ADDIS ABABA UNIVERSITY

COLLEGE OF HEALTH SCIENCE, SCHOOL OF MEDICINE

DEPARTEMENT OF ANATOMY

Prevalence of Neural Tube Defect and Associated Risk Factors at Debre Berhan Referral Hospital, North Shewa, Ethiopia, 2019.

Hospital based Case Control study

BY: ZERIHUN KINDIE (BSc)

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ACRONYMS AND ABBREVIATIONS

AAU	Addis Ababa University
ANC	Ante Natal Care
BMI	Body Mass Index
CI	Confidence Interval
CNS	Central Nervous System
DBRH	Debre Berhan Referral Hospital
DM	Debates Militias
FA	Folic Acid
HTN	Hypertensive
Hx	History
ICU	Intensive Care Unit
MRN	Medical Record Number
MTHFR	Methylene Tetrahydrofolate reductase
NGOs	Non-Governmental Organizations
NTD	Neural Tube Defect
OR	Odds Ratio
P-Value	Probability Value
SES	Socioeconomical Status
SPSS	Statistical Package for Social Sciences
UTI	Urinary Tract Infection
WHO	World Health Organization

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ABSTRACT

Background*: -* ***Neural tube defect (****NTD) is a structural defect of the central nervous system that affects the brain, spine and spinal column of the developing embryo during the first month of developmental process and most common congenital malformations, are potentially preventable cause of perinatal morbidity and mortality**. Worldwide, around 10% of infant mortalities are due to nervous system defects. It is estimated that approximately 300,000 babies are born each year with NTDs worldwide. In Ethiopia there is very limited published data regarding the prevalence and associated risk factors as well as established preventive strategy of NTDs. Therefore, the purpose of this study was to assess the associated risk factor and prevalence of NTDs among pregnant outcome in Debreberhan Referral Hospital.*

Objective:- *To assess the prevalence and risk factors of NTDs among pregnant outcome at DBRH, North Shewa, Ethiopia, 2019.*

Methods:- *Hospital based unmatched case control study was conducted among 50 cases (neonate born with NTDs) and 100 controls (neonate born without NTDs) for associated risk factors and retrospective cross sectional, descriptive study of registration of all birth outcome for prevalence of NTDs were revised. The study was carried out at DBRH from August30/2017 to August 30/2019.*

The data was checked for completeness and consistencies, and cleaned, coded and entered using Epi data version 4.2 and was exported to statistical package for social sciences (SPSS) software version 20 for analysis. Variables having P<0.05 was consided as statistically significant

Results*:-* *The total prevalence of NTDs was 10.9 per 1000 pregnancy and the prevalence of each NTD type was Anencephaly 5.6 per 1000 pregnancies, spina bifida 3.5 per 1000 pregnancies, encephalocele 0.9 per 1000 pregnancies and both spina bifida and anencephaly 0.7 per 1000 pregnancies. Logistic regression analysis showed that maternal age, < 20 (P=0.046), maternal age 31-35 (P =0.028), no education (P=0.014), Family history of NTDs (P=0.034), history of multiparous (P=0.002), history of abortion (P=0.024) and still birth (P=0.009), No ANC follow up, un planned pregnancy (P=0.000), history of medical illness (P=0.001), Hyperthermia (P=0.000), used antipyretics drugs (P=0.002), pericoceptional folic acid and Oral contraceptive use (P=0.001), maternal exposure to smoking (P=0.001) and pesticides (P=0.000), and use of local alcohol "Arki" (P=0.000) had significant association with NTDs.*

Conclusion*:-* *the prevalence of neural tube defects in this study is among the highest globally reported and family history of NTDs, coffee and alcohol consumption, maternal no education, maternal hyperthermia and disease, maternal antipyretic use, maternal age <20 and 31-35, exposure to pesticides and smoking were associated with the increasing the risk of NTD. Comprehensive preventive strategies focused on the identified risk factors should be established and early preiconceptional maternal screening for genetic factors and medical illness as well as periconceptional folic acid supplementation are the effective possible approaches to bring about the required reduction in NTD*

Key words: - Neural tube defect, Folic acid, maternal factors, Central Nervous System

1: INTRODUCTION

1.1 Background

Birth defects are congenital anomalies that are present at birth of the baby, and can be structural, functional, or metabolic. Structural birth defects are those that affect the development of body part. Birth defects usually begin during organogenesis. Birth defects may be visible before birth, at birth or later in life, and are major causes of children's hospital admissions and deaths.(1)

Neural tube is part of the central nervous system (CNS), which begins to form during the third week post conception. The formation of the nervous system starts with the formation of the neural plate, a thickening of ectoderm located along the middle dorsal region of the embryo. In humans, the neural plate develops into the neural tube via a two-step process i.e. primary and secondary neurulation. During primary neurulation, the lateral edges of the neural plate elevate as cells differentiate and migrate toward the neural plate edges to form the neural folds. As the neural folds continue to elevate, they meet at the midline and fuse to form the neural tube and primary neurulation and closure of the neural tube concluded by four weeks post conception. Secondary neurulation or canalization occurs at the caudal end of the primary neural tube when mesenchymal cells form a space that connects to the lumen of the primary neural tube. Failure of the neural tube to close at different regions results in the clinical variation seen in the anatomical location NTDs. These defects are classified as open and closed type, Open NTDs occur when the neural folds fail to meet and fuse during primary neurulation, whereas incomplete secondary neurulation leads to closed NTDs, in which the neural tissue is exposed to the environment or covered only by a membrane (e.g. anencephaly and spinal bifida encephalocele, meningocele), whereas closed NTDs, the neural tissue is not exposed and the defect is covered by skin (e.g. Lipomylomeningocele, lipomeningocele) among all NTDs spinal bifida and anencephaly are the two most common forms (2).

NTDs are structural defects of the central nervous system that affects the brain, spine and spinal column of the developing embryo during the first month of developmental process and they are most common congenital malformations, are potentially preventable cause of perinatal morbidity and mortality(3).

It is caused through multifactorial disorders, arising from a complex combination of genetic determinants and environmental factors involving nutritional deficiencies, parental education, maternal ages and occupations, smoking, alcohol consumption, maternal reproductive history, a previous NTD affected pregnancy, use of anti-seizure medication, obesity, and maternal use of caffeine and traditional medication, and exposure to radiation and hyperthermia during early pregnancy (4).

Worldwide, around 10% of infant mortalities are due to nervous system defects. However, the incidence of neural tube defects (NTD) is coming to decline in recent years in industrialized countries, while it still remains high in the less developed countries of Latin America, Africa, the Middle East and Far East Asia (5). NTDs is estimated that approximately 300,000 babies are born each year with NTDs worldwide (5).

Studies which were done Texas–Mexico border, that have a high occurrence of NTDs are more common in community who have risk factors of folic acid deficiency, B12 deficiency obesity, or diabetes (6).

Unlike developing countries, including Ethiopia the identification of the risk factors (maternal nutritional deficiency, chemical exposure, medical and fever illness and life style) in decreasing the burden of NTDs is well established in the developed world (7).

1.2 Statement of the problem

NTDs are serious birth defects that occur when the neural tube fails to close properly. Any woman who is capable of becoming pregnant could have risk of NTD-affected pregnancy. It is impossible to tell which women will have a pregnancy affected by NTD. Ninety five percent of women with NTD-affected pregnancies have no personal or family history (8).

Globally it occurs widely among diverse populations, due to varying levels of economic development, and geographic difference. Contrary to the declining mortality and morbidity due to diarrhea and infection, under five morbidity and mortality due to birth defect is increasing among low and middle income countries. Nevertheless, data on the prevalence of NTDs are limited in low income countries despite the WHO resolution on birth defect surveillance (9).

Screening tests, periconceptional consumption of folic acid and vitamin, and folic acid fortification in staple food in many countries have been proven to reduce the risk of NTD affected pregnancy by 50% to 70%. However, a large number of NTDs cases still occur and the cause is idiopathic. Therefore better understanding and additional research of risk factors will improve interventions aimed at reducing NTD prevalence (10, 11) .

There is deficiency of data mainly in low and middle income countries concerning on associated risk factors. This lack of data comes from unimproved recording system, poor diagnostic capacity, and lack of capability of countries to capture birth defects and established birth defect surveillance system. In sum-up, prevalence estimates related to birth defect may not involve birth defects related to stillbirths, elective terminations and aborted fetus because no organized data is collected these types of pregnancy losses. As a result, the global tool of birth defects estimation is poorly estimated.

In Ethiopia there is very limited published data regarding the prevalence and associated risk factors as well as established preventive strategy of NTDs. Specifically, there is no documented data on associated risk factor and prevalence of NTDs in North Shewa particularly in Debrerbrhan Referral Hospital.

Therefore, the purpose of this study was to assess the prevalence of NTDs and associated risk factors among pregnancy outcome at Debrebrhan Referral Hospital.

1.3 Significance of the study

Worldwide, there is information gap regarding risk factor, prevalence and preventive strategy about NTDs in addition to absence of well-organized data recording mainly in Asia, Africa and Latin America, so this study give the benefit to become one part of information regarding NTDs in Sub-Saharan Africa, particularly in Ethiopia.

In Africa, studies mainly focused on identifying the possible risk factors involved as contributor factors for NTDs are still low, so this study can have great contribution to fill the information gap.

For Ethiopia, this study would give a fertile soil of information concerning NTDs to change police and program implementing through fortification of food with folic acid, counseling and screening as well as health education.

For all, this study was provided pertinent information on associated risk factors and prevalence of NTDs and expected to be important inputs to heaths planners, policy makers, in their end overs to monitor and prevents NTDs along with associated risk factors. This may be true for governmental as well as Non-Governmental Organizations (NGOs).

The study might also asses the health professional in designing interventional projects mean to improving maternal and child health.

As the first study conducted on NTDs in North Shewa, this study was also gave base line data on prevalence and preventive strategy on associated risk factors of NTD for North Shewa area as well as the nation at large. The study will also become baseline data for others investigator who want to search further on related issues.

2. LITERATURE REVIEW

2.1 Prevalence of NTD

Worldwide, more than 10% of neonatal mortality is caused by embryological malformation of the nervous system (12) and it is estimated that more than 300,000 babies are born each year with NTDs, resulting in approximately 88,000 deaths and 8.6 million (2).

Although the incidence of NTDs has declined recently in the developed nations, Incidence of NTDs in developing countries has been increased to be up to fourfold higher than in developed ones (13). The most common NTDs cases are Anencephaly and spina bifida and anencephaly is a fatal NTDs type, but babies with spina bifida often survive following surgical interventions (14).

One scientific survey from eighteen countries in six world health organization (WHO) regions related that the prevalence of the NTDs based on live births to be 1.67/1000 births for total NTD prevalence, 1.13/1000 births for spina bifida, 0.25/1000 for anencephaly and 0.15/1000 for encephalocele in low and middle-income countries (15).

Some evidence suggest that presence of modern technology which leads to early detection and termination of NTDs, improvement of folic acid supplementation and better socioeconomically status/living standard leads to reduction in prevalence of NTDs worldwide (16).

In Africa, the reported incidence of spina bifida was variable, for example, it was 0.47/1000 births (17), 1.74/1000 births (18), and 3.48/1000 births in Malawi, Cape town, and Sudan (19) respectively .

In Ethiopia, hospital based cross sectional and unmatched case control study at Addis Ababa three Teaching Hospital show that NTDs prevalence is 55/8677 (20) , and a retrospective chart review from Addis Ababa teaching hospitals revealed the overall prevalence of NTDs to be 6.1/1000 births (21).

Another case control study which was done in Tigray region suggest that prevalence of anencephaly in 6.6/1000,spina bifida in 6.4/1000 and encephalocele in 0.67/1000 and the total prevalence is 13.8/1000 live births(7).

2.2. Associated risk factors for developments of NTDs

Congenital abnormalities can develop at any time of the pregnancy, from conception to birth. But frequently can develop at the time of organogenesis periods (from third –ninth weeks).

Neural tube defects are commonest malformations of the brain and spinal cord. Genetic factors, maternal exposure to environmental factors (hazards), maternal sociodemographic factors, maternal medical condition and drug uses, have some contribution for developments of NTDs, but number of cause is unknown (22).

Among contributory factors genetic and environmental factors have a great impact on development of NTDs, but nutritional deficiency, obesity and drugs have small contribution for occurrence of NTDs (23).

2.2.1 Socio Demographic Factors

Many study show that NTD risk is higher among families of lower SES, nutritional deficiency that come through poverty and poverty related problems could predispose these mothers to NTDs affected pregnancy. Furthermore maternal age, parental low educational status, female gender of the offspring, occupational and residential exposure to chemical pollutants are also incriminated as risk factors for NTDs (24). According to study in Catania, Italy identified that the average maternal age at birth was 27.9 years (7.5% of cases < 20 years; 24.5% were 20±24 years; 25.5% were 25±29 years; 28.7% were 30±34 years; 12.8% were 35±39; and only 1% were more than 40 years old) (25).

Hospital based case–control study conducted at Genoa, Italy, shows that maternal low educational level (OR=4.87), median annual family income (OR=3.35), maternal age < 25 years (OR=3.36) and >35 years; (OR=5.21), second and third birth order (OR=2.15) and (OR=3.93) respectively were among the significant risk factors of NTDs (24).

A study in California indicate that lower SES and residence in lower SES neighborhood increase the risk of having a neural tube defect-affected pregnancy, (OR =1.5to2.4) (26) .

Studies done in Riyadh and Algeria hospitals show that female gender of offspring's had great NTDs prevalence than male (36.6% and70% respectively) and maternal age 25-29 was associated to higher predisposition to NTDs (27).

The case control study on maternal risk factor and associated factor in Tigray region identified that maternal age >35 (OR=2.5, P=0.004) have a significant association with NTDs (7). Other study was done in Addis Ababa three teaching hospitals on magnitude and risk factor of NTD show that family annually cash income (P= 0.032), pregnancy BMI (P=0.041) and annual cash family income less than $1,300 USD (OR, 2.5; 95%, 1.2–5.5), $1,300–1,800 USD (OR, 2.8; 95%, 1.3–5.8), and $1,801–2,700 USD (OR, 2.6; 95%, 1.2–5.8) was found to be risk factors compared to income greater than $2,700 USD (20).

2.2.2 Genetic Factors

Neural tube defects (NTDs) are common, severe congenital malformations whose cause involves multiple genes and environmental factors. Methylene tetrahydrofolate reductase (*MTHFR*) plays an important role in folate metabolism. The *MTHFR*677 TT Geno-types in infants and mothers have been associated with increased risk for NTDs (19).

Some genetic syndromes such as Trisomy 13, Trisomy 18 and other aneuploidy are thought to be associated with NTDs. Positive family history of mother also can have 2% -5% rise of risk of occurrences of NTDs and 50 fold increase from the general population (28).

2.2.3 Maternal Nutrition and Folic Acid Consumption

Studies showed that being overweight/obese in women was significantly associated with an increase of NTD's rate. Glucose homeostasis affect neural tube closure and it is indicated that mothers with poorly regulated glucose levels are likely to have changes in the intrauterine environment. That can be the cause to abnormal organogenesis**.** Massive consumption of sucrose-containing and high-glycemic index foods was associated with an increased risk of NTD, especially among obese women (29).

Maternal folate status have a great role in determining the risk of NTDs. Folates are integral to intracellular one carbon metabolism, which produces pyrimidines and purines for DNA synthesis.. The micronutrient form of folate 5-methyltetrahydrofolate (MTHF) which circulates in plasma, is the physiologically active form of folate that serves as a cofactor for enzymatic reactions. Demands for folate increase during pregnancy because it is also required for growth and development of the fetus. Maternal supplementation with folic acid during pregnancy reduces NTD frequency whereas reduced serum folate and/or elevated homocysteine (an inverse indicator of folate status) is observed in some mothers of NTD affected fetuses, and is considered risk factors for NTDs (28).

WHO recommends periconceptional folic acid supplementation, however, studies showed that many women still do not follow the recommendations, particularly women of low socioeconomic status (30).

A case-control studies conducted in Italy and Algeria shows that significant association to NTDs was observed in mothers who didn't take pre-conception folic acid (OR=27 CI, 9.31–78,& 86%) respectively (24, 27).

According to study conducted in Iran, maternal obesity (OR: 5.4, CI: 1.3-21.8) was significantly associated with NTD. A six year period hospital based case-control study conducted among all pregnancies with NTD affected newborns (n=91) in Kasha, center of Iran, indicated that maternal obesity (OR: 5.4) was significantly associated with NTDs (24).

The study done in three teaching hospital of Addis Ababa show that preconception folic acid supplementation (P: 0.033), or only 7.8% were folic acid supplementation (20). Other study that was done at two hospital of Addis Ababa and hospital based study of Tigray region suggest that 85.3% (151/177), only less than 1% (2/177) of the mothers started taking folic acid supplementation pre-conceptionally and none of the respondents had a practice of periconceptional folic acid supplementation in Tigray region respectively (7, 20).

2.2.4 Maternal Reproductive History

Studies shows that maternal reproductive history (history of abortion, still birth, early neonatal loss, multiparty) is associated with increase of risk factors for NTDs. According to Catania, Italy study multiparous (84.8%), spontaneous abortion (21%), still birth (16%), early neonatal death (4.2%) are

associated with mothers whose birth outcome are NTD (14). A 4-year period case-control study conducted at King Khalid University Hospital, Riyadh conclude that significantly higher proportion case mothers had ,history of stillbirth (P=0.02) (5).

A six year period hospital based case-control study conducted among all pregnancies with NTD affected newborns (n=91) in Kashan, center of Iran, indicated that maternal history of abortion (OR: 4.9), was significantly associated with NTDs (31).

Hospital based case control study in Tigray suggest that the maternal age, and residency, birth order 3 and 4, unplanned pregnancy, history of breastfeeding above 2 years, history of stillbirths, history of male gender predominance fetus were found to have a strong association with an occurrence of NTDs (p = 0.000) (7, 21). Study that was done in three hospital of Addis Ababa show that there is significant association between multiparty (P: 0.042) and NTDs (20).

2.2.5 Maternal Chemical Exposure

Chemicals used as pesticides (insecticides, herbicides, and fungicides) can cross the placenta and impact embryonic development. In animal studies, these chemicals have been shown to alter neuroepithelial cell proliferation and differentiation during neurulation and lead to excessive neuroepithelial cell death that adversely impacts closure of the neural tube. Pesticide exposure can occur in both home and the workplace receiving (2).

With adjustment for maternal education, smoking, and folate intake, women who reported using pesticides in their homes or yards were two times more likely to have NTD-affected pregnancies than women without these reported exposures. Increase of pesticide exposure can increase risk of NTDs. The adjusted ORs and 95% CIs for one, two, and three or more exposure sources were 1.2 (0.69–1.9), 2.3 (1.3–4.1) and 2.8 (1.2–6.3) respectively, and this positive trend was stronger for risk of anencephaly than of spina bifida (31).

Italian study of use of pesticides or solvents (OR=10.62), residence near waste sites or polluting industries (OR=3.57) were proved to be a risk factor for spina bifida (25).

One case control study in Zewditu Referral Hospital show that maternal external exposure to cigarette smoke was strongly associated (OR 2.49 CI 1.15 -5.11 P=0.02). Significantly strong association was revealed for ≥3 cups/day caffeine intake (OR 8.14 CI 4.02 -16.4 P .000) and coffee was the most common source of caffeine (OR 34.17 CI 12.08 -96.6 P.000) (32).

2.2.6 Maternal Febrile Illness

Maternal hyperthermia can arise from either febrile illnesses or external exposure to heat. A recent meta-analysis evaluated that the NTD risk was increased almost three fold in cases of maternal fever during the first trimester. Therefore, maternal fever in early pregnancy as a risk factor for NTD-affected pregnancies and also heat that adversely affects development, can leads to increased cell death, decreased

proliferation, disruption of gene expression, and damage to the embryonic vasculature, results induction of apoptosis, inhibition of proliferation/ slow differentiation (33, 34).

Case control study in California show that maternal febrile illness episode in the first trimester was associated with an increased risk for having a NTD-affected pregnancy (OR 1.91 CI) and (OR 2.02) respectively(35) . Study of Northern China shows that history of a fever (OR 3.36), use of antipyretic drugs (OR 4.89,) were associated to NTDs (36).

A case control study in Zewditu Referral Hospital, Ethiopia suggest that maternal periconceptional hyperthermia was associated to NTDs risk (OR 65.5, CI 4.48 – 957.9 P .002) (32).

2.2.7 Maternal Medical and drug factors

Maternal pregestational diabetes mellitus is an important risk factor for the development of CNS anomalies. It has been found to cause a 2-fold to 10-fold increase in risk of CNS malformations among the offspring of affected women, relative to the general population (37).

Maternal epilepsy is associated with a 1% to 2% risk for offspring with NTDs and an overall two- to three-fold increased risk for congenital anomalies in the offspring, due to anticonvulsant use, in particular valproic acid and carbamazepine (24).

Other factors including use of oral contraceptives, medical illness (such as hyperthyroidism), and use of anti-seizure drugs may also predispose offspring to NTDs (35). One study in Zewditu Memorial Hospital indicate that there is a significance association present between contraceptive use (OR: 7.14, P: 0.000), antipyretic use (OR: 2, P: 0.029) and NTDs (37).

2.2.8 Maternal Life Style

Maternal life style factors such as alcohol consumption, caffeine consumption, smoking and external exposure to tobacco smoke are associated to NTDs. Meta-analyses review show that maternal factors confirms a small increased risk for spina bifida (34).

Hospital based study conducted in Italy high caffeine intake (≥3cups per day) (OR=7.78), maternal smoking habits (OR=1.91) and alcohol intake (OR= 3.69,) were proved to be a risk factor for spina bifida (24). In addition to this Northern China study indicated that daily passive exposure to cigarette smoke (OR 1.60) was associated to NTDs(36).

In other study of Addis Ababa, Ethiopia show that maternal external exposure to cigarette smoke was strongly associated (OR 2.49, P.020). Significantly strong association was revealed for ≥3 cups/day caffeine intake (OR 8.14, P .000) and coffee was the most common source of caffeine (OR 34.17, P.000) (32).

3. OBJECTIVES

3.1. General Objective

- To assess the prevalence and associated risk factors of NTDs among pregnancy outcome at DBRH, North Shewa, Ethiopia, 2019.

3.2. Specific Objectives

- ✓ To assess the prevalence of NTDs at DBRH, North Shewa, Ethiopia, 2019
- ✓ To identify maternal socio demographic factors associated to NTDs at DBRH, North Shewa, Ethiopia, 2019
- ✓ To determine maternal reproductive and obstetric factors associated to NTDs at DBRH, North Shewa, Ethiopia, 2019
- ✓ To determine maternal environmental exposure associated with NTDs at DBRH, North Shewa, Ethiopia, 2019.
- ✓ To identify maternal medical and drug history associated with NTD at DBRH, North Shewa, Ethiopia, 2019.
- ✓ To assess maternal life style associated with NTDs at DBRH, North Shewa, Ethiopia, 2019

4. METHODS AND MATERIALS

4.1 Study Design

This stud had two study designs:-

- ✓ **For risk factors:-** Hospital based unmatched case control study was be conducted among cases, neonate born with NTDs and controls, neonate born without NTDs in the study period at DBRH, North Shewa, Ethiopia, 2019.
- ✓ **For prevalence:-** retrospective cross sectional, descriptive study of registration of all pregnant outcome from August 30, 2017 to August 30, 2019 at DBRH, North Shewa, Ethiopia, 2019.

4.2 Study Area and Period

Amhara region is one of the 9 regional states of FDRE. The region is divided into 10 zonal administrations and Semien Shewa (North Shewa) is one of these 10 administrative zone. Based on the 2007 Census conducted by the Central Statistical Agency of Ethiopia (CSA), this Zone has a total population of 1,837,490, an increase of 17.72% over the 1994 census, of whom 928,694 are men and 908,796 women (38).

Debrebrhan is zonal city of North Shewa, which is located 130km North East of Addis Ababa. It has nine kebeles with a total population of 94 829 individuals, 50.8% of whom are female. In Debre Berhan, one governmental referral hospital, one private general hospital, four health centers and seventeen private clinics provide healthcare services (39). This study was conducted at DBRH which has a 150-bed facility with a catchment population of 2.8 million people (40).

The services at DBRH include ear, nose and throat, surgery, outpatients department, emergency, tuberculosis and HIV, gynaecology and obstetrics, delivery, abortion care, pediatrics and neonatal intensive care unit (NICU), adult ICU, maternal and child health, physiotherapy, dental, radiology, psychiatry, and internal medicine. Even though DBRH is not affiliated with any of the teaching colleges or universities, it is a teaching hospital for nursing, health officer, medicine, midwifery and pharmacy undergraduate and graduate students in partnership with DBU and other health colleges.

The hospital has a total of 334 healthcare employees: 38 physicians, 180 nurses, 26 midwives, 7 anaesthetists, 31 laboratory technicians, 2 physiotherapists, 4 dentists, 6 radiographers, 4 optometrists and 36 pharmacists (41).

Particularly, Gynecology and Obstetrics ward has 13 bed, 4 gynecologists, 1 emergency surgeon, 9 nurses and 1 abortion room and also a delivery room with 4 waiting beds, 4 delivery coach, ,19 midwives. Neonatal intensive care unit has also 26 bed, 3 pediatrician, 3 general practionars, and 8 nurses.

So this study was carried out at DBRH from March 1 to August 30, 2019.

4.3. Population

4.3.1 Source Population

- For risk factors- all neonate born and medically terminated in DebreBerbrhan Referral Hospital in the study period.
- For prevalence: - all birth and medically terminated fetus registered from August 30, 2017 to August 30, 2019 in the DBRH.

4.3,2 Study Population

- For risk factors: - all neonate and aborted, who full fill the inclusion criteria, born in DBRH for both case and controls in the study periods.
- For prevalence: - all live birth and abortuses who full fill the inclusion criteria from August 30, 2017 to August 30, 2019 in DBRH.

4.3,3 Study Unit

- ✓ For risk factor:- all mother of both case and control groups
- ✓ For prevalence:- medical charts

4.3.4 Inclusion and Exclusion Criteria

4.3.4.1 Inclusion Criteria

For risk factors:-

- ➢ For case: - all neonate born with NTDs in both sex and all medically terminated confirm to had NTDs with gestational age of >12 weeks in DBRH, North Shewa, Ethiopia, 2019.
- ➢ For control: - all neonate born without NTDs and other congenital anomalies in DBRH, North Shewa, Ethiopia, 2019.

For prevalence:-

- ➢ All delivered and medically terminated fetus (gestational age > 12 weeks medical chart) from August 30, 2017 to August 30, 2019 in DBRH, North Shewa, Ethiopia, 2019.

4.3.4.2 Exclusion Criteria

For risk factors: -

- ❖ Respondents who could not hear and talk and critically ill during the data collection, and children's admitted with other care giver/other than the mother will be excluded.
- ❖ Cases with any ambiguity or multiple congenital anomalies were excluded.
- ❖ Gestational age of medically terminated fetus <12 weeks were not involved.
- ❖ A control group with other congenital anomalies other than NTDs would also be excluded.

For prevalence: -

- ❖ Charts with no registration number or incomplete information
- ❖ Cases with other documented congenital anomalies or multiple congenital anomalies

- Gestational age less than 12 weeks

4.4 Sample Size Determination and Sampling Procedure

4.4.1. Sample Size Determination

Sample size of the study was calculated separately for both the prevalence and risk factors to NTDs. For the prevalence, considering the prevalence of NTDs to 50% due to absence of similar research in the study area and using a single proportion formula at 95% CI and 2.5% margin of error, a total of 1537 minimum sample was calculated, but due to rare case 8862 medical delivery charts were conveniently revised in the study periods.

For assessing risk factors for NTDs, using Epi Info, Stat Calc, and considering 95% confidence level (CI), 80% power, control to case ratio of 2, maternal abortion history of similar study which give large sample size compared to other exposure leads to estimate case exposure status 20% and control exposure status 5%. By considering 5% nonresponding rate, we can calculate minimum sample of 50 cases of NTDS and 100 controls.

4.4.2. Sampling Procedures

As it is described in the study area section above, among the North Shewa Hospitals, DBRH (which has high case flow) was purposively selected. To assess risk factors of NTDs, for each confirmed case of NTDs, two mother baby's deliver with normal was selected. Systematically from that same days of delivery list of DBRH. NTDs was defined as case of anencephaly, spinal bifida and meningeocephalocele (42) among infants of any gestational age and medically terminated NTDs. The cases were ascertained by a senior gynecologist who confirmed the diagnosis through gross appearance fetus. To assess prevalence of NTDs, Medical delivery chart from August 30, 2017 to August 30, 2019, which fulfill inclusion criteria was reviewed. It was done through a retrospective review of charts of all mothers who were admitted to DBRH and delivered, live or neonates with NTDs. Additionally medically terminated NTDs affected abortuses were also be included from the abortion**.** Their Medical Record Numbers (MRN) recorded on the admission log books in the maternity ward and Neonatal Intensive Care Unit (ICU) was used to retrieve medical records/charts.

Total prevalence of NTDs was defined as number of NTD cases of live births, still births, technically terminated and abortuses among a total of live birth, still births, and terminated cases (greater than 12 weeks of gestation).

4.5 Data Collection

4.6.1 Data Collection Instruments

All intern doctors, residents, midwives, and clinical nurses informed to report to the principal investigator whenever they encounter suspected cases of NTDs in study period. Data was collected using pretested a semi structured questionnaire. Standard questionnaire was adapted from the WHO birth defect surveillance tool (42).

In order to address the objectives of the study, contents of data extraction format was reviewed by senior Gynecologist. The questioner was developed first in English which was later translated to local language (Amharic).

The sociodemographic and clinical information on the study participants, for example, maternal age, obstetric history, history of medical illnesses, drug intake, exposure to radiation, occupation, level of education, history of congenital anomalies in the family, history of NTDs, residential area, maternal exposure to pollutants such as smoking, exposure to herbicides or pesticide, paternal history, and history of periconceptional folic acid supplementation was collected but, paternal history was obtained from their midwives as secondary information.

Five trained BSc midwives conducted face to face interview with parents of the neonates. Respondent got a brief orientation on the purpose of the study and its significance

Medical delivery chart from August 30, 2017 to August 30, 2019 was revised based on well-structured questionnaire through trained BSc midwives for prevalence of NTDs.

4.5.2 Pretest

Before data collection, pretest was done in 5% (15) of the sample size population (5 cases and 10 controls) at Wollo Referral Hospital, which was not include the study area before the actual data collection period and necessary adjustments, was done on the study tool.

4.6 Data Quality Control

To increase the quality of data properly designed, pretested data extraction tool was used. Training was given to data collectors by principal investigator about the objectives of the study, data collection instruments, data collection procedures and the ethical considerations during data collection.

Each respondent's questionnaire was checked for its completeness and its consistency at the time of data collection by collectors and principal investigators. Data coding, entry, and cleaning was performed by the principal investigator.

4.7 Study Variables

Dependent variable

- ✓ NTDs

Independent variable

- ❖ Sociodemographic
  - Parental education status
  - Maternal age at pregnancy
  - Maternal occupation
- ❖ Maternal reproductive factors
  - Still birth
  - Abortion
  - Parity
  - ANC visit
  - Planning of pregnancy
- ❖ Maternal life style
  - Alcohol
  - Smoking status
- ❖ Maternal exposure to environment
  - Chemical exposure
  - Radiation exposure
- ❖ Preconception factors
  - Maternal weight
  - Folic acid supplementation
- ❖ Genetic factors
  - Previous history of NTDs or congenital anomalies
  - Family history of NTDs history of NTDs or congenital anomalies
- ❖ Maternal medical and drugs factors
  - Maternal drug use
  - Maternal herbal use
  - Maternal medical history
- ❖ Neonatal characteristics
  - Gender
  - Birth index

4.8. Operational Definition

- **Birth defect**: are congenital anomalies that are present at birth of baby, can be structural, functional, metabolic
- **NTDs**: are structural defect of the central nervous system that affects the brain, spine and spinal column during the first month of embryonic development.
- **Spina bifida occulta:** is defect the outer part of some of the vertebrae is not completely closed, Spinal cord does not protrude and Skin may be normal, or some hair growing from it.
- **Spina bifida meningocele:** is meninges herniate between the vertebrae and the nervous system remains undamaged.
- **Spina bifida myelomeningocele:** is a condition where the spinal cord and the tissues covering it protrude out of an opening in the back.
- **Anencephaly** is the absence of a major portion of the brain, skull, and scalp that occurs during embryonic development.
- **Encephalocele:** is a kind of neural tube defect that results in a sac like protrusion of the brain and its surrounding membranes through an opening in the skull.
- **Abortion** is the ending of a pregnancy by removal or expulsion of an embryo or fetus before 28 weeks in Ethiopian heath sit up.
- **Stillbirth** is the delivery, after the 28^{th} week of pregnancy, of a baby who has died according Ethiopian health set up.
- **Alcohol Consumption:** Averagically two or more bottle per day had significant to risk factors NTDs.
- **Caffeine Consumption:** Averagiccally, two or more coffee or tea which had risk of NTDs.
- **Smoking:** According to WHO survilances exposure to smoking or smoking of more than one pack per day.
- **Radiation Exposure:** Any history of Abdominal X-ray or chest x-ray without protection of other body part.
- **ANC follow up:** a woman who had regular antenatal care during pregnancy.
- **Periconceptional period:** a time one month before conception until12 weeks of gestation.
- **Pericoceptional folic acid supplementation:** standard recommendation of folic acid (400µg/day) for all women from the moment they begin trying to conceive until 12 weeks of gestation should take a folic acid supplements.
- **Consanguineous marriage:** a union between two individuals who are related by birth as second cousins or closer (familial marriage).

- **Maternal fever/febrile illness:** a febrile condition that increases degree of body temperature > 38^0C either internally caused by illness (febrile illness) or external exposures (hot tab use, heated beds….).
- **Close family member:** are individuals who are related by blood and it includes grandparents, parents, siblings and children.
- **Body Mass Index (BMI):** BMI is a measurement of persons weight(kg) with respect to his/her height (Meter square) based on the WHO BMI score is classified as;
    - **<18.5- underweight**
    - **18.5-24.9 - normal weight**
    - **25-29.0 - over weight**
    - **>30.0 - obese**
- **Parity:** the number of times the woman give birth
- **Multiparous:** woman who has given one more birth
- **Living near to waste disposal area**: residents located less than 100 meter from waste disposal area or polluting industries.
- **Cases:** for this study NTDs includes spinal bifidia, mylomeningocele and Encephalecle occurring either in isolation or in combination with other congenital malformations.

4.9 Data Processing, Analysis, Presentation

The data was checked for completeness and consistencies, cleaned, coded and entered using Epi data version 4.2 and was exported to statistical package for social sciences (SPSS) software version 20 for analysis.

Odds ratio (OR) with 95% CI will be used to assess the relationship between factors associated with the occurrence of outcome variable.

Descriptive statistics was used to describe the study population in relation to relevant variable. The prevalence of NTDs was calculated for both birth prevalence and total prevalence per 1000 based on birth outcomes [live births, abortion, stillbirths, and terminated pregnancy].

Logistic regression model was used mainly multinomial and bivariate logistic regression was done to determine association between each independent variable with outcome variable.

Variables having $P<0.05$ was consider as statistically significant.

4.10 Ethical Consideration

After approval of the document by Addis Ababa University (AAU), Collage of Health Science, Ethical clearance was obtained from AAU Collage of Health Science Research and Community Service directorate ethical committee. Then supportive letter was taken from North Shewa Zone health bureau and DBRH for permission to conduct the research. After getting permission to conduct the study, each study subject's selected parents was asked for their willingness by explaining objectives of the study and its significance.

During data collection, the respondents were participated voluntarily and given freedom to withdraw any time they want. Participant's confidentiality was strictly held by research team. Study participants was not face any physical and emotional harm resulting from participating in the study.

5. RESULTS

7.1 Prevalence of Neural Tube Defects

During the study period, there were a total of 8,862 card (out of them, 7920 were delivery, 722 were abortion and 220 were medically terminated) of pregnancy after the 12th week of gestation were assessed. From those 97 pregnancy (out of case of NTDs,60 cases from delivery,26 cases from abortion and 11 cases from medically terminated) were with NTDs.

Among live births, aborted, stillbirths and medically terminated cases, the birth prevalence of NTDs was 10.9 per 1000 pregnancy. Including medically terminated cases, 50 cases per 8862 were encephala, 31 cases per 8862 were spina bifidia, 10 cases were encephalocelly and 6 cases per 8862 were both spina bifid and anecephalocelly.

Prevalence of Anencephaly was 5.6 per 1000 pregnancies, spina bifida was 3.5 per 1000 pregnancies, encephalocele was 0.9 per 1000 pregnancies and both spina bifida and anecephally were 0.7 per 1000 pregnancies.

In this study, cases of anencephaly were the most common types of the NTDs. Among types of NTDs most of anencephaly cases were aborted and become still birth and spina bifid and encephallocelly cases were live birth. [Figure 1].

From a total of 97 cases, maternal occupation was house wife 21.6% (21/97) and farmer 28.9% (28/97). From 97 NTDs case, 87.6 % (85/97) and 95.6% (93/97) were not taken folic acid prior to conception and were not taken folic acid at any time, respectively.

Multiparous and prim parous were the commonest gravidities each accounting for 71 % (69/97) and 16.5% (16/97), respectively. Most of NTDs, 33% (32/97), 23.7% (23/97) and 16.5% (16/97) gestational age were 37-40 weeks, 32-36 weeks and <28 weeks, respectively.

As shown in table 1; when we compare types of NTDs with gender, 74% (37/50) anencephaly and 67.7% (21/31) spina bifida were common in male and female, respectively.

Majority of the mothers, 47.4% (46/97) had no ANC follow up and 28.9% (28/97) were start ANC follow up after 3rd trimester. [Table 2].

From 97 NTDs, 31.9 % (31/97) spina bifida and 22.7% (22/97) anencephaly occurs among multiparous mothers and 16.5 % (16/97) and 12.4 % (12/97) anencephaly occurs from prim parous and nulliparous, respectively. [Table 3].

Anencephaly of 20.6 % (20/97) and 12.4% (12/97) mainly presented with in gestation age of 32-36 weeks and <28 weeks, respectively. [Table 4].

Among 97 NTDs, 20.6% (20/97) and 21.6% anencephaly were <20years and 31-35 years maternal age, respectively. Maternal age of >35 years of mothers had 28.9% (28/97) spina bifida, 10.3% enecephalocelly and 6.2% (6/97) both anencephaly and spina bifida. [Table 5].

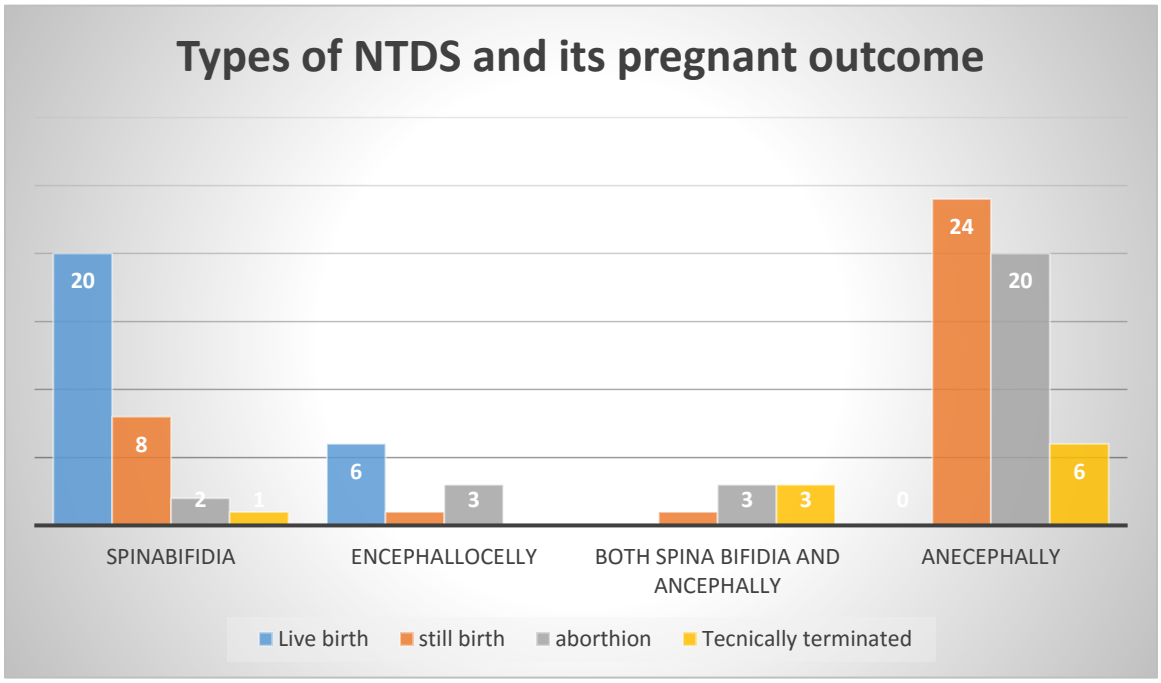

Figure 1:- Types of NTDs and its pregnant outcome in Debrebrhan Referral Hospital, from August 30, 2017 to August 30, 2019 (n = 97).

Table 1: Types of NTDs and Genders at Debrebrhan Referral Hospital, North Shewa, Ethiopia, 2019. [N=97]

		Types of NTD				Total
		Anencephaly	**Spina bifda**	**Encephalocele**	**Both anencephally and spinal bifidia**	
Gender	Male	**37**	10	3	1	51
	Female	13	**21**	**7**	**5**	46
Total		50	31	10	6	97

Table 2: Socio demographic characteristics and folic acid history of pregnant women whom their child were NTDs at Debrebrhan Referral Hospital, North Shewa, Ethiopia, 2019. [N=97]

	Variables	Frequency	Percent %	Cumulative percent
Religion	Orthodox	70	**72.2**	72.2
	Muslim	14	14.4	86.6
	Protestant	7	7.2	93.8
	Others	6	6.2	100.0
	Total	97	100.0	
Ethnicity	Amhara	51	**52.6**	52.6
	Oromo	25	25.8	78.4
	Tigray	12	12.4	90.7
	Others	9	9.3	100.0
	Total	97	100.0	
Occupational status	House wife	28	**28.9**	28.9
	Labor	11	11.3	40.2
	Merchant	6	6.2	46.4
	Professional	7	7.2	53.6
	Farmer	21	**21.6**	75.3
	Factory	17	17.5	928
	Others	7	7.2	100
Took folic acid supplement prior to conception	Yes	4	4.1	4.1
	No	93	**95.6**	100.0
Took folic acid supplement at any time	Yes	12	12.4	12.4
	No	85	87.6	100.0
Parity	Nulliparous	12	12.4	12.4
	Prim parous	16	**16.5**	28.9
	Multiparous	69	**71.1**	100.0
ANC initiation time	1st trimester	10	10.3	10.3
	2nd trimester	13	13.4	23.7
	3rd trimester	28	**28.9**	52.6
	No ANC	46	**47.4**	100.0
	<20	20	20.6	20.6
	20-25	9	9.3	29.9

	31-35	23	**23.7**	53.6
	>35	44	**45.4**	100.0
Gestational age of NTDs	<28 weeks	16	16.5	16.5
	28-31 weeks	14	14.4	30.9
	32-36 weeks	23	**23.7**	54.6
	37-40 weeks	32	**33.0**	87.6
	>40 weeks	12	12.4	100.0

Table 3: Types of NTDs and Parity at Debrebrhan Referral Hospital, North Shewa, Ethiopia, 2019. [N=97]

		Parity			Total
		Nullipa rous	Prim parous	Multipa rous	
Types of NTD	Anencephaly	12	16	**22**	50
	Spina bifda	0	0	**31**	31
	Encephalocele	0	0	10	10
	Both anencephally and spinal bifidia	0	0	6	6
Total		12	16	**69**	97

Table 4: Types of NTDs and Gestational age at Debrerbrhan Referral Hospital, North Shewa, Ethiopia, 2019. [N=97]

		Gestational age of NTD					**Total**
		<28 weeks	**28-31 weeks**	**32-36 weeks**	**37-40 weeks**	**>40 weeks**	
Types of NTD	**Anencephaly**	**16**	14	**20**	0	0	50
	Spina bifda	0	0	3	**28**	0	31
	Encephalocele	0	0	0	4	6	10
	Both anencephally and spinal bifidia	0	0	0	0	6	6
Total		16	14	23	32	12	97

Table 5: Types of NTDs and Maternal age at Debrerbrhan Referral Hospital, North Shewa, Ethiopia, 2019. [N=97]

		Types of NTD				Total
		Anencephaly	Spina bifda	Encephalocele	Both anencephally and spinal bifidia	
Maternal Age	<20	**20**	0	0	0	20
	20-25	9	0	0	0	9
	31-35	**21**	3	0	0	24
	>35	0	**28**	**10**	**6**	44
Total		50	31	10	6	97

7.2. Logistic Regression Results of Factors and NTDs

7.2.1 Socio Demographic Factors of Cases & Controls Associated With NTDs

A total of 150 (50 cases and 100 controls) were included with 100 % response rate. As it is shown in the table 6; the socio demographic factors, maternal age, < 20 (OR =3.09, CI= 0.95 – 10.08, P=0.046), maternal age 31-35 (OR= 0.53, CI=0.16 – 1.7, P =0.028) were strongly associated to risk of NTDs. No difference between case and control in maternal age of 20-25 and 26-30. Compare to maternal education of case to control, no education (OR=2.98, CI=0.95-9.35, P=0.014) were significant association with NTDs. However, in other education level there were not association between control and case association with NTDs. Paternal educational status no education (OR=0.65, CI=0.08-5.14, P=0.994), maternal occupational status house wife (OR=0.96, CI=0.15-6.19, P=0.657), gender differences female (OR=0.58, CI=0.22-1.57, P=0.287), and birth order first order (OR=0.36, CI=0.21-0.60, P=0.220) were insignificantly association with NTDs.

Table -6 Socio demographic factors of cases & controls and NTDs at DRH, North Shewa, Ethiopia, 2019

Variables	Case. N (%)	Control. N (%)	OR	CI (95%)	P- value
1.Maternal age					
<20	17 (34)	11 (11)	3.09	0.95 - 10.08	**0.046**
20-25	6 (12)	27 (27)	0.44	0.125 - 1.58	0.210
26-30	11 (22)	14 (14)	1.57	0.47 - 5.23	0.461
31-35	9 (18)	34 (34)	0.53	0.16 - 1.70	**0.028**
>35	7 (14)	14 (14)	Ref		
2. Maternal educational status					
No education	16(32)	17 (17)	2.98	0.95-9.35	**0.014**
Primary school	15 (30)	20 (20)	2.37	0.76-7.39	0.136
Secondary school	11 (22)	24 (24)	1.45	0.45-4.64	0.530
Occupational training	2 (4)	20 (20)	0.32	0.06-1.77	0.190
Diploma/degree /other	6 (12)	19 (19)	Ref		
3. Paternal educational status					
No education	6 (12)	7 (7)	0.65	0.08-5.14	0.994
Primary school	11 (22)	13 (13)	2.01	0.51- 7.98	0.680
Secondary school	9 (18)	33 (33)	0.36	0.09- 1.45	0.320
Occupational training	8 (16)	24 (24)	0.28	0.06 – 131	0.152
Diploma/degree /other	16 (32)	23 (23)	Ref		
4**. Maternal occupation**					
House wife	21 (42)	33 (33)	0.96	0.15 - 6.19	0.657
Farmer	7 (14)	20 (20)	0.53	0.07 - 3.82	0.961
Factory workers	14 (28)	7 (7)	3.00	0. 40 - 22.30	0.525
Office workers	6 (12)	36 (36)	0.25	0.03 - 1.82	0.283
Others	2 (4)	3 (3)	Ref		
5. Gender					
Female	27 (54)	52 (52)	0.58	0.22 - 1.57	0.287
Male	23 (46)	48 (48)	Ref		
6. Birth order					
First	12 (24)	41 (41)	0.36	0.21 - 0.60	0.220
Second	11 (22)	35 (35)	0.38	0.32 - 0.67	0.440
Third and above	27 (54)	24 (24)	Ref		

Ref:-Reference point which given by Logistic regression software

7.2.2 Maternal Reproductive and Genetic Factors Associated With NTDs

As it is shown in the table 7; Family history of NTDs (OR=4.21, CI=1.11-15.88, P=0.034) and history of multiparous (OR=3.96, CI=1.60-9.28, P=0.002) had significant association with NTDs. Regarding abortion (OR=1.78, CI=0.68-4.60, P=0.024) and still birth (OR=3.92, CI=1.33-11.51, P=0.009) were risk factors for NTDs. Mother who had unplanned pregnancy (OR=6.29, CI=2.95-13.44, P=0.000) and no ANC follow-up were strong significance association with occurrence of NTDs. There was no difference between the cases and controls in terms of husband and wife relationship (OR=1, CI=0.09-11.30, P=1.000), previous history of pregnancy with NTDs (OR=6.58, CI=0.91-47.68, P=0.062) and neonatal loss (OR=2.81, CI=1.33-11.51, P=0.170)

Table - 7; Maternal Reproductive, Obstetric and Genetic history association with NTDs, at DRH, North Shewa, Ethiopia, 2019

Variables	*Case. N (%)*	*Control. N (%)*	*OR*	*CI (95%)*	*P- value*
1. Maternal Genetic Factors					
1.1 Family Hx of NTDs					
Yes	17 (34)	12 (12)	4.21	1.11 - 15.88	**0.034**
No	33 (66)	88 (88)	Ref		
1.2 Consanguinity					
Yes	1(2)	2 (2)	1.00	0.09 -11.30	1.000
No	49(98)	98 (98)	Ref		
1.3 Hx of NTDs					
Yes	10 (20)	3 (3)	6.58	0.91 - 47.68	0.062
No	40 (80)	97 (97)	Ref		
2. Maternal reproductive and obstetric history					
2.1 Parity					
Nulliparous	12 (24)	42 (42)	Ref		
Multiparous	26 (52)	22 (22)	3.96	1.68 - 9.28	**0.002**
2.2 History of abortion					
Yes	14 (28)	10 (10)	1.78	0.68 – 4.6	**0.024**
No	36 (72)	90 (90)	Ref		
2.3 History of still birth					
Yes	8 (16)	5 (5)	3.92	1.33 – 11.51	**0.009**
No	42 (84)	95 (95)	Ref		
2.4 History of neonatal loss					
Yes	4 (8)	3 (3)	2.81	1.33 – 11.51	0.170
No	46 (92)	97 (97)			
2.5 Planned pregnancy					
Yes	21 (42)	82 (82)	Ref		

No	29 (58)	18 (18)	6.29	2.95-13.44	**0.000**
2.6 ANC follow up					
Yes	16 (32)	78 (78)	Ref		
No	34 (68)	22 (22)	7.53	3.53-16.1	**0.000**

7.2.3 Maternal Medical and Drug History Factors Associated With NTDs

From table 8; A significance risk of NTDs was high among mothers who had history of medical illness (OR=3.8, CI=1.75-8.25, P=0.001), such as history of HTN, hyper/hypothyroidism, UTIs, Epilepsy, and DM and drug use associated to each medical illness (OR=2.72, CI=1.94-10.67, P=0.000). All mothers (99% cases and 98% controls) did not use sauna/hot tab. Regarding cases of mother 74% and controls of mothers 17% were develop fever (OR=13.89,CI=6.12-31.54, P=0.000) due to different cause. Among them 62% case of mothers and 43% controls of mothers who had fever during pregnancy, used antipyretics drugs (OR=11.96, CI=5.21-27.45, P=0.002) for relief of fever. There was a significant difference (OR=4.65, CI=1.32-16.43, P=0.001) between cases (90%) and controls (71%) for never use of folic acid in preconception period. A significantly high risk association was observed for maternal periconceptional oral contraceptive use (OR=3.4, CI=1.67-7.05, P=0.001).

Table 8 Maternal Medical and Drug History factors & NTDs, at DRH, North Shewa, Ethiopia, 2019

Variables	Case. N (%)	Control. N (%)	OR	CI (95%)	P- value
1.Disease history					
Yes	21 (42)	16 (16)	3.8	1.75-8.25	**0.001**
No	29 (58)	84 (84)	Ref		
1**.1 Medication taken**					
Yes	18 (85.7)	11 (68.75)	2.72	1.94-10.67	**0.000**
No	3 (14.3)	5 (31.25)	Ref		
2**.History of fever**					
Yes	37 (74)	17 (17)	13.89	6.12-31.54	**0.000**
No	13 (26)	83 (83)	Ref		
2.1. Anti-pyretic use					
Yes	31 (62)	43 (28.7)	11.96	5.21-27.45	**0.002**
No	19 (38)	88 (71.3)	Ref		
3. Oral contraceptive use					
Yes	26 (52)	24 (24)	3.4	1.67-7.05	**0.001**
No	24 (48)	76 (76)	Ref		

4. Folic acid consumption					
Never	45 (90)	71 (71)	4.65	1.32 – 16.43	**0.001**
Periconceptional intake	2 (4)	7 (7)	2.09	0.29 – 15.19	0.464
After 3 months of conception	**3 (6)**	**22 (22)**	**Ref**		

7.2.4 Maternal Environmental Factors Associated With NTDs

As it is shown in table 9; Factors which were maternal exposure to radiation (OR=1.35, CI=0.22-8.33, P=0.748), living near to waste disposal (OR=1.79, CI=0.76-1.79, P=0.176), tradition medication use (OR=2.04, CI=0.90-4.62, P=0.083) and maternal underweight or overweight were not NTDs risks. There was strong significant association found for maternal exposure to smoking (OR=3.35, CI=1.62-6.91, P=0.001) and pesticides (OR=7.81, CI=3.61-16.76, P=0.000) with NTDs. Regarding to maternal consumption of alcohol mainly local alcohol "Arki" (OR=9.33, CI=4.28-20.32, P=0.000) and caffeine source mainly coffee and tea (OR=42.67, CI=9.8-18.98, P=0.000) were strong significant association with the occurrence of NTDs. However, maternal cigarette smoking (OR=1.89, CI=0.85-4.26, P=0.117) was not associated with NTDS.

Table 9: Environmental factors and NTDs, at DRH, North Shewa, Ethiopia, 2019

Variables	Case .N (%	Control. N (%)	OR	CI (95%)	P-value
1. Sauna/hot bath use					
Yes	1 (2)	1 (1)	2.02	0.37-6.15	0.615
No	49 (98)	99 (99)	Ref		
2. Pesticides exposure					
Yes	35 (70)	23 (23)	7.81	3.61-16.76	**0.000**
No	15 (30)	77 (77)	Ref		
3. Radiation exposure					
Yes	2 (4)	3 (3)	1.35	0.22-8.33	0.748
No	48 (96)	97 (97)	Ref		
4. Living near to waste disposal					
Yes	12 (24)	15 (15)	1.79	0.76-1.79	0.176
No	38 (76)	85 (85)	Ref		
5. Herbal medication use					
Yes	14 (28)	16 (16)	2.04	0.90-4.62	0.083
No	36 (72)	84 (84)	Ref		
6. Prepregnancy weight					
Under weight	7 (14)	52 (52)	1.10	0.16 – 6.97	0.850
Normal weight	17 (34)	42 (42)	0.932	0.27 – 5.25	0.954
Over weight	24 (48)	6 (6)	1.21	0.28 - 6.18	0.787
Obese	2 (4)	0 (0)	Ref		
7. Caffeine taken					
Yes	48 (96)	36 (36)	42.67	9.80 -18.98	**0.000**
No	2 (4)	64 (64)			
8. Caffeine source					
Coffee	32 (64)	26 (26)	39.38	8.79 - 176.42	**0.000**
Tea	16 (25)	10 (10)	51.20	10.19 - 257.17	**0.000**
NA	2 (4)	64 (64)	Ref		
9. Alcohol consumption					
Yes	35 (70)	20 (20)	9.33	4.28-20.32	**0.000**
No	15 (30)	80 (80)	Ref		
10. Tobacco smoking					
Yes	14 (28)	17 (17)	1.89	0.85-4.26	0.117
No	36 (72)	83 (83)	Ref		
11. Exposure to Smoking					
Yes	25 (50)	23 (23)	3.35	1.62-6.91	**0.001**
No	25 (50)	77 (77)	Ref		

8. DISCUSSION

This study was done through two study design which were unmatched hospital based case control study consisted of 50 case and 100 control and retrospective cross sectional study consists of reviewing 8862 pregnant mothers card from those 97 pregnancies were NTDs. Because birth defects are a major cause of mortalities before five years of age and Neural tube defects (NTDs) are one of the most common major birth defects next to congenital heart diseases (43), adequate surveillance data are needed to develop effective prevention strategies.

In this study, the total prevalence for all types of NTDs was found to be 10.9 per 1000 pregnancy, which were compared to six times more prevalent than study done in six World Health Organization (WHO) 1.67/1000 (15), and three times and one point eight (1.8) times more prevalence in studies done in Sudan 3.48/1000 (19), and in three teaching hospital of Addis Ababa 6.1/1000 (20), respectively, but this prevalence was less than the prevalence of NTDs in Tigray region 13.8/1000 (7). The specific finding of anencephaly (5.6/1000) is higher than prevalence of NTDs reported in in Africa and Ethiopia, and much more higher than reports from six World Health Organization stud site in Africa 0.25/1000 (14), Malawi 3.1/1000 (16), and in three teaching hospital Addis Ababa 4.2/1000 (20). In other types of NTDs, this study had higher prevalence of spinal bifida (3.5/1000) compare to six World Health Organization site in Africa 1.13/1000 (14), Malawi 0.47/1000 (16) and Cape Town 1.74/1000 (17), except in Tigray region 6.4/1000 (7). NTDs were observed to occur almost equal in male-female proportional, 52.6% in male and 47.4% in female, but 74% anencephaly common in male and 67.7% spinabifida common in female which is not comparable to reports from a case-control study based on the Oxford Record Linkage about 70 percent of the children with anencephaly and 60 percent of the children with spina bifida were females (44).

Variable such as periconceptional oral contraceptive use, no folic acid consumption, maternal age <20 and 31-35, no ANC follow up, maternal no education, history of abortion and still birth, exposure to smoking, alcohol and caffeine consumption and family history of NTDs show significant association with NTDs.

This study revealed that maternal age, < 20 (OR =3.09, CI= 0.95 – 10.08, P=0.046), maternal age 31-35 (OR= 0.53, CI=0.16 – 1.7,P =0.028) were strongly associated to risk of NTDs, but it had age differences study done in Genoa show that maternal age < 25 years (OR=3.36) and >35

years; (OR=5.21) Italy (19) and Tigray region >35 years (OR=2.5,P=0.0004) (7). Possible correlation can be established between Pregnancy at lower age groups<20 and NTDs risks, this might be due to low socioeconomic status, low educational level, lack of knowledge regarding potential risk factors and preventive measures.

Hospital based case–control study conducted at Genoa, Italy, shows that maternal low educational level (OR=4.87) (28) had association to NTDs similar to this study which was no education (OR=2.98, CI=0.95-9.35, P=0.014).

The high prevalence of this study was might be results from nutritional factors, family history of NTDS (OR=4.21, CI=1.11-15.88, P=0.034), lack of routine folic acid supplementation (OR=4.65, CI=1.32-16.43, P=0.001), and absence of folic acid fortification programs. So, in this study there was no periconceptional folic acid supplementation in 90% of case of mothers and 71% of control mothers and it had consistence with study of Italy (OR=27 CI, 9.31–78) (28), 86% in Algeria (25), 92.2% and 85.3% of case not receive folic acid study was done three teaching hospital of Addis Ababa and Tigray region, respectively.

According to this study the maternal reproductive history of abortion 28% (OR=1.78, P=0.024), still birth 84% (OR=3.92, P=0.009) and multiparous 52% (OR=3.96, P=0.002) were associated with mothers whose birth outcome are NTD, this result is inconsistent with study conducted in Catania, Italy study multiparous (84.8%), spontaneous abortion (21%),and still birth (16%) (24), and somewhat it had similarity in study done in Tigray multiparous (P=0.000) and three hospitals of Addis Ababa multiparous (P=0.049) (7, 20). Regarding this study, 68% of pregnancy did not have ANC follow up (P=0.000) and 52% of pregnancy were unplanned (P=0.000) of case of mothers with NTDs. The differences were come due to lack of knowledge, inadequacy of infrastructure, poor folic acid supplementation and low ANC follow up.

This study show that maternal hyperthermia was associated to NTDs risk, 74% cases mothers develop fever during early pregnancy (OR=13.89, CI=6.12-31.54, P=0.000) due to different causes. This finding is consistence with other case control studies conducted in California (OR=2.02) (35), China (OR=3.36) (36) and Zewditu Referral Hospital, Ethiopia (OR 65.5, CI 4.48 – 957.9 P .002) (32). Meta-analyses review show that maternal fever in early pregnancy as a risk factor for NTD-affected pregnancies and also heat that adversely affects development, can leads to increased cell death, decreased proliferation, disruption of gene expression, and damage

to the embryonic vasculature, results induction of apoptosis, inhibition of proliferation/ slow differentiation (33, 34). Related maternal hyperthermia, 62% case of mothers and 43% controls of mothers who had fever during pregnancy, used antipyretics drugs, but 99% of case mothers and 98% control mothers not used sauna/hot tabs.

This study showed that coffee consumption was significantly associated with NTDs risk (OR=42.67, CI =9.8-18.98, P =0.000) which is supported by study conducted in Italy (24) and had similarity in study done in Addis Ababa, Ethiopia (OR 2.49, P.020) (32). The major caffeine source is coffee (OR=42.67, CI=9.8-18.98, P=0.000). Several study show that coffee consumption during pregnancy can cause abortion, decrease folate absorption and metabolism and disturbed maternal food intakes.

Regarding to maternal consumption of alcohol (mainly local alcohol "Arki") (OR=9.33, CI=4.28-20.32, P=0.000) associate with NTDs and it had consistencies in study done in other area such as Italy (OR= 3.69) (24).

Concerning to maternal exposure to smoking had risk to NTDs ((OR=1.89, CI=0.85-4.26, P=0.117) which had similar consistency finding that was done in China (OR=1.6) (36) and Addis Ababa, Ethiopia (OR 2.49, P.020) (32).

9. CONCLUSION

In conclusion, the prevalence of neural tube defects in this study is among the highest globally reported (Africa, Europe, America, and many Asian countries as well as reported from Addis Ababa Hospitals). Results indicated that the most prevalent NTD being anencephaly and spina bifida.

This study showed that family history of NTDs, coffee and alcohol consumption, maternal no education, maternal hyperthermia and disease, maternal antipyretic use, maternal age <20 and 31-35, exposure to pesticides and smoking were associated with the increasing risk of NTD.

The practice of periconceptional folic acid supplementation in our setup is negligible, and most of case of mother had no ANC follow up.

10. STRENGTH OF THE STUDY

This study had several strengths;

- It involve all pregnancy outcome >12 gestational age (Live birth, Still birth, Aborted fetus and medical terminated)
- It tries to examine several potential risk factors
- The study is done in two study design, which are case control to associate risk factors and retrospective cross sectional to for prevalence of NTDs, separately based on the its advantage of the study design.
- Analyses were based on all pregnancies outcomes (Live birth, still birth, Aborted fetus and medical terminated).

11. LIMITATION OF THE STUDY

- Since it is a hospital based study it makes it difficult to represent for the general population.
- Because of retrospective study, participants recall bias is participant recall bias.
- It is a hospital-based study and could have missed many deliveries which occurred outside hospitals.

12. RECOMMENDATION

Considering the findings of this study; the following recommendations are set for:-

- **Ministry of health, Regional health bureau and North Shewa health Offices**
  - ✓ NTDs prevention should be incorporated as part of ANC follow up in order to achieve periconceptional maternal counseling, screening for potential medical illness and periconceptional folic acid supplementation to early prevent NTDs.
  - ✓ Implementing preventive strategy of NTDs should incorporate in HEPs.
  - ✓ Fortification of food with folic acid should be done at national level.
- **Health care workers**
  - ✓ Health education about NTDs risk factors and preventive measures should be given for all family and women's of child bearing age (15-49).
  - ✓ Periconception counseling, screening and folic acid supplementation should be done before mothers become pregnant.
- **For Researchers**

✓ Large scale prospective studies are needed to have reliable estimates on burden of NTDs and associated factors

✓ Other study can be done in cost benefit analysis of screening and preconception folic acid administration for Ethiopian set-up.

✓ Further research should be conducted on factors such as genetic studies (like genotype screening)

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ANNEXES

ANNEX- 1: Information Sheet (English Version)

My name is Zerihun Kindie, I am MSc student at Addis Ababa University, Collage of Health Sciences, Department of Anatomy, and I am conducting a research on burdens and associated risk factors of neural tube defect in Debrebrhan Referral Hospital, North Shewa, Ethiopia, 2019.

The objective of the study is to collect information regarding burdens and associated risk factors of neural tube defect in the study. The study will not give any direct benefits to you. Howe ever the information which will be obtained from you will help the researcher to recommend the concerned bodies to design appropriate interventions to address the problems. The study will not affect and your participation is depend on your permission and also you can quite from giving answers to the questions any time you want. Be Sure that your information will not be accessed by anyone other than the study team and will be confidential, and if you are willing the interview may take 15-25 minute and we can proceed.

Consent Form

I give my consent to participate in this study. I have been given the necessary information about the research in a language I understand. I have also understood that I can withdraw my consent any time without penalty or loss of benefits.

1. If yes, proceed to the next page
2. If no, thank you, and skip to the next participant

Name of data collector _____ Signature_____

Name of supervisor _____ Signature_____

Date of interview _____

ANNEX- 2: Questionnaire (English Version)

The questionnaire has seven components: Part one is about Maternal socio-demographic factors and child characteristics, Part II: maternal genetically factors, Part III: maternal reproductive and obstetric factors, Part IV: maternal medical and drug history, Part V: maternal chemical exposure, Part VI: maternal nutritional and folic acid consumption, finally, Part VII: maternal lifestyle

Part I: parental socio demographic factors

1.0 Maternal age at index pregnancy_____

1.2 Parental educational status

1. No education
2. Primary school
3. Secondary school
4. Occupational training
5. Diploma/degree /other

Mother_____ Paternal____

1.3 Maternal occupation_____

1. Housewife
2. Farmer
3. Factory worker
4. Office worker
5. Other_____

1.4 Child characteristics

1.4.0 Gender Female ◯ Male ◯

1.4.1 Birth order First ◯ Second ◯ Third and above ◯

1.4.2 Type of diagnosis (only for case)_____

Part II: Maternal Genetical Factors

2.0 Is there any close family member with history of NTDs /other congenital anomalies?

Yes ◯ No ◯

2.1 Do you have history of NTDs affected pregnancy? Yes ◯ No ◯

2.2 Do you have genetically relationship with your husband? Yes ◯ No ◯

PART III: Maternal reproductive and obstetric history

3.0 Parity_____

3.1 Do you have any history of Abortion ◯ still birth ◯ early neonatal loss ◯

3.2 Was the pregnancy for the index child planned? Yes ◯ No ◯

3.3 Had ANC follow up? Yes ◯ No ◯

Part IV: Maternal Medical And Drug History

4.0. Have you ever been told by a doctor that you had any of these diseases

Hypertension, Hyper/hypothyroidism, bladder /urinary tract infection, tumor, epilepsy, DM

Yes ◯ No ◯

4.1 If the answer is yes did you take any medication (for the medical condition)?

Yes ◯ No ◯

4.2 If "yes" list the medication name_____

4.3 Any Oral contraceptive use? Yes ◯ No ◯

4.4 Have you experienced any fever/ febrile illness Yes ◯ No ◯

4.5 If "yes" did you take any antipyretic (fever reducing medication)? Yes ◯ No ◯

Part V: Maternal Chemical Exposure

5.0 Is there waste disposal site or industry near to your residence? Yes ◯ No ◯

5.1 If "yes" state the distance (meter) _____

5.2 Have you used any pesticides (insecticides, herbicides or fungicides) at home /work place?

Yes ◯ NO ◯

5.3 Have you ever used sauna/hot tub? Yes ◯ No ◯

5.4 If yes for the above question for how long in average(in minutes)_____

5.5 Have you experienced any diagnostic/therapeutic radiations?

Yes ◯ No ◯

Part VI: Maternal nutritional and folic acid consumption

6.1 What was your prepregnancy weight (Kg)_____, height (meter)_____

6.2 Did you take Folic acid supplements?

Never ◯ Periconceptional intake ◯ After 3 months of conception ◯

Part VII: maternal lifestyle

7.1 Did you take any caffeine Yes ◯ No ◯

7.2 If your answer is yes which caffeine source Coffee ◯ Tea ◯

7.3 How many cups per day (in average)? _____

7.4 Other traditional herbal use (specify)? _____

7.5 Do you drink alcohol? Yes ◯ No ◯

7.6 If your answer yes (amount in average)

>500ml/day ◯ <500ml/day ◯ occasional (<500ml/week) ◯

7.7 Do you smoke? Yes ◯ No ◯

7.8 Did you had any home/occupational exposure to tobacco smoke?

Yes ◯ No ◯

ANNEX 3: Information Sheet (Amharic Version)

የጥናቱ ተሳታፊ አጠቃላይ መረጃ

ሰላም

ስሜ ዘሪሁን ክንዴ ይባላል ። በአዲስ አበባ ዩኒቨርሲቲ ፣ የሰዉ አካል አወቃቀር ት/ምህርት ክፍል የድህረ ምረቃ ተማሪ ስሆን በደብረብርሃን ሪፈራል ሆስፒታል ዉስጥ በህብረ-ሰረስር ትቦ አለመግጠም ተዛማጅ ምክንያቶች እና ስርጭቱ ዙሪያ የጥናትና ምርምር ስራ በመስራት ላይ እገኛለሁ ።

የጥናቱ አላማ ፡- በህብረ- ሰረስር ትቦ አለመግጠም ተዛማጅ ምክንያቶችና ስርጭቱን መለየትና ችግሩን ለሚመለከተዉ አካል ማሳወቅ ነዉ ።

ጥናቱ ፡- ለተሳታፊዎች ቀጥተኛ የሆነ ጥቅም የለዉም ። ነገር ግን ከእርስዎ የሚገኝ መረጃ ችግሩን ለመቅረፍ ወሳኝነት አለዉ ።

ጥናቱ በተሳታፊዎች ላይ ምንም አይነት አደጋ የማያስከትል ሲሆን የሚከናወነዉም በተሳታፊዉ ፍቃደኝነት ላይ ተመስርቶ ነዉ እንድሁም የጥናቱ ተሳታፊ በፈለገዉ ጊዜ ምላሽ አለመስጠት ይችላል የጥናቱ ተሳታፊ የሚሰጡት መረጃ ከአጥኝዉ ቡድን ዉጭ ለሌላ አካል ተላልፎ አይሰጥም ። ስለዚህ ፍቃደኛ ከሆኑ ቃለመጠይቁ ከ15-25 ደቂቃዎች ዉስጥ ማከናወን ይችላል ። ስለሆነም መቀጠል እችላለን ።

የመረጃ ሰነድ

ስለጥናቱ በቂ የሆነ መረጃ አግኝቻለሁ በምችለዉ ቋንቋ በጥናቱ በመሳተፍ ተስማምቻለሁ። በጥናቱም ላይ ምንም አይነት ጉዳት እንዲሁም ቀጥተኛ ጥቅም እንደማላገኝ ተረድቻለሁ።

አዎ ከሆነ ወደሚቀጥለዉ ጥያቄ ሂድ / ሂጅ

አይደለም ከሆነ ተሳታፊዉን በማመስገን ወደሚቀጥለዉ ተሳታፊ ሂድ / ሂጅ

መረጃ ሰብሳቢዉ ስም ---------- ፊርማ ----------

መረጃዉ የተሰጠበት ቀን ----------

ANNEX 4: Questionnaire (amharic Version)

መጠይቅ

ክፍል 1 የእናት ማህበራዊ እና ኢኮኖሚያዊ እና የህፃኑ ባህርያት

1.1 የወላጅ የትምህርት ደረጃ

1. አልተማሩም
2. የመጀመሪያ ደረጃ ወይም ከ1-8
3. ሁለተ ኛ ደረጃ ከ9-12
4. የሙያ ስልጠና
5. ዲፕሎማ ወይም ዲግሪ ወይም ሌላ ----------
6. እናት ---------- አባት ----------

1.2 የናት የስራ ድርሻ----------

1. የቤት እመቤት
2. የግብርና ስራ
3. የፋብሪካ ሰራተኛ
4. የቢሮ ሰራተኛ
5. ሌላ ----------

1.4 ህፃኑን የሚመለከቱ መጠይቆች

1.4.0 ፆታ ሴት ወንድ

1.4.1 ስንተኛ ልጅ ነዉ ወይም ናት

1.4.2 የበሽታዉ አይነት (ህብረ ስራስር ትቦ አለመግጠም ላለባቸዉ ብቻ)

ክፍል 2 የዘር ምክንያቶች

2.1 ህፃኑ ወይም ህፃኗ ሲረገዝ የእናትየዋ እድሜ ስንት ነዉ?

2.2 የህብር ስራስር ትቦ አለመግጠም ችግር ያለባት የቅርብ ዘመድ አለ

አዎ ☐ የለም ☐

በባለፉት የእርግዝናዎ የህብረ ስራስር ትቦ አለመግጠም ገጥመዎት ያዉቃል

አዎ ☐ የለም ☐

2.2 ከባለቤትዎ ጋር ዝምድና አለዎት አዎ ☐ የለም ☐

ክፍል 3 የናት የወሊድና የስነ ተዋልዶ ምክንያቶች

3.0 ስንት ልጅ ወልደዋል ----------

3.1 ከሚከተሉት ዙሪያ ታሪክ አለዎት

ፅንስ ማቋረጥ ☐ ሞቶ የተወለደ ☐ ጨቅላ ህፃን የሞተ ☐

3.2 አቅደዉ ነዉ ያረገዙት አዎ ☐ የለም ☐

3.3 ቅድመ ወሊድ የእርግዝና ክትትል ነበረዎች አዎ ☐ የለም ☐

ክፍል 4 የእናት በእርግዝና ወቅት የህክምና እና የመደሀኒት ታሪኮች

4.0 ከሚከተሉት የህመም አይነቶች ዉስጥ በዶክተር ተነግረዎት ያዉቃል የደም ግፊት ፣የእንቅርት መብዛትና ማነስ ፣የሽንት ትቦ ህመም ፣እጢ ፣የሚጥል በሽታ ፣የስኪር ህመም አዎ ☐ የመል ☐

4.1 አዎ ከሆነ የወሰዱትን መድሃኒት ይጥቀሱ ----------

4.2 ማንኛዉም አይነት የወሊድ መከላከያ ተጠቅመዉ ያዉቃሉ አ☐ የለም ☐

4.3 ህክምና አድርገዉ ያዉቃሉ --------

4.4 ትኩሳትና ከትኩሳት የሚመጡ ህመሞች ታመዉ ያዉቃሉ አዎ☐ የለም ☐

4.5 አዎ ከሆነ የወሰዱትን አለ አዎ ☐ የለም ☐

ክፍል 5 እናት በኬሚካል የተጋላጭ ነት መጠን መጠይቅ

5.0 በመኖሪያ አካባቢዎ የቆሻሻመጣያ ወይም ኢንዱስትሪ አለ አዎ ☐ የለም ☐

5.1 አዎ ከሆነ ከመኖሪያ ቤትዎ ምን ያህል ይርቃል ----------

5.2 ከቤት ስራ /ቦታዎ /ማንኛዉም ቦታ ፀረ ተባይ / አረም መዲሀኒት ተጠቅመዉ ያዉቃሉ ☐ አይደለም ☐

5.3 ሳሙና .ሙቅ ዉሃ ተጠቅመዉ ያዉቃሉ አ☐ የለም ☐

5.4 አዎ ከሆነ ምን ያህል ደቂቃ

5.5 የህክምና ጨረር ተደርጎለዎት ያዉቃል አዎ ☐ የለም ☐

ክፍል 6 የእናቶች የስነ -ምግብና ፎሊክ አሲድ አወሳሰድ መጠይቅ

6.0 ከማርገዝዎ በፊት ክብደትዎ ምን ያህል ነበር ----------

6.1 ማንኛዉም አይነት ፎሊክ አሱድ ወስደዉ ያዉቃሉ በፍፁም ከፅንስ በፊት 3 ወር የመጀመሪያ እርግዝና ወራት ከፀነስኩ ከ3 ወር በኋላ

ክፍል 7 የእናት አኗኗር ዘይቤ መጠይቅ

7.0 ቡና ወይም ሻይ ወስደዉ ያዉቃሉ አዎ ☐ የለም ☐

7.1 አዎ ከሆነ የትኛዉን አይ ነት ቡና ☐ ሻይ ☐

7.2 በቀን ምንያህል ኩባያ ይወስዳሉ

7.3 የባህል መድሃኒት በእርግዝና ወቅት ወስደዉ ያዉቃሉ ----------

7.4 የአልኮል መጠጥ ወስደዉ ያዉቃሉ አዎ ☐ የለም ☐

7.5 መልስዎ አዎ ከሆነ ምንያህል ቀን

ከ500 ሚሊ ሊትር በላይ ☐ በቀን ከ500 ሚሊ ሊትር በታች ☐ 500 ሚሊ ሊትር በታች ☐

7.6 ትንባሆ ያጨሳሉ አዎ ☐ የለም ☐

7.7 በቤትዎ ወይም በስራ ቦታዎ ሲሆኑ ትንባሆ ጭስ ተጋላጭ ነዎት አዎ ☐ አይደለሁም ☐

ጨርሰናል
አመሰግናለሁ !!(1)