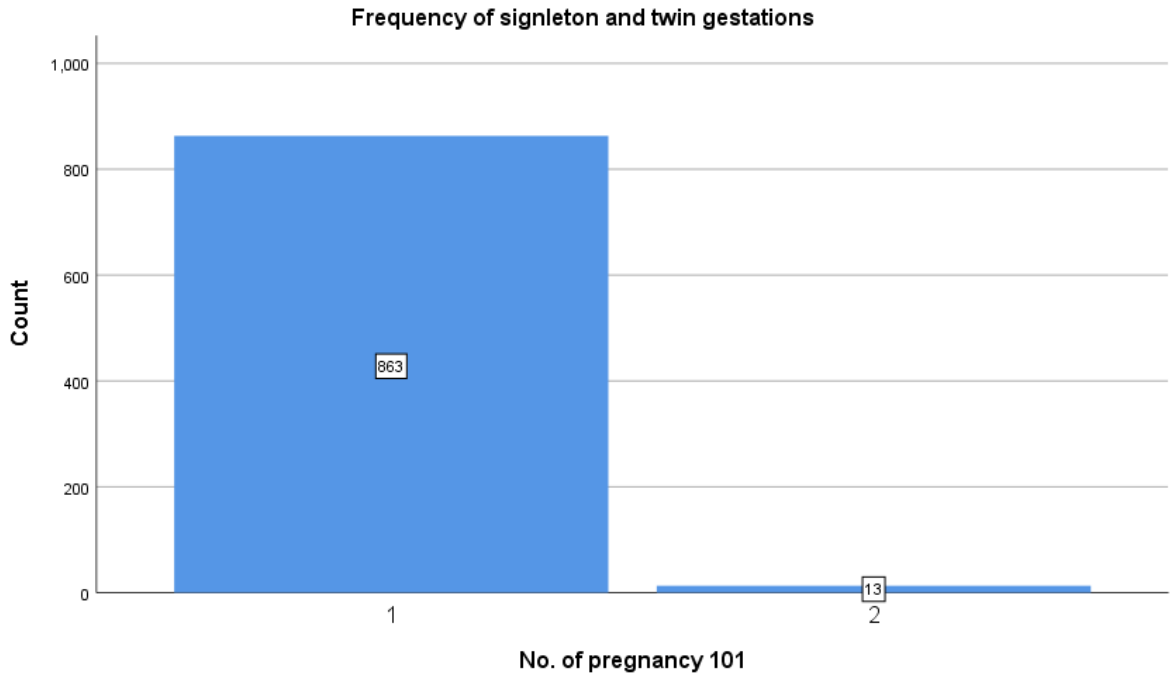
About 3 participants told that they had previous pregnancy with neural tube defect. This however not among the current cases in our study. Another 10 participants (1.1 %) gave a history of family member with pregnancy complicated with neural tube defect. None of these however had NTD in their evaluation with ultrasound. About 10 pregnant women are known diabetics (i.e. 1.1 % of respondents). None of these however have NTD during their current pregnancy.

Table 8: History of previous NTD and family history of NTD of study participants at TikurAnbessa specialized Hospital during Oct 2018 and Apr 2019.

Previous NTD			
Response	**Frequency**	**Valid Percent**	**Cumulative Percent**
Yes	3	.5	.5
No	549	99.5	100.0
Total	552	100.0	
Family member with NTD			
Response	**Frequency**	**Valid Percent**	**Cumulative Percent**

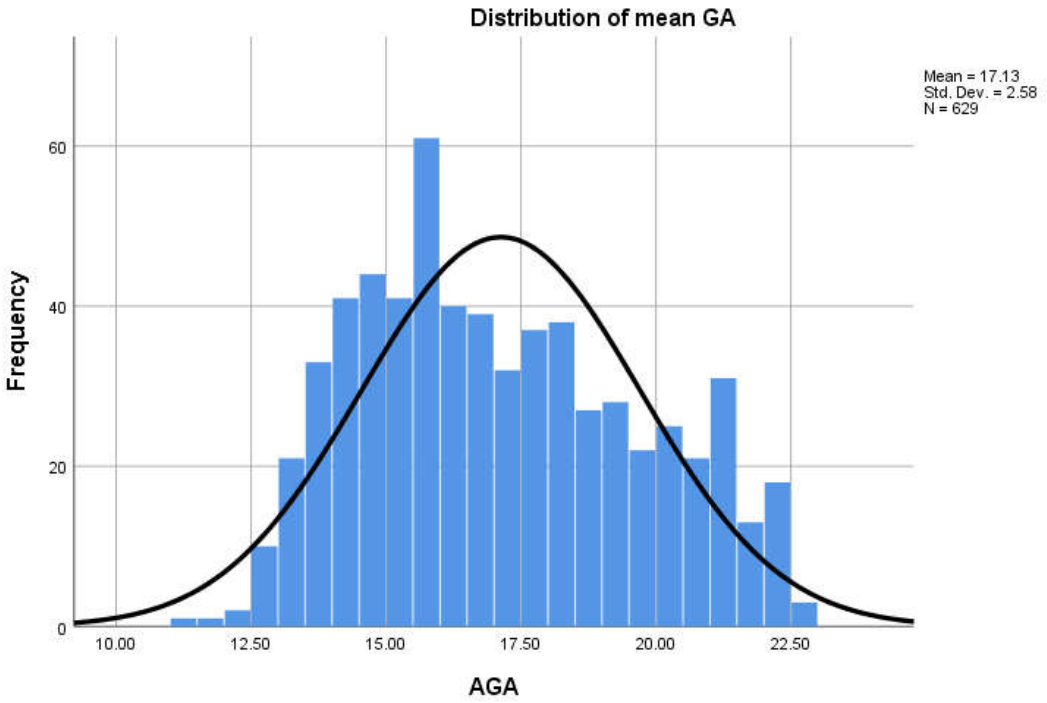
Yes	10	1.1	1.1
No	894	98.9	100.0
Total	904	100.0	

The gestational age of the fetuses during the study period as determined by theaverage of BPD, FL and HC. Out of the total 876 patients 863 had singleton pregnancies and 13 of them had twin gestations; giving a twinning rate of 1.48 %.



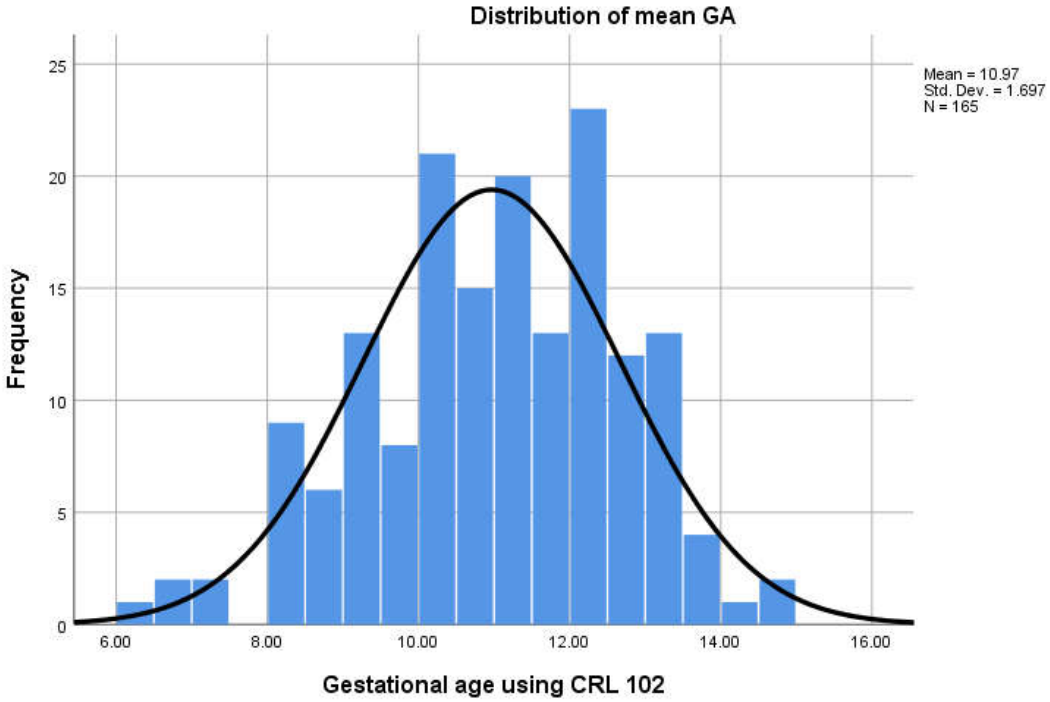
Graph 4: Number of pregnancy study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Mean average gestational age for second trimester patients was 17.12 weeks with standard deviation of 2.58 weeks.



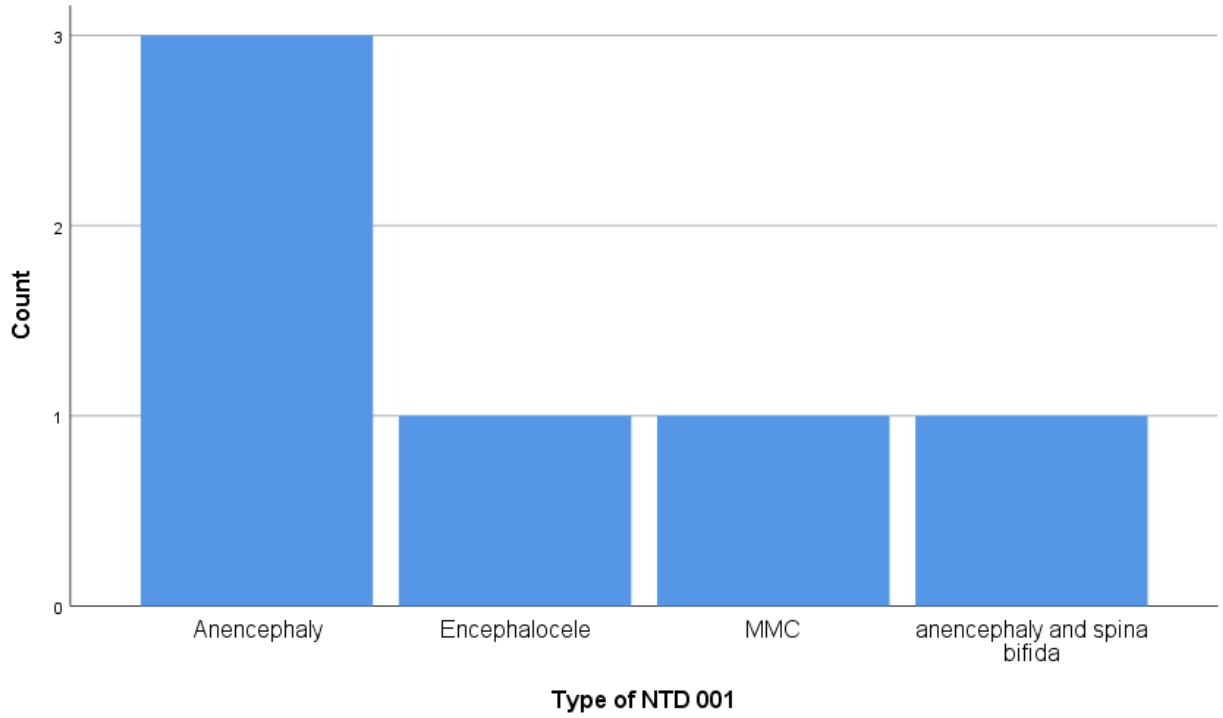
Graph 5: Distribution of mean gestational age for second trimester pregnancies study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Mean GA of those patients for whom CRL was used was 11.9 weeks with standard deviation of 0.6 weeks.



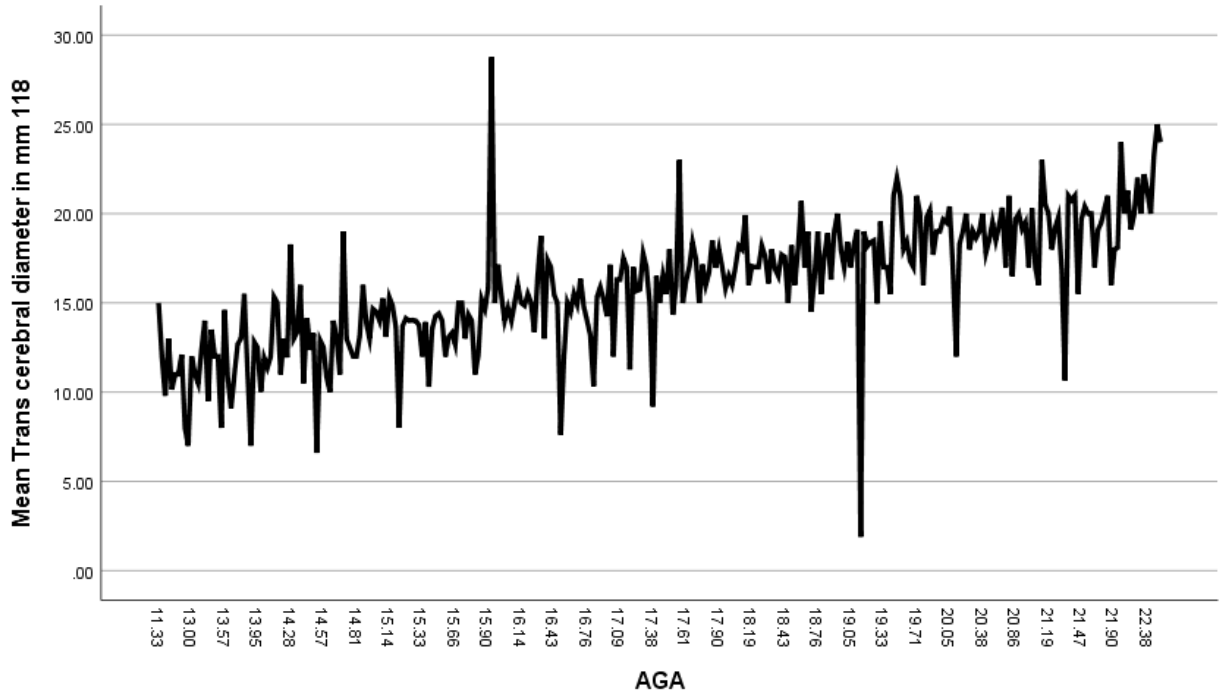
Graph 6: Distribution of mean gestational age for first trimester pregnancies study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Out of the total 876 patients for whom ultrasound evaluation was done during the study period 6 cases were identified as having neural tube defect (corresponding to 0.7 % incidence or 70 cases per 10,000). Out of the six cases identified 3 were cases of anencephaly, one had chiari II malformation, another one had both anencephaly with spina bifida and the other had occipital encephalocele.



Graph 7: Types of NTD's study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Trans-cerebellar diameter (TCD) showed positive correlation with average gestational age (p value of 0.01). Trans-cerebellar diameter showed linear relationship with the calculated average GA.



Graph 8: Trans-cerebellar diameter vs. Average gestational age study participants at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

(AGA: Average Gestational Age)

The mean and standard deviation for measurements of the lateral ventricle size at the thalamic level have comparable results.

Table 9: Lateral ventricle size of the fetus of women included in the study at TikurAnbessa specialized Hospital during Oct 1 2018 and Apr 30 2019.

Lateral ventricle size					
	N	Minimum	Maximum	Mean	Std. Deviation
Left ventricle size at thalamus 113b	676	1.20	12.00	6.2434	1.66238
Right ventricle size at thalamus in mm 113a	679	1.00	15.60	6.2783	1.80334
Total	904	94.5	100.0		

Discussion

The three sub-cities (namely Addis Ketema, AkakiKality and Kirkos) accounted for great majority of the participants. Meanwhile bole,Arada and KolfeKeranio sub cities were among the least contributors (with cumulative percentage of 4.5 percent).This variation may be attributable to socioeconomic variability amongst the localities.As the screening ultrasound is done free of charge; it is expected sub cities with lower socioeconomic group would have greater percentage of participants. This discrepancy may have its own implication in generalization of the research results to the city population at large. No of subjects as well as proportion of participants is limited in number for adequate description of differences among the sub cities thus searching for possible local contributing factors.

The percentage of women who cannot read and write (i.e 10 %) is indicative of a fairly higher rate of illiteracy among the study participants. About third of the participants only finished primary education; another indicator of poor literacy of participants. This can be taken as an important contributing factor for occurrence of birth defects. The data for the husbands shows a higher level of education for males; about quarter of them finishing a higher (college/University) education.

A significant proportion of the study participants are young women; about 7.7 are teenagers while a little over 40 % of the pregnancy, women are below the age of 25 years. As usual this data is subject to bias due to mis-registration of participant age. The majority of the participants' young age might be a soft indicator for unmet need for family planning and increasing the likelihood of lack of proper support for the pregnancy.

About 95% of the participants are married; a figure correlating with better support for pregnancy. A significant proportion however is single mothers (4.2 %).

According to the 2015/2016 national absolute poverty line was 7184 per year per adult person (food poverty line was 3772 per year per adult person). About 10.1% of the population is living below the national poverty line (National plan commission; an interim report on 2015.2016 poverty analysis study.)

About a little over third of the women gave history of previous abortion. This is aggregate figure of the induced and spontaneous abortion. This high rate of abortion is another indicator of an unmet need for contraception and other potential contributing factors should be sought.

WHO recommends daily folate supplementation for pregnant women; routine dose of 0.4mg for low risk and higher dose for those with higher risk (Eg: Previous NTD, drugs associated with increased occurrence of NTD's). It is recommended that it be started before conception (about one month according to some guidelines). (WHO.-Daily iron and folic acid supplementation during pregnancy). In our study only 55% of the women took Iron with folate supplement and of these nearly half (43%) took the supplement for a maximum of 1 week. This is indicative of poor coverage of the supplement and the figure would be expected to be lower for the rural or smaller towns where there is poor literacy and health access. Supplementation of Iron with and folic acid has additional benefits including prevention of maternal anemia, puerperal sepsis' low birth weight as well as preterm delivery. This is underlines the urgency to give due notice for expanding the current low coverage of the supplementation.

About 876 pregnant women were included in our study and a total case of 6 corresponding to a prevalence of 70 cases per 10,000 population was found. The burden of the disease represented by this figure might not be a true representative of nationwide prevalence; thus giving a false impression of a total prevalence comparable (even lower) to the rest of African estimates or worldwide prevalence. Women in the city have comparatively better access to health care and education which have significant impact in predicting the occurrence of neural tube defects.

Of the total pregnant women included in our study we've found about 6 cases a figure corresponding to a prevalence of 0.7 % or 70 cases per 10,000. The findings were communicated with the respective mothers with explanation given regarding the outcome of the pregnancy. Spouses were involved when available. They were then sent to their respective institutions to get the appropriate care and for possible referral where their referring center didn't provide the options of treatment. This figure is a little lower than the findings in a prospective cross-sectional hospital-based case-control study done in three hospitals in the city; namely TikurAnbessa specialized Hospital (TAH), Zewditu Memorial Hospital (ZMH) and Ghandi Memorial Hospital (GMH) in 2016.The frequency of NTD's was calculated to be 116 out of 10,000 births after 12 weeks of pregnancy. Planned pregnancy, Male sex, normal or underweight BMI, Folate/multivitamin supplement during the first trimester were found to correlate with reduced

risk of occurrence of NTD's. Out of the total cases of NTDs, 54.1% were cases of anencephaly; 40.5%,spina bifida; and 5.4%, encephalocele. Diagnosis of NTDs was made by ultrasound before birth for 96/111 (86.5%) cases, while the remaining cases were diagnosed at birth even though there was at least one ultrasound scan during ANC. Anencephaly was also the commonest Type of NTD in our research. There are however small number of total cases to extrapolate the relative frequency of the types of NTD's for the population at large. [15]

Another retrospective cross sectional study in 2 hospitals in the city; namely TikurAnbessa specialized hospital and Ghandimemorial hospital conducted between 2009 and 2012; obstetric neonatal records were evaluated and a frequency of the incidence of NTD's was 61 per 10,000 deliveries. 12% of the cases were diagnosed before 28 weeks gestational age and mean age at diagnosis was 33.8 weeks. About 72% were diagnosed with ultrasound before delivery. About 85% of the pregnant women with baby with NTD never received folic acid supplementation while only 1 % (2/177) received pre-conceptional folic acid. Myelomeningocele contributed for a little over half of the NTDs. Anencephaly contributed to 43.5% of the NTDs followed by encephalocele and meningiocele. [16]

In another hospital based cross sectional research done to assess the prevalence of NTD's in Tigray an overall incidence of 131 cases per 10,000 population was found among both live and still births. Anencephaly was still a little over thanspina bifida in incidence. Majority of the cases however had male sex in this research.[17]

A 10 –year retrospective case-control study of NTDs In a federal teaching hospital in south east Nigeria between January, 2006 and December, 2015 18 NTDs out of 21, 067 births giving a prevalence rate of 0.85 per 1000 births. Neural tube defects were significantly more common among the neonates of un-booked rural women, women who suffered from diabetes mellitus and those who used herbal medicines during the pregnancy. Spina bifida was the most common type of NTD in this study.

A systematic review to estimate the world wide prevalence of NTD's amongst live births ;which included 75 countries indicted a large variation among the regions of the world (between range: 1.2–124.1 per 10,000 births) approximately 80% of reported prevalence estimates above 6.0 per 10,000 births. Lowest median prevalence value (6.9 per 10,000 births) were registered for western pacific region. Among studies that included spina bifida and at least one other NTD, the lowest prevalence was 3.3 per 10,000 births. Among African Region eight countries were

included (11 studies). The lowest reported NTD prevalence for the region was reported in Nigeria (5.2 per 10,000 births) and the highest was reported in Algeria (75.4 per 10,000 births) [19]. The median NTD prevalence was 11.7 per 10,000 births. Data from this region were primarily gathered from hospital-based retrospective case reviews. [1]

A facility based cross-sectional study was conducted in eight randomly selected major public hospitals from the Tigray region of Ethiopia, Out of the 14,903 births (live and stillbirths) during the study period, a total of 195 (126 males and 69 females) infants had NTDs. The incidence of infants with anencephaly and spina bifida was 66.4 and 64.4 per 10,000 births, respectively. The overall occurrence of NTDs was 131 per 10, 000 births (95% CI, 113–150.6). Anatomical location of the spina bifida cases was cervical 29% (28/96), thoracic 28% (27/ 96) and lumbar 43% (41/96) regions, respectively.

Trans-cerebellar diameter showed strong positive correlation with mean gestational age (as calculated average of the other 3 parameters in our study). This is in agreement with a hospital based study done in Nigeria which showed than TCD correlated more significantly with menstrual age than the other biometrics as well as higher predictive accuracy as compared with FL, BPD and AC. [18]In another hospital based study in India which included women of gestational ages 15-40 weeks; for normal pregnancy as well as in IUGR pregnancies mean difference between estimated and actual gestational age was minimum in TCD as compared to other parameters; still underlining the important role of TCD in predicting gestational age even with IUGR fetus. Sub division of the gestational age ranges showed correlationbetween TCD measurements and gestational age was strong at 16-20, >20-24 and >24-28 weeks. It wasmoderate at >24-28 weeks and mild at >32-35 and >36-40 weeks. [19]

Conclusion and recommendation

The estimated magnitude of incidence of neural tube defects from our research is a figure equivalent (even higher) with highest reported rates among other African countries and the rest of the world. The low coverage of IFA supplement during pregnancy and the observed low literacy rate as well as poor socioeconomic factors can be considered potential factors contributing to these findings. Studies to elucidate the true burden of the defects within the country at large are important to plan further preventive strategies and guide the way for studying possible contributing factors among different regions within the country. Poor coverage of peri-conceptional IFA supplement is an issue that demands urgent attention and devising comprehensive strategy to improve the coverage is critical.

Limitations of the study

The findings of the study might not be applicable to the general population taking into consideration the socio-cultural, economic and environmental as well as genetic variability of the country's population and the type of sampling used. As the study is being conducted in Addis Ababa where the relative access for early prenatal counseling and early supplementation of folate has better coverage than the other parts of the country, the findings of this research might not show the real magnitude of the problem nationwide

Lack of high quality ultrasound machine as well as limited experience with the use of ultrasound for screening is a limiting factor to screen for signs of occult NTD's. Thus true incidence of aggregate of both types of NTD's may be higher than the figure we found.

Women participating in our study might not be representative of the real socioeconomic status of the population of the city. Groups with better income are expected to seek care from the private institutions with better quality care.

The relatively lower sample size affects the accuracy of overall estimated incidence as well as the pattern of occurrence and correlation with different contributing factors.

.

References

1. Zaganjor, I., et al., *Describing the prevalence of neural tube defects worldwide: a systematic literature review.* PloS one, 2016. **11**(4): p. e0151586.
2. Santos, L.M.P., et al., *Prevention of neural tube defects by the fortification of flour with folic acid: a population-based retrospective study in Brazil.* Bulletin of the World Health Organization, 2016. **94**(1): p. 22.
3. Salih, M.A., W.R. Murshid, and M.Z. Seidahmed, *Classification, clinical features, and genetics of neural tube defects.* Saudi medical journal, 2014. **35**(Suppl 1): p. S5.
4. Grosse, S.D. and J.S. Collins, *Folic acid supplementation and neural tube defect recurrence prevention.* Birth Defects Research Part A: Clinical and Molecular Teratology, 2007. **79**(11): p. 737-742.
5. Wald, N., H. Cuckle, and K. Nanchahal, *Amniotic fluid acetylcholinesterase measurement in the prenatal diagnosis of open neural tube defects. Second report of the Collaborative Acetylcholinesterase Study.* Prenatal diagnosis, 1989. **9**(12): p. 813-829.
6. Habib, Z.A., *Maternal serum alpha-feto-protein: its value in antenatal diagnosis of genetic disease and in obstetrical-gynaecological care.* Acta Obstetricia et Gynecologica Scandinavica, 1977. **56**(sup61): p. 1-91.

7. Wald, N.J., et al., *Maternal serum-alpha-fetoprotein measurement in antenatal screening for anencephaly and spina bifida in early pregnancy. Report of UK collaborative study on alpha-fetoprotein in relation to neural-tube defects.* Lancet (London, England), 1977. **1**(8026): p. 1323-1332.
8. Cameron, M. and P. Moran, *Prenatal screening and diagnosis of neural tube defects.* Prenatal Diagnosis: Published in Affiliation With the International Society for Prenatal Diagnosis, 2009. **29**(4): p. 402-411.
9. Robinson, H.P., et al., *Diagnostic ultrasound: early detection of fetal neural tube defects.* Obstetrics and Gynecology, 1980. **56**(6): p. 705-710.
10. Chan, A., et al., *The sensitivity of ultrasound and serum alpha-fetoprotein in population-based antenatal screening for neural tube defects, South Australia 1986–1991.* BJOG: An International Journal of Obstetrics & Gynaecology, 1995. **102**(5): p. 370-376.
11. Forest, C.P., D. Goodman, and R.G. Hahn, *Meningomyelocele: early detection using 3-dimensional ultrasound imaging in the family medicine center.* J Am Board Fam Med, 2010. **23**(2): p. 270-2.
12. Chaoui, R. and K. Nicolaides, *From nuchal translucency to intracranial translucency: towards the early detection of spina bifida.* Ultrasound in Obstetrics and Gynecology: The Official Journal of the International Society of Ultrasound in Obstetrics and Gynecology, 2010. **35**(2): p. 133-138.
13. Roberts, C., et al., *Diagnostic effectiveness of ultrasound in detection of neural tube defect: the South Wales experience of 2509 scans (1977-1982) in high-risk mothers.* The Lancet, 1983. **322**(8358): p. 1068-1069.
14. Nicolaides, K., et al., *Ultrasound screening for spina bifida: cranial and cerebellar signs.* The Lancet, 1986. **328**(8498): p. 72-74.
15. Gedefaw, A., S. Teklu, and B.T. Tadesse, *Magnitude of neural tube defects and associated risk factors at three teaching hospitals in Addis Ababa, Ethiopia.* BioMed research international, 2018. **2018**.
16. Sorri, G. and E.M. Kassa, *PATTERNS OF NEURAL TUBE DEFECTS (NTDs) AT TWO TEACHING HOSPITALS IN ADDIS ABABA, ETHIOPIA A THREE YEARS RETROSPECTIVE STUDY.* Ethiopian medical journal, 2015. **53**(3).
17. Berihu, B.A., et al., *High burden of neural tube defects in Tigray, Northern Ethiopia: Hospital-based study.* PloS one, 2018. **13**(11): p. e0206212.
18. Adeyekun, A. and M. Orji, *Predictive accuracy of transcerebellar diameter in comparison with other foetal biometric parameters for gestational age estimation among pregnant Nigerian women.* East African medical journal, 2014. **91**(4): p. 138-144.

19. Dashottar, S., et al., *Transcerebellar diameter: an effective tool in predicting gestational age in normal and IUGR pregnancy.* International Journal of Reproduction, Contraception, Obstetrics and Gynecology, 2018. **7**: p. 4190.

ANNEXES

ADDIS ABEBA UNIVERSITY CHOLEGE OF HEALTH SCIENCES

SCHOOL OF MEDICINE DEPARTMENT OF RADIOLOGY

A QUSTIONAIRE ON ASSESMENT OF LATE FIRST TRIMESTER NEURAL TUBE DEFECT

Section 16: Ultrasound results

A. Obstetric ultrasound, 9-12 wks			
No.	**Measurements**	**Coding**	**Remark**
1601	No of intra-uterine pregnancy	_____	
1602	Gestational age using CRL length (mm), (for each fetus if multiple)	_____	
1603	Is there is any cystic mass besides the yolk sac?	1. Yes 2. No	
1604	Is trans cranial lucency visible?	1. Yes 2. No	
1605	Is the cisterna magna effaced?	1. Yes 2. No	
1606	Nuchal translucency distance- (mm)	_____ mm	
B. Obstetric ultrasound between 18 - 22			
1607	Gestational age (for each fetus if multiple)	BPD _____ FL _____ HC _____	
1608	Is the supra orbital skull vault formed?	1. Yes ⟶ 2. No	If Yes, skip to **Q 1610**
1609	If No,	1. Cerebral parenchyma absent 2. Present but disorganized	

1610	Is there is any spinal defect in continuity?	1. Yes 2. No →	If No, skip to **Q 1612**
1611	If yes, length of spinal defect (in terms of number of spine)	_____	
1612	If yes to 1608, describe the shape of frontal bone	1. Oval 2. Concave anterior or lemon shaped	
1613	Lateral ventricle atrium size at the level of thalamus (mm)	1. Right _____ mm 2. Left _____mm	
1614	Third ventricle size	1. Slit like____ 2. If dilated (mm)____	
1615	Shape of posterior horn of lateral ventricle	1. Right a. Pointed b. Round 2. Left a. Pointed b. Round	
1616	Cerebral cortex	1. Normal → 2. Abnormal	Skip to Q 1618
1617	If abnormal, specify the type of abnormality		
1618	Trans cerebellar diameter (mm)	_____mm	
1619	Cerebellar shape	1. Normal 2. Crescent shape 3. Other, Specify_____	
1620	Cisterna magna	1. Normal appearing 2. Effaced	
1621	Herniation of cerebellar tonsil below foramen magnum:	1. Yes 2. No	
1622	Is there is any spinal defect?	1. Yes 2. No →	If No, skip to Q 1626
1623	If yes,	1. Anterior	

		2. Posterior	
1624	If yes,	1. Cervical 2. Thoracic 3. Thoraco-lumbar 4. Lumbar 5. Lumbo-sacral	
1625	Is overlying skin	1. Intact 2. Defective	
1626	Is there is any overlying cystic mass in communication with spinal canal through the defect?	1. Yes 2. No →	If No, skip to Q 1630
1627	If yes, Size of the sac	1. Length_____mm 2. Width_____mm 3. AP diameter_____mm	
1628	If yes, Contents of the sac	1. Anechoic 2. Contains neural tissue	
1629	If yes, Underlying spinal cord appears	1. Normal 2. Tethered 3. Flattened	
1630	Is there is sign of syringomyelia?	1. Yes 2. No →	If No, skip to Q 1633
1631	If yes, location?	_____	
1632	If yes, Length (no. of spinal vertebra)	_____	
1633	Is there a soft tissue mass posterior to spine?	1. Yes 2. No →	If No, skip to Q 1635
1634	If yes, size?	1. Length_____mm 2. Width_____mm 3. AP_____mm	
1635	Level of conusmedullaris	1. At or above L3	

		2. Below L3	
1636	If the spine is normal for above anomalies, is there is sign of isolated spinal cord tethering?	1. Yes 2. No	
1637	Any other systemic defect?	1. Yes 2. No →	
1638	If yes, specify	1. MSK 2. CVS 3. Respiratory 4. GIS 5. Hepatobiliary 6. GU 7. Others Specify _____	