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Association between ABO Blood groups, Socio-demographic and Behavioral factors with risk of Hypertension in Dangila and Durbete hospitals, northwest Ethiopia

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DEPARTMENT OF BIOLOGY

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MSc THESIS

BY:

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BAHIR DAR, ETHIOPIA

BAHIR DAR UNIVERSITY
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ASSOCIATION OF ABO BLOOD GROUPS, SOCIO-DEMOGRAPHIC AND BEHAVIORAL FACTORS WITH RISK OF HYPERTENSION IN DANGILA AND DURBETE HOSPITALS, NORTH WEST ETHIOPIA

A THESIS SUBMITTED TO COLLEGE OF SCIENCE, BAHIR DAR UNIVERSITY, IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF MASTERS OF SCIENCE IN BIOLOGY (GENETICS)

BY:

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OCTOBER, 2020

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DECLARATION

I declare that this thesis is my original work in partial fulfillment for the requirements for the degree of Master of Science in Biology (Genetics). All the sources of the materials used for this thesis and all people and institutions who gave support for thesis work are fully acknowledged.

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APPROVAL SHEET

As a thesis research advisor, I hereby certify that I have read and evaluated this thesis prepared under my supervision, by Abebe Kindie entitled as "Association of blood groups, socio-demographic and behavioral factors with risk of hypertension in Dangila and Durbete Hospitals, North west Ethiopia". I recommended the paper to be submitted as fulfilling the requirement for the Degree of Master of Science in Biology (Genetics).

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Advisor	Signature	Date

As members of the board of examiners for the MSc thesis open defense examination, we certify that we have read and evaluated the thesis prepared by Abebe Kindie and examined the candidate. We recommended the thesis to be accepted as fulfillment for the requirements of the Degree of Master of Science in Biology (Genetics).

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External Examiner	Signature	Date

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DEDICATION

‘‘I dedicate this thesis to my family especially my brother who lives in abroad for his continuous follow up, financial support and initiation until the completion of this work”.

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LIST OF ABBREVIATIONS

AH	Arterial Hypertension
AOR	Adjusted Odds Ratio
BP	Blood Pressure
CHD	Coronary Heart Disease
CHF	Congestive Heart Failure
CNS	Central Nervous System
COR	Crude Odds Ratio
CSA	Central Statistics Agency
CVD	Cardiovascular Disease
DALY	Disability- Adjusted Life Years
DASH	Dietary Approaches To Stop Hypertension
DBP	Diastolic Blood Pressure
DM	Diabetes Mellitus
HFD	Health Fitness Dynamics
HIV/ AIDS	Human Immuno-Deficiency Virus/ Acquired Immune Deficiency Syndrome
HTN	Hypertension
NCD	Non-Communicable Disease
OPD	Outpatient Department
OR	Odds Ratio
RH	Rhesus Factor
SBP	Systolic Blood Pressure
SPSS	Statistical Package For Social Science
SSA	Sub-Saharan Africa
WHO	World Health Organization

ABSTRACT

Hypertension has become a growing public health concern in developing countries. Hypertension is one of the most common complex genetic disorders, with genetic heritability averaging 30%. Many factors contribute to the development of hypertension which can broadly be categorized into two; modifiable and non-modifiable. The present study was performed to assess the association between ABO blood types and other factors with hypertension in Dangila and Durbete hospitals. A Hospital-based case control study was conducted from June to July 2020. Data was collected from a total of three hundred eighty four individuals who visited the hospital for medical attention. By using simple random sampling techniques, 192 case and 192 control subjects were selected. Necessary information on associated factors of hypertension was collected using a questionnaire.ABO and Rh blood group types were determined by taking a venous blood sample from the participants. Based on multivariate logistic regression, Age, marital status, residence, educational level, alcohol drink, animal source food consumption and physical activity were significantly associated with hypertension ($p < 0.05$) but there was no statistically significant association between ABO blood group (X^2=1.98, 3, N=384 and p=0.577) and Rh factor (X^2 =0.62, 1, n=384 and p=0.557) with hypertension. Blood group O was the most prevalent blood group and AB was the least prevalent both in case and control study group. Under behavioral factors, alcoholic people were2.26 times hypertensive than non-alcoholic ones (p=<0.001, AOR 95% CL=2.26(1.38, 3.71)). Physically inactive people were 2.19 times more likely to develop the disease than physically active ones (p=0.006, AOR 95% CL= 2.19 (1.27, 3.54)).Regarding animal source food consumption, those who eat animal source food were1.48 times more susceptible than who do not eat animal source food (p= 0.014, AOR 95% CL=1.48(0.90, 2.41)). Hence, healthy lifestyle changes like change alcoholic use, promotion of balanced diet complemented by regular physical activity should be promoted to reduce the risk of hypertension.

Key Words: Association, Behavioral Factor, Blood groups, Hypertension, Socio-demographic

1. INTRODUCTION

1.1. Background

In modern medicine, relationship of blood groups to disease has become an interesting area of research because of a known genetic connection of specific blood groups to certain diseases, in certain population. Thus far, several reports have suggested important associations between ABO blood groups and various diseases, for example, peptic ulcer, gastric cancer, pernicious anaemia (Clarke, 1959), Certain bacterial and viral infections (Anstee, 2010), hypertension (Miller *et al*., 1979; Sachdev, 2011 and Nishi *et al*., 2012) and ischemic heart diseases (Nemesure *et al*., 2006; He *et al*., 2012).

Some studies have been done to discern the role of genetic factors in the pathophysiology of hypertension. One such important genetic factor is individual's blood group system. Blood groups are genetically determined and hypertension is one of the most common complex genetic disorders, with genetic heritability averaging 30%. Although our understanding of the pathophysiology of blood pressure has increased, in 90 to 95% of cases the aetiology is still largely unknown. Many factors contribute to the development of hypertension which can broadly be categorized into two; modifiable and non-modifiable. Modifiable risk factors such as adiposities, age, stress, high salt intake, overweight and obesity are strongly correlated with high blood pressure whereas genetic factors remain as important non–modifiable predisposing factors in the development of hypertension (Braunwald *et al*., 2001).

A study from Himachal Pradesh, India, reported that the incidence of hypertension was highest in O group, followed by A, B and AB, suggesting O blood group individuals have more risk of developing Hypertension (Lester, 2019). A tribal-population based study from Rajasthan, India, found that people having B blood group were more susceptible for hypertension (Sachdev, 2011).

Study done in African population stated that there was no significant association between systolic Blood Pressure (BP) and any of the blood type, however they observed high diastolic (BP) was associated with B, O and Rhesus Factor (Rh) D type (WHO, 2001). Another study in White population could not find any significant difference in frequency distribution of blood groups between essential hypertensive patients and controls and it was concluded that, although an inherited factor in essential hypertension is accepted, there was no association with ABO inheritance (Maxwell and Maxwell, 1955).

Hypertension has become a growing public health concern in developing countries. For the developed countries, the trend emerges somewhat in the character of a paradox: remarkable successes achieved in under nutrition and infectious disease control within the last century has produced a sharp decline in infant mortality, as a result of which life expectancies have increased considerably. However, non communicable diseases have replaced infectious diseases as the major causes of morbidity (Perpetua, 2003). Given current urbanization and industrialization efforts in Ethiopia (Lester, 2019) however, together with improvements in health services, a demographic transition in which the proportion of adults and aged population increase significantly the younger age group. Expectedly, this would lead to a rise in the prevalence of chronic diseases that are generally more common among older age populations (Lester, 2019). Little is known about the magnitude and factors of hypertension in Ethiopia, however, recent evidences indicate that hypertension and raised blood pressure are increasing partly because of the increase in risk factors including smoking, obesity, and use of alcohol and lack of exercise (Fikru Tesfaye*et al*., 2007).

1.2. Statement of the problem

The prevention and control of hypertension has not received attention in many developing countries. In these countries health care resources are overwhelmed by other priorities including HIV/AIDS, tuberculosis, and malaria. However, the rapid urbanization of this country has created a change in the lifestyles of population in terms of nutrition, physical activities and behaviors such as smoking, alcohol and drug use among urban dwellers, which may impact on the risk of developing Non-communicable Diseases (NCDs). Despite its high prevalence in sub-Saharan Africa (SSA), hypertension was not given due attention. An increasing burden of hypertension in this region could thus result in grave consequences, as only very few people get treatment, and control is likely to be low. Through early detections, simple life style modifications and with the help of modern medical treatment, the problem of hypertension, ″the silent killer", can be largely controlled and patients with hypertension can lead a prolonged and healthy life. However, researches done in relation to this problem were very limited in Ethiopia. Therefore, the present study was performed to assess the relation between ABO and Rh blood types, socio-demographic and behavioral factors with hypertension in Dangila and Durbete hospitals.

1.3. Significance of the study

The World Health Assembly of the World Health Organization in 2000 passed a resolution, with a goal to support Member States in their efforts to reduce the toll of morbidity, disability and premature mortality related to NCDs. Specific strategies adapted at the meeting focus on assisting Member States in mapping the emerging epidemics of NCDs in their respective countries and analyzing the socio-economic, behavioral and political determinants of observed trends. This will provide guidance for policy, legislation and the financial commitment of nations towards NCDs. An appropriate first step towards initiating NCDs risk factor surveillance system therefore, is for countries to conduct a „baseline" nationwide survey, where this has not been done and provide an effective source of evidence to support and guide the development and implementation of specific prevention and control program. By offering up to date information on the status of hypertension, it may help the prevalence and risk factors of the diseases and thus facilitate a future assessment of their burden and impact. In turn, this should enable relevant

health administrators to develop comprehensive and appropriate community-based health promotion strategies to encourage healthy lifestyles among its populations, detect cases early enough, and choose. This study provides a baseline data on the association and possible factors of hypertension in Dangila and Durbete hospital, Northwest Ethiopia.

1.4. Objectives of the study

1.4.1. General objective

- The general objective of this study was to assess the association between ABO and Rh blood groups, socio-demographic and behavioral factors with the risk of hypertension in Dangila and Durbete Hospitals.

1.4.2. Specific objectives

The specific objectives of this study were:

- ✓ To assess the distribution of blood groups (ABO and Rh) among case and control study groups.
- ✓ To identify socio-demographic factors that increases the risk of hypertension in the study area.
- ✓ To identify behavioral and life style factors that increases the risk of hypertension in the study area.

2. LITERATURE REVIEW

2.1. Meaning of Hypertension

World Health Organization (WHO) defines hypertension as systolic blood pressure≥140 mmHg or a diastolic blood pressure≥90 mmHg or prior diagnosis of hypertension and taking antihypertensive drugs (Mathers *et al*., 2009;Weber *et al*., 2014;Kasper*et al*., 2015). About 80–90% of hypertension is primary hypertension and has no known cause. Secondary hypertension takes a share of 5–20% of hypertension cases and has different causative factors that could be preventable(Weber *et al*., 2014).Blood pressure is summarized by two measurements, systolic and diastolic, which depend on whether the heart muscle is contracting (systole) or relaxed between beats (diastole). Normal blood pressure at rest is within the range of 100-140mmHg systolic (top reading) and 60-90mmHg diastolic (bottom reading). High blood pressure is said to be present if it is persistently at or above 140/90 mmHg. Hypertension is a leading risk factor of mortality, followed by tobacco use and diabetes mellitus (DM). It is the fifth cause of Disability-adjusted life years (DALYs) lost globally. Hypertension doubles the risk of Cardiovascular Disease(CVD): coronary heart disease (CHD), congestive heart failure (CHF), ischemic stroke and hemorrhagic stroke, renal failure and Peripheral arterial disease (PAD). Systolic blood pressure causes 51% and 45% of deaths due to stroke and ischemic heart disease respectively (Mathers *et al*., 2009;Weber *et al*., 2014;Kasper *et al*., 2015).

2.2. Epidemiology of hypertension

Arterial Hypertension (AH) is the most common cardiovascular disease and is a major public health problem in both developed and developing countries. It produces a marked effect on patients, relatives and society, either because of hypertension present or through its complications (stroke, heart attack, ischemic heart disease, renal dysfunction and heart failure) which can produce premature death or permanent disability. The risk of developing a cardiovascular complication is higher when the individual combines hypertension with other risk factors such as hyper cholesterolemia/dislipidemia or smoking. It is known that more than 95%

of hypertensive patients in the community are of essential or idiopathic/unknown an etiology and only a small percentage have an identifiable cause (secondary hypertension). Epidemiological evidence shows that there are several factors which play an important role in the development, evolution and prognosis of arterial hypertension, some of them non-modifiable, such as age, sex, ethnicity and heredity, and others modifiable, such as body weight, alcohol intake and sedentary life (Antezana *et al*., 1998).

In the year 2000, nearly one billion people or 26% of the adult population of the world had hypertension. It was common in both developed (333 million) and developing (639 million) countries; however rates vary in different regions as low as 3.4% (men) and 6.8% (women) in rural India and as high as 68.9% (men) and 72.5% (women) in Poland (5 Study done in Vietnam reported that the prevalence of hypertension was 14.1%, of whom, only 17.4% were aware of their hypertensive status. Men had higher hypertensive than women and age was positively associated with hypertension. Another study reported that an overall of 26.4% of the adult population in 2000 had hypertension (26.6% men and 26.1% women), and 29.2% were projected to have this condition by 2025. The number of adults with hypertension in 2025 was predicted to increase by about 60% to a total of 1.56 billion (Kearney *et al*., 2005).

A study in Tibet reported that the crude prevalence of hypertension was 55.9%. The age standardized prevalence was 57.1% for the whole Sample using the 2001 WHO World Standard Population (Zhao *et al.,* 2012). A study in US National Health and Nutrition Examination Survey 2007–2010 study reported that there was a statistically significant difference in the age-adjusted prevalence of hypertension between those with abdominal obesity (35.0%) and without abdominal obesity (21.0%). Consistently across all levels of the demographic and other covariates, participants categorized as abdominally obese had a higher prevalence of hypertension when compared with participants categorized as not abdominally obese. These differences, with the exception of the 80 or more years of age category, were all statistically significant (Ostchega *et al*., 2012).

2.3.Hypertension in Africa

It has been suggested that the prevalence of cardiovascular disease and hypertension was increasing rapidly in sub-Saharan Africa (SSA). The current prevalence in many developing countries, particularly in urban societies, was said to be already as high as those seen in developed countries (Addo *et al.,* 2007). Studies in sub Saharan Africa reported that hypertension was the most prevalent risk factor for CVD. The crude prevalence of hypertension ranged from 19.0% in Tanzania to 32.0% in Namibia. The age adjusted prevalence was 19.3% in Nigeria, 21.4% in Kenya, 23.7% in Tanzania, 38.0% in Namibia and 18.2% in Sudan (Hendriks *et al.,* 2012).

The prevalence of hypertension was estimated by a population-based survey in 1798 Cameroonian subjects aged 25-74 years which showed the age-standardized prevalence of hypertension to be significantly higher in the urban than the rural area (16.4% in urban men and 12.1% in urban women, and 5.4% in rural men and 5.9% in women) (Mbanya *et al*., 1998).

A survey on detection, management, and control of hypertension in 12 villages in Ashanti, Ghana established an adult prevalence rate of 28.7% overall and comparable in men and women, but higher in semi-urban villages (32.9% versus 24.1%), and increased with age. A study in Ghana showed that the prevalence of hypertension ranged from 19 to 48% between studies. Sex differences were generally minimal whereas urban populations tended to have higher prevalence than rural population in studies with mixed population types. Factors independently associated with hypertension included older age group, over-nutrition and alcohol consumption (. A national cross-sectional study in Tunisia established the prevalence of elevated BP was 35.1%. And a community-based study done in Uganda showed the age-standardized prevalence of hypertension was 30.5 % (Wamala *et al*., 2009).

Hospital-based cross-sectional study, which took place at the Out-Patients Department of St. Mary's Catholic General Hospital in Nigeria, showed that out of the 405 subjects that participated in the study, 124 (30.6%) were hypertensive. The study revealed that age, occupation, body mass index, waist circumference and hip circumference were significantly associated with high blood pressure. A study in Eritrea reported that the prevalence of

hypertension in the general population was 16%, with the highest levels in unemployed people and local merchants (24%) and the lowest levels in students (7.1%)(Mufunda *et al.*, 2006).

2.4. Hypertension in Ethiopia

Very few studies in hypertension epidemiology have been undertaken in Ethiopian. A population-based prevalence study conducted in Addis Ababa, Ethiopia reported a 31.5% and 28.9% prevalence of hypertension among males and females respectively (Fikru Tesfaye *et al*., 2009).

Another study done in Gondar showed that the overall prevalence of hypertension was 28.3%, of whom more than a third (37.0%) did not know they had hypertension (Akilew Awoke *et al.*, 2012).

A comparative study in Buta jira showed that the prevalence of hypertension was significantly higher among Khat chewers (13.4%) than non-chewers (10.7%) (Workineh Getahun *et al*., 2010).

A study conducted in Gilgel Gibe Field Research Center reported that the overall prevalence of CNCD was 8.9% (7.8% men and 9.8% women). The specific observed prevalence was 0.5% for diabetes mellitus (DM), 2.6% for hypertension, 3.0% for cardiovascular diseases, 1.5% for asthma and 2.7% for mental illness (Ayalew Tegegn Muluneh *et al*., 2012).

2.5. Risk Factors for Hypertension

2.5.1. Blood groups

All human blood may perhaps look alike, but it's not so. Based on the presence or absence of antigens on erythrocyte membrane, blood is categorized into different groups. So far, International Society of Blood Transfusion has approved thirty-six major blood groups in humans (Storry *et al*., 2019).The first blood group, ABO, was discovered in 1900 by Austrian scientist, Karl Landsteiner. The ABO blood group antigens (A, B, AB and O) are widely expressed on the membranes of red cells and a variety of tissue cells (Hosoi, 2008).Apart from their importance in blood transfusion process, the ABO and Rh blood groups are useful in other fields as well, like genetics, certain medico legal issues and to study population migration patterns. In modern medicine, relationship of blood groups to disease has become an interesting area of research because of a known genetic connection of specific blood groups to certain diseases, in certain population. Thus far, several reports have suggested important associations between ABO blood groups and various diseases, for example, peptic ulcer, gastric cancer, pernicious anaemia (Clarke, 1959) , certain bacterial and viral infections, hypertension and ischemic heart diseases (Nemesure *et al*., 2006) .

Some studies have been done to discern the role of genetic factors in the pathophysiology of hypertension. One such important genetic factor is individual's blood group system. Blood groups are genetically determined and hypertension is one of the most common complex genetic disorders, with genetic heritability averaging 30%. Although our understanding of the pathophysiology of elevated arterial pressure has increased, in 90 to 95% of cases the aetiology is still largely unknown. Many factors contribute to the development of hypertension which can broadly be categorized into two; modifiable and non-modifiable. Modifiable risk factors such as adiposities, age, stress, high salt intake, overweight and obesity are strongly correlated with high blood pressure whereas genetic factors remain as important non– modifiable predisposing factors in the development of hypertension (Braunwald *et al*., 2001).

Many efforts have been made in India and other parts of the world to understand the blood type and elevated BP relationship. A study from Himachal Pradesh, India, reported that the incidence

of hypertension was highest in O group, followed by A, B and AB, suggesting O blood group individuals have more risk of developing Hypertension (Nishi *et al*., 2012).

A tribal-population based study from Rajasthan, India, found that people having B blood group were more susceptible for hypertension (Sachdev, 2011). Study done in African population stated that there was no significant association between systolic BP and any of the blood type, however they observed high diastolic BP was associated with B, O and Rhesus (Rh) D type (Nemesure *et al.,* 2006) .Another study in White population could not find any significant difference in frequency distribution of blood groups between essential hypertensive patients and controls and it was concluded that, although an inherited factor in essential hypertension is accepted, there was no association with ABO inheritance. As it is evident, the results from previous studies are somewhat inconclusive and it may be due to variance in the study population, study design, regional and racial differences that might have affected the results (Maxwell and Maxwell, 1955).

2.5.2. Socio-Demographic factors

The study that was conducted in Bangladesh showed that the prevalence of Hypertension was high among Urban and married than Rural and single respectively. The risk of systolic Hypertension increases as the age of individual increases. According to one study, men have a higher prevalence of hypertension compared to women until the sixth decade of life (Sullivan, 2012).

2.5.3. Behavioral factors

Alcohol drinking can increase the risk of Hypertension by different mechanisms. There is a strong link between alcohol and NCDs, particularly cancer, cardiovascular disease (Hypertension). According to a study in rural South Africa, the Prevalence of Hypertension was higher among alcohol drinkers. The prevalence of pre-hypertension is more among alcohol drinkers in Uganda (Nuwaha and Musinguzi, 2013).

Cigarettes smoking- Both maternal and paternal smoking of ≥15 cigarettes/day during pregnancy was associated with increased risks of hypertension (OR 1.19, 95% CI 1.09 to 1.29, and OR 1.18, 95% CI 1.12 to 1.25, respectively). Large Mendelian randomization Meta analysis suggests

that smoking is causally related to higher level of resting heart rate, but not to alterations in blood pressure and risk of hypertension. Active smoking in the third trimester was associated with reduced odds of preeclampsia and gestational hypertension with the strongest association among continuous smokers (for preeclampsia, OR = 0.57 [95% CI = 0.46–0.70]) (Engel *et al*., 2013).

Khat chewing- khat chewing is among the known risk factors of hypertension based on the study conducted in Yemen. Another study revealed that khat chewing aggravates the development of Hypertension and lose of memory. Khat chewing is also amongst the known risk factors of Heart failures and hypertension as well (El –Menyar*et al*., 2015).

Family history of Hypertension- The prevalence of hypertension was significantly higher in those with a Family History of Hypertension based on evidence from Sri Lanka. Based on the study that was conducted in Japanese Children, Hypertension was more prevalent among those who have family History of Hypertension. Majority of the Hypertensive participants have family History of Hypertension. Another study from Japan and China showed the same result with the above (Igarashi *et al*., 2016).

Physical activity –Physical activity is defined as any bodily movement produced by contraction of skeletal muscles that increases energy expenditure above resting levels and comprises of routine daily tasks, such as commuting, occupational tasks, or household activities, as well as purposeful health- enhancing movements/activities. Ten thousand steps per day are needed in order to reduce high Blood Pressure. Physical activity is associated with low risk of hypertension. Physical activity is an advisable intervention for the majority of Hypertensive patients (Brooks and Ferro, 2012).

Heavy Salt consumption – Salt restriction lowers mean BP in Chinese adults with the strongest effect among Hypertensive. Low sodium high potassium salt-substitutes effective in lowering both Systolic and Diastolic blood pressure and offers a simple, low-cost approach for Hypertension control among Tibetansin China. Hypertension and excessive sodium intake in adults are major public health problems in Shandong Province, China. Both relatively high levels of dietary sodium intake and gradual increases in dietary sodium are associated with future increases in blood pressure and the incidence of hypertension in the Japanese general population (Takase *et al*., 2015).

Fat consumption – in Japanese Americans that change in the intra-abdominal adipose tissue deposit is the critical region related to the development of Hypertension. Although high- fat dairy (e.g. whole milk, cheese) consumption was not associated with BP change in the present study, the significantly inverse association found between total high- fat dairy intake and Hypertension (HTN) incidence in the discrete-time hazard model may indicate that keeping up an appropriate level of high- fat dairy consumption in the context of an energy-balanced, healthy dietary pattern may have a small, cumulative benefit on HTN prevention over the long run. Health Fitness Dynamics (HFD) did not increase baseline BP, but enhanced the Hypertensive response to Ang II compared to a normal fat diet. Dietary strategies for the prevention of Hypertension include reducing sodium intake, limiting alcohol consumption, increasing potassium intake and adopting an overall dietary pattern such as the DASH (Dietary Approaches to Stop Hypertension) diet or a Mediterranean diet. Reducing total fat intake is a priority for many people seeking to reduce or maintain a lower body weight. Herbs and spices improve liking of lower fat versions of foods, such as French toast, chicken and vegetables (Polsky *et al*., 2014).

3. MATERIALS AND METHODS

3.1. Description of the study area

The research was conducted in Dangila and Durbete hospitals which are found at Dangila and Durbete towns, respectively.

Dangila is one of the administration towns in Awi Zone of northwestern Ethiopia which is found about 489km northwest of Addis Ababa and 78km south east of Bahir Dar city. Dangila town is bordered on the south by Faggeta Lekoma, on the southwest by Guangua on the northwest by the Jawi and on the northeast by the west Gojjam Zone. Dangila town administration is situated at an altitude of 2200m above sea level with latitude and longitude of 11°16′N and 36°5′E respectively. Dangila town has total area of about 918.4km^{2}and estimated total population of 26,704. The town consists of 10 kebeles with five urban and five rural kebeles. From the population, 12,916 were men and 13,788 were women (Central Statistics Agency (CSA), 2007).

Durbete town is capital city of South Achefer woreda at north western of Ethiopia. South Achefer woreda is located at 502 kilo meters from Addis Ababa, the capital city of Ethiopia. It is geographically located at 11^{0}25'32" N latitude and 36^{0}17'42" E longitudes and has an area of 170,000 km^2.The altitude of the district ranges from 1,500 to 2,500 meters above sea level and the mean annual rainfall varies between 1,400 to 1,594 mille litre (ml) with the average annual temperature range from 25c^o to 29c^o. The district has many banks of rivers, streams, ponds, stagnant marshy water bodies and irrigation canals (CSA, 2007).

3.2. Study design

Hospital-based case control study design was used to assess the association between ABO blood group and other factor with hypertension in Dangila and Durbete primary Hospital during the study period.

3.3. Study population

Cases were patients who had a confirmed diagnosis of hypertension attending a regular OutPatient Department (OPD) follow-up in Dangila and Durbete primary hospital. Eligible cases were all patients who came to the hospital at the time of data collection which last a month as they visit the hospital monthly based on their follow up card schedule. Consecutive sampling technique was used to include cases. Based on the above criteria, 192 cases (hypertensive) study population were selected.

A control is defined as an individual who visited the hospital for other than hypertension diseases. By using simple random sampling technique 192 control (normotensive/non-hypertensive study population was recruited and for the selection of control, a person blood pressure level was primarily measured. After checking the participant's non-hypertensiveness, other required data are collected.

3.4. Inclusive and exclusive criteria

3.4.1. Inclusion criteria

- All Hypertension disease patients who have a regular follow up in the hospital, those people who visited the hospital for other medical attention age above 18 years and have consent to participate in the study were included.

3.4.2. Exclusion criteria

- Pregnant women
- Diabetes Mellitus patients
- Person who are not willing to participate in the study are excluded.

3.5. Sample size determination

The representative samples were selected using random sampling technique from individuals who address to give socio-demographic information, behavioral factors and blood samples for the diagnosis of blood types. For estimating the sample size (N) statistical formula at 5% level of significance was used and since the prevalence of hypertension with specific blood group and

other associated factor was not known in the study area, P was taken to be 50% for the calculation. So the formula is as follows:

$N=Z^2P\ (1-P)/d^2$ (Niang *et al.*, 2006)

1-P=q

N=the minimum required sample size

Z=1.96 at 95% confidence interval

d= margin of sampling error (5% margin error used)

p=prevalence of the association between ABO/Rh blood group and other associated factors with hypertension.

$N= (1.96)^2\ 0.5\ (1- 0.5) / (0.05)^2 =384.$

3.6. Data collection

3.6.1. Questionnaire survey

As recommended in the WHO STEP wise Approach guidelines on NCD risk factor surveillance, the survey comprises interview-structured questionnaire to assess the socio-demographic factors and behavioral characteristics of the study subjects. Questionnaires were designed to collect data on independent variable that may have an association with hypertension. Before prepare the questionnaire literatures were consulted. Then questionnaire was prepared and translated in to Amharic version by professional language translator before the data collection. Socio-demographic characteristics and behavioral factors for hypertension disease were collected from the participants by face to face interview using Amharic version of the structured questionnaire.

3.6.2. Blood sample collection procedures

Data was collected from a total of three hundred eighty four individuals who visited the hospital for medical attention. When the participants were volunteer to participate in the study, a 0.5ml of venous blood was taken as part of the usual diagnosis at the hospital, then blood group ABO and Rh type was identified for both hypertension patients and non-hypertensive subjects. The blood pressure was measured by mercury sphygmomanometer two times and mean was calculated for accuracy. Data was collected in collaboration with professionals working in the OPD rooms.

For blood sample collection, the study participant's finger was lanced by qualified health care professionals in the hospital. The collected blood from each study participants was dropped on 3 clean microscopic slides labeled A, B and D. Then anti-A, anti-B and anti-D antibodies were added on the slides respectively and stirred. After 2 minute the samples were checked for agglutination under microscope to determine the blood group and Rh factor.

When a slide labeled A shows agglutination, the study participants blood group was determined to be blood group A. When a slide labeled B agglutinates, the blood group of study participants was recorded as blood group B. When both A and B slides agglutinate, the blood group of study participants was recorded as blood group AB and if blood cells of both slides A and B don't agglutinate, the blood group of study participants was recorded as blood group O.

In the same way Rh group was determined by the presence or absence blood cells clumping (agglutination) on slide labeled as D. When the blood cells on a slide labeled D clump together the study participant's blood was determined to be Rh positive but if no agglutination is seen on a slide labeled D, the study participant's blood was determined to be Rh negative. Safety precautions were taken while handling blood and disposing it.

3.7. Study Variables

Dependent variable: Hypertension is the dependent variable and it's defined as a systolic blood pressure at or above 140 mmHg and/or a diastolic blood pressure at or above 90 mm Hg.

Independent variables:

Blood groups: A, B, AB, O;

Rh factor: Rh+, Rh-;

Socio-demographic characteristics: Sex, Age, Marital status, Residence, Educational level, family history of hypertension.

Behavioral factors: Coffee drinking, Alcohol consumption, chewing khat, Cigarette smoking, Animal source fat consumption and Physical exercise;

3.8. Data Analysis

Collected data was compiled and analyzed using Statistical Package of Social Science (SPSS) software version 22. Chi-square test was used to find group association. P-value of < 0.05 was considered to be highly significant. Descriptive statistics like frequency, percentage and graphs were used. Both the bi-variable and multivariable logistic regression analyses were performed to assess the association between dependent and independent variables.

3.9. Ethical Clearance

Subjects who were volunteers to participate in the study were included after signing a written agreement. The protocol and the written consent were approved by the research community service office of college of science Bahir Dar University.

3.10. Operational Definitions

Hypertension: Defined as systolic blood pressure (SBP)≥140 mm of Hg and or diastolic blood pressure (DBP)≥90 mm of Hg. An individual previously diagnosed as hypertensive and presently under treatment was also considered as hypertensive.

Case: a hypertensive patient diagnosed and followed up in the hospital.

Control: patients or companions of patient not suffering from hypertension or diabetes visiting the hospital.

Coffee drinking: refers to drinking of coffee on a daily basis.

Alcohol use: refers to the consumption of local or manufactured alcohol beverages on a daily basis.

Khat chewing: Individuals who reported khat (Catha edulis leaves) use for 5 days or more in a week were considered to be regular khat chewers.

Smoking cigarette: was based on patients' history of using manufactured or locally-made tobacco 3 days or more a week.

Animal source food consumption: Eating food from animal sources and saturated fat at least 3 times a week.

Physical activity: Walking, jogging, swimming and cycling at least 30 minutes per day and other activities (Esaiyas kebede Gudina *et al*., 2013).

4. RESULTS

4.1. Socio-Demographic Factors

A total of 384 subjects were participated in this study of which 192 were hypertensive study group and 192 were non-hypertensive study group. Out of these 181(47.1%) were male and 203(52.9%) were females. From those hypertensive group, 85(44.3%) were males and 107(55.7%) were females. From non-hypertensive study group, 96(50%) were males and 96(50%) were females. Comparatively female participants were a few higher than male participants in the hypertensive study group (Table 1).

Regarding age, from the total study groups, 51(13.3%) were in the age between 18-28, 101(26.3%) were between 29-39, 97(25.3%) were between 40-50 and 135(35.2%) were above 50. Out of hypertensive study participants, the most prevalent age were above 50 years (87(45.3%)) and least prevalent age were 18-28 years(8(4.2%)). From non-hypertensive study groups, the age between 29-39 were more prevalent i.e. 64(33.3%). Least prevalent ages were between 40-50 age groups that is 37 (19.3%)(Table 1).

From the total 384 study participants, 65.6% have got married, 23.2% were single and 11.2% were either divorced or widowed. In both hypertensive and non-hypertensive study groups, 121(63%) and 131(68.2%) were married respectively. Most of the participants were married and from non-hypertensive study populations 2.6% of the participants were either divorced or widowed which is very few in number (Table 1).

From the total study group,209(54.4%) were urban dwellers whereas 175(45.6%) were rural dwellers. Within the hypertensive study group, 115(59.9%) out of 192 were live in urban area. In the non-hypertensive study population, 98(51%) were live in rural area (Table 1).

Concerning educational level, from total of 384 study populations, 82(21.4%) can’t read and write, 101(26.3%) can read and write, 83(21.6%) had learnt from grade 1-8, 58(15.1%) had learnt from grade 9-12, 60(15.6%) of the participants had an educational level of diploma and above. From the hypertensive study populations, participants who can’t read and write are more frequent (53(27.6%)), less frequent participants were having diploma and above 20(20.8%).

From normotensive study groups, participants who can read and write were higher 59(30.7%) whereas participants who can't read and write were less 29(15.1%)(Table 1).

In the hypertensive family member, 20(5.2%) had hypertensive family member and 364(94.8%) did not have hypertensive family member. From 192 hypertensive study populations, 11(5.7%) have hypertensive family member and 181(94.3%) did not have hypertensive family members. From 192 normotensive (non-hypertensive) study populations, 9(4.7%) had hypertensive family member and 183(95.3%) did not have hypertensive family members (Table 1).

Table 1: socio-demographic factors of study population in Dangila and Durbete Hospitals.

Variables	Category	Hypertension		
		Case	Control	total
Sex	Male	85(44.3%)	96(50%)	181(47.1%)
	Female	107(55.7%)	96(50%)	203(52.9%)
Age	18-28	8(4.2%)	43(22.4%)	51(13.3%)
	29-39	37(19.3%)	64(33.3%)	101(26.3%)
	40-50	60(31.3%)	37(19.3%)	97(25.3%)
	>50	87(45.3%)	48(25%)	135(35.2%)
Marital status	Single	33(17.2%)	56(29.2%)	89(23.2%)
	Married	121(63%)	131(68.2%)	252(65.6%)
	Divorced/widowed	38(19.8%)	5(2.6%)	43(11.2%)
Residence	Urban	115(59.9%)	94(49%)	209(54.4%)
	Rural	77(40.1%)	98(51%)	175(45.6%)
Educational level	Can't read and write	53(27.6%)	29(15.1%)	82(21.4%)
	Read and write	42(21.9%)	59(30.7%)	101(26.3%)
	Grade 1-8	51(26.6%)	32(16.7%)	83(21.6%)
	Grade 9-12	26(13.5%)	32(16.7%)	58(15.1%)
	Diploma and above	20(10.4%)	40(20.8%)	60(15.6%)
Hypertensive family	Yes	11(5.7%)	9(4.7%)	20(5.2%)
	No	181(94.3%)	183(95.3%)	364(94.8%)

From the socio-demographic factors, age is one of the major factor for hypertension based on spss analysis ($p<0.001$). In this study, we classify age into four categories i.e 18-28, 29-39, 40-50 and above 50(Mihretie Kiber *et al*., 2019). So in this case participants whose age is above 50 are assumed as old age in which it accounts the highest percentage (45.3% for hypertensive and 25% for non-hypertensive participant) with a total of 35.2% from the whole study population. 29-39 age category ranks second in this study due to the presence of many participants in the control study group for this category (Figure 1).

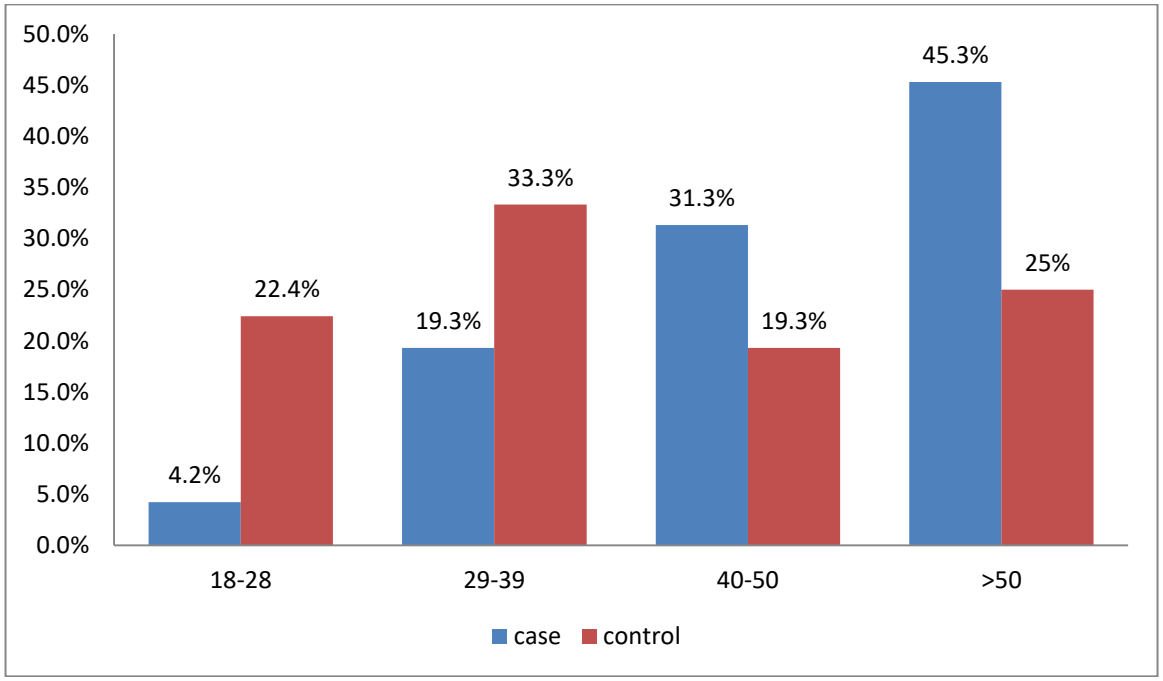

Figure 1: Distribution of age category in case and control study group of Dangila and Durbete Hospitals.

4.2. ABO and Rh Blood Types

From a total of 384 abo blood group sample collections, blood group O was highest which is 142(37%) followed by A (115(29.9%)), B (99(25.8%)), AB (28(7.3%)) respectively. From both hypertensive and normotensive study populations, blood group O was more prevalent and blood group AB was less prevalent. The percentage of blood group distribution in the hypertensive study group were 32.3%, 24.5%, 8.3 % and 34.9% for A, B, AB, O respectively. The percentage of blood group distribution in non-hypertensive study group were 27.6%, 27.1%, 6.3% and 39.1%for A, B, AB, O respectively.92.7% of the total study populations are Rh+ participants and 7.3% of the participants are Rh-. Majority of the participants are Rh+ and few participants are Rh- (Table 2).

Table 2: ABO and Rh Blood Types of study population in Dangila and Durbete Hospitals.

Variables	Category	Hypertension		
		Case	Control	Total
Blood group	A	62(32.3%)	53(27.6%)	115(29.9%)
	B	47(24.5%)	52(27.1%)	99(25.8%)
	AB	16(8.3%)	12(6.3%	28(7.3%)
	O	67(34.9%)	75(39.1%)	142(37%)
Rh factor	Rh +	180(93.8%)	176(91.7%	356(92.7%)
	Rh -	12(6.3%)	16(8.3%)	28(7.3%)

Blood groups are considered as one of the genetic factor for hypertension. But in our study blood group didn't statistically associate with hypertension (X^2=1.98, 3, N=384 and p=0.577). Blood group O accounts the highest percentage in both study population (34.9% for case and 39.1% for control) followed by A, B and AB blood groups(Figure 2).

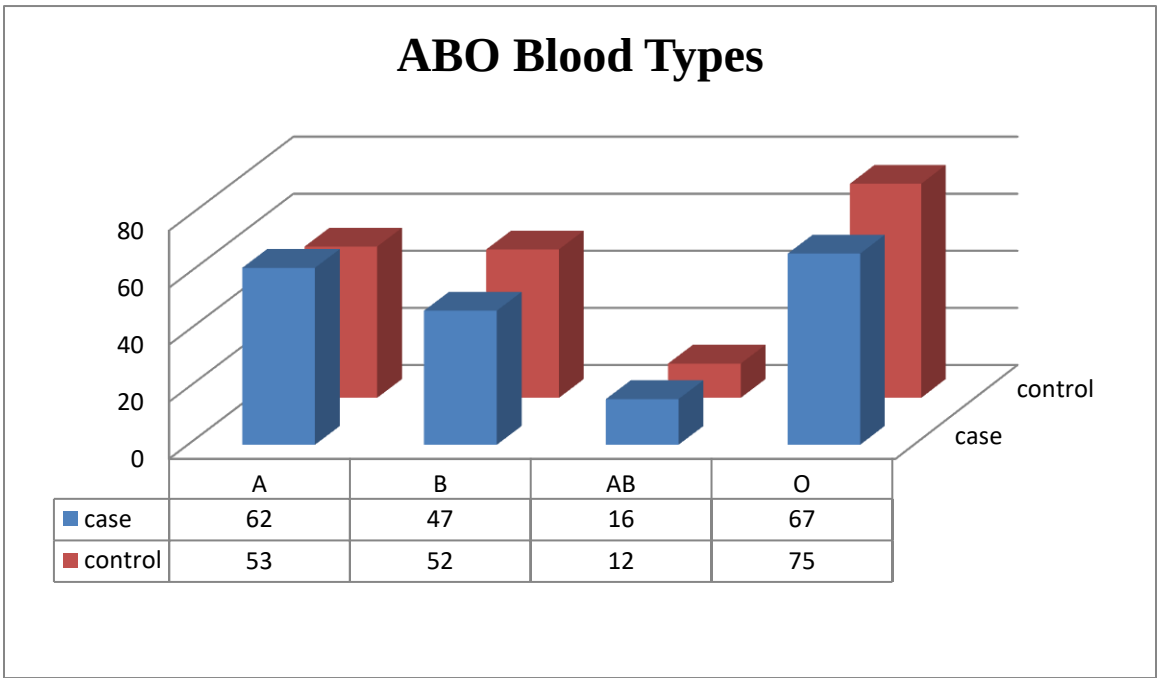

Figure 2: Distribution of ABO Blood types in case and control study group of Dangila and Durbete Hospitals.

Rh (Rhesus) factor was the second genetic factor for hypertension. From the total populations 92.7% of them were Rh positive. The rest 7.3% of them were Rh negative.

The chi-square value between Rh factor and hypertension was X^2 (1, n=384) =0.62, p=0.557. Based on this p value, Rh factor has no significant association with hypertension. Even if Rh factor is not statistically significant with hypertension, its odds ratio value is 1.36(Crude Odds Ratio (COR)=1.36) that means Rh+ individuals are 1.36 times more likely to be hypertensive than Rh- individuals (Figure 3).

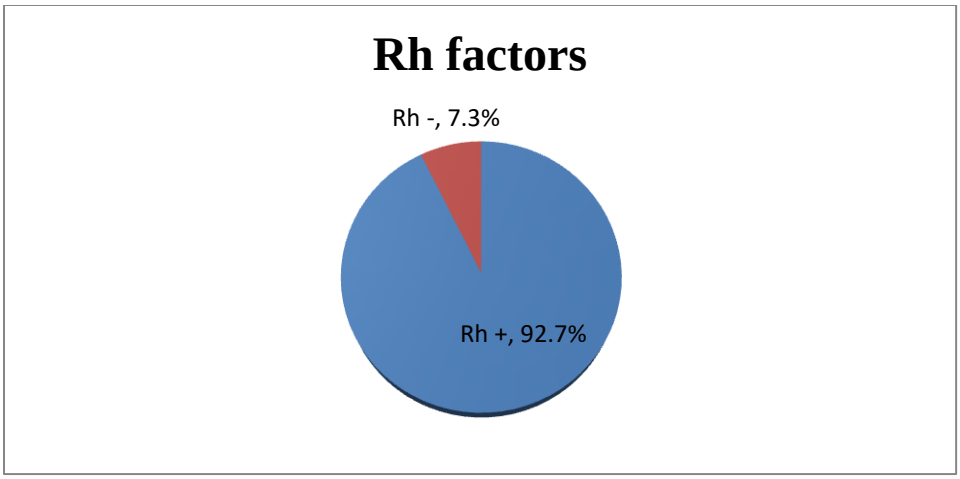

Figure 3: Distribution of Rh Blood Types in the total study group of Dangila and Durbete Hospital.

4.3. Behavioral And Life Style Characteristics

Based on the data collected on coffee drinking, most of the participants are drinking coffee in their way of life. Of the total population 74.2% of the participants are drinking coffee and 25.8% are not drinking coffee. In hypertensive study populations, as physicians prohibited the hypertensive patients not to drink coffee, we are asked their drinking habit before the occurrence of hypertension. So 148(77.1%) of hypertensive participants were have past history of drinking coffee and 44(22.9%) were not drinking coffee. In the non-hypertensive study populations, 137(71.4%) were current coffee drinkers and 55(28.6%) were not drinking coffee at all (Table 3).

In the alcohol consumption, from the total study populations, 52.9% were drinking alcohol and 47.1% were not drinking alcohol. There are more alcohol drinkers in the hypertensive populations than in the non-hypertensive populations. 122(63.5%) of the participants were alcohol drinker in the past in hypertensive study group and 81(42.2%) were also current alcohol drinkers from non-hypertensive study group (Table 3).

Chat chewing was less practiced in the study area based on the collected data. 91.4% of the total participants did not chew chat while 8.6% of the participant chews chat (Table 3).

The same is true for cigarette smoking even if it is a little bit more practiced than chat chewing. From the total study participants, 11.7% of the participant smokes cigarette but 88.3% of the participants did not smoke cigarette (Table 3).

Being one factor for hypertension, animal source food consumption was included in the questionnaire for data collection. From the total 384 study populations, 210(54.7%) had animal source food in their diet in a regular basis and 174(45.3%) did have animal source food in their diet in a regular basis. From hypertensive study group, people eating animal source foods were higher than people that did not eat which 117(60.9%) and 75(39.1%) respectively. In the non-hypertensive study group, it is vice versa, 93(48.4%) for people eat animal source food and 99(51.6%) for those who did not eat animal source food (Table 3).

Physical activity is also one of the other main associated factors for hypertension. As this is so, we have the following data on our study. 144(37.5%) from the total population did a regular physical exercise and 240(62.5%) of them were not doing regular physical exercise. Above have of the hypertensive study population didn’t exercise a regular physical exercise (Table 3).

Table 3: Behavioral factors of study population in Dangila and Durbete Hospitals.

Variables	Category	Hypertension		
		Case	Control	Total
Coffee drinking	Yes	148(77.1%)	137(71.4%)	285(74.2%)
	No	44(22.9%)	55(28.6%)	99(25.8%)
Alcohol drinking	Yes	122(63.5%)	81(42.2%)	203(52.9%)
	No	70(36.5%)	111(57.8%)	181(47.1%)
khat chewing	Yes	16(8.3%)	17(8.9%)	33(8.6%)
	No	176(91.7%)	175(91.1%)	351(91.4%)
Cigarette smoking	Yes	20(10.4%)	25(13%)	45(11.7%)
	No	172(89.6%)	167(87%)	339(88.3%)
Animal source food consumption	Yes	117(60.9%)	93(48.4%)	210(54.7%)
	No	75(39.1%)	99(51.6%)	174(45.3%)
Physical activity	Yes	59(30.7%)	85(44.3%)	144(37.5%)
	No	133(69.3%)	107(55.7%	240(62.5%)

4.4. Factors Associated With Hypertension

The binary logistic regression analysis started with unadjusted analysis in which each potential risk factor was assessed separately for its association with hypertension. Variables with p-values<0.25 on the unadjusted analysis were entered into a multivariable binary logistic regression model to find out risk factors independently associated with hypertension adjusting for other factors in the model (Table 4).

Table 4: Bivariate analysis of factors associated with hypertension in Dangila and Durbete Hospitals.

Factor	Category	Frequency	COR 95% CL	p- value
Sex	Male	183	1	0.262
	Female	201	1.26(0.84, 1.88)	
Age	18-28	51	1	<0.001
	29-39	101	1.12(0.65, 1.92)	
	40-50	97	3.14(1.83, 5.36)	
	>50	135	9.74(4.24, 22.41)	
Marital status	Single	89	1	<0.001
	Married	252	12.89(4.62, 36.01)	
	Divorced/widowed	43	8.23(3.14, 21.59)	
Residence	Urban	209	1.56(1.04, 2.33)	0.031
	Rural	175	1	
Educational level	Can't read and write	82	1	<0.001
	Read and write	101	2.57(1.41, 4.68)	
	Grade 1-8	83	1.15(0.61, 2.16)	
	Grade 9-12	58	2.25(1.13, 4.47)	
	Diploma and above	60	3.66(1.81, 7.38)	
Hypertensive Family	Yes	20	1.24(0.50, 3.05)	0.647
	No	364	1	

Blood group	A	115	0.76(0.47, 1.25)	0.577
	B	99	0.99(0.59, 1.65)	
	AB	28	0.67(0.29, 1.52)	
	O	142	1	
Rh factor	RH +	356	1.36(0.63, 2.97)	0.557
	RH -	28	1	
Coffee drinking	Yes	285	1.35(0.85, 2.14)	0.200
	No	99	1	
Alcohol drinking	Yes	203	2.39(1.58, 3.60)	<0.001
	No	181	1	
Chat chewing	Yes	33	0.94(0.46, 1.91)	0.856
	No	351	1	
Cigarette smoking	Yes	45	0.78(0.42, 1.45)	0.429
	No	339	1	
Animal source food consumption	Yes	210	1.66(1.11, 2.49)	0.014
	No	174	1	
Physical activity	Yes	144	1.79(1.18, 2.72)	0.006
	No	240	1	

Multivariate logistic regression was done for those factors that were significantly associated in binary logistic regression. The analysis showed that there was statistically significant association with the socio-demographic factors of age, marital status, residence and educational level with the disease hypertension. With behavioral and life style factors, hypertension is statistically associated with alcohol consumption, animal source food consumption and physical inactivity. According to this study, the age 50 and above are 8.66 times more susceptible for hypertension than the age between 18 and 28(p=<0.001, AOR 95% CL = 8.66(3.42, 21.96)). Participants who have got married are 11.17 times more likely to be hypertensive than those are single (p= <0.001, AOR 95% CL = 11.17(3.48, 35.81)). Urban dwellers are around 3.5 times more susceptible than

rural dwellers for hypertension (p=0.031, AOR 95% CL =3.49 (1.89, 6.46)). Under educational level, people who have learnt diploma and above are 9.25 more vulnerable than those who cannot read and write (p= <0.001, AOR 95% CL = 9.25(3.59, 23.79)).alcoholic people are 2.26 times hypertensive than non-alcoholic ones (p=0.003, AOR 95% CL=2.26(1.38, 3.71)). Physically inactive people are 2.19 time more likely to develop the disease than physically active ones (p=0.006, AOR 95% CL= 2.19(1.27, 3.54)).regarding animal source food consumption, those who eat animal source food are 1.48 times more susceptible than who do not eat animal source food (p= 0.014,AOR 95% CL=1.48(0.90, 2.41)). Even though blood group O was found to have higher distribution among hypertension disease patients than in the control group, there is no statistically significant association between blood group and hypertension (Table 5).

Table 5: Multivariate analysis of factors associated with hypertension in Dangila and Durbete Hospitals.

Risk factors	Case	Control	COR (95% CL	AOR (95% CL)	P-value
Age group					
18-28	8	43	1	1	<0.001
29-39	37	64	1.12(0.65, 1.92)	1.09(0.59, 2.03)	
40-50	60	37	3.14(1.83, 5.36)	2.49(1.36, 4.60)	
>50	87	48	9.74(4.24, 22.41)	8.66(3.42, 21.96)	
Marital status					
Single	33	56	1	1	<0.001
Married	121	131	12.89(4.62, 36.01)	11.17(3.48, 35.81)	
Divorced/widowed	38	5	8.23(3.14, 21.59)	8.13(2.76, 23.93)	

Residence					
Urban	115	94	1.56(1.04, 2.33)	3.49(1.89, 6.46)	
Rural	77	98	1	1	0.031
Educational level					
Can't read and write	53	29	1	1	<0.001
Read and write	42	59	2.57(1.41, 4.68)	3.11(1.53, 6.31)	
Grade 1-8	51	32	1.15(0.61, 2.16)	1.41(0.67, 2.99)	
Grade 9-12	26	32	2.25(1.13, 4.47)	4.60(1.86, 11.41)	
Diploma and above	20	40	3.66(1.81, 7.38)	9.25(3.59, 23.79)	
Alcohol drinking					
Yes	122	81	2.39(1.57, 3.60)	2.26(1.38, 3.71)	
No	70	111	1	1	0.003
Physical activity					
Yes	59	85	1.79(1.19, 2.72)	2.19(1.27, 3.54)	
No	133	107	1	1	0.006
Animal food					
Yes	117	93	1.66(1.11, 2.49)	1.48(0.90, 2.41)	
No	75	99	1	1	0.014

5. DISCUSSION

5.1. Blood Group And Rh Factor

This study was conducted to assess the association between ABO blood group and other factors with the disease Hypertension. The associated factors include behavioral characteristics like coffee drinking, alcohol consumption, khat chewing, cigarette smoking, animal source food consumption, physical activity and socio-demographic factors.

From these factors, age, marital status, residence, educational level, alcohol consumption, animal source food consumption and physical activity were factors that were significantly associated with hypertension (Table 5).

Hypertension is dependent on modifiable and non-modifiable factors. Familial patterns of hypertension suggests genetic factor as another important non-modifiable predisposing factor and ABO blood group is one such factor. The findings of this study revealed that the ABO blood group distribution among hypertensive patients was 32.3% for blood group A, 24.5% for blood group B, 8.3% for blood group AB and 34.9% for blood group O while among control groups it was 27.6% for blood group A, 27.1% for blood group B, 6.3% for blood group AB and 39.1% for blood group O indicating higher percentage of hypertension among patient with O blood group than other blood group type but statistically it is not significant(Table 4). This result agreed with the study done in India (Ambareesha *et al.*, 2014). We can conclude that this result mean that hypertension is not related with abo blood group as a genetic predisposing factor while Nishi *et al*. (2012), reported that a significant difference was present.

This study is consistent with a research done in India (Kaur *et al*., 2016) in which ABO blood group doesn't statistically associate with hypertension. This result is also similar to study in Saudi Arabia that demonstrated blood group O is more prevalent both in control and hypertensive groups (38.1%) as compared with B (30.1%), A(26.0%) and AB (5.8%) blood group (Alanazi *et al*., 2018).

A study from India by Kaur *et al*., (2016) found that among the study population 92.6% were Rh+ve, 7.4% were Rh-ve. In addition, there was no significant association between hypertension and Rh factor ($P \geq 0.05$).This finding is similar with our study.

5.2. Socio-Demographic Factors

Increased age was identified as a major factor for hypertension in this study. Similar findings were also reported from the study conducted at different parts of Ethiopia in Hawassa University (Atkilt Esaiyas *et al*., 2018), Tigray (Alemayehu Bayray *et al*., 2018), Addis Ababa (Kassamar Angaw *et al*., 2015),Bahir Dar city (Zelalem Alamrew *et al*., 2015), Debre Markos (Mihretie Kiber *et al*., 2019), Durame town (Tsegab Paulose *et al*., 2014), Gondar city (Akilew Awoke *et al*., 2012) and Bedele town (Feyie Bonsa *et al*., 2014). Other studies have similar finding done in Netherlands (van de Vijver *et al*., 2014). Similarly, studies conducted in the United States and China reported results similar to the current study (Yoon *et al*., 2015; Gao *et al*., 2013).

This could be due to the physiological change of blood vessels as the age is increased; in which blood vessels flexibility might be lost (hardening of the arteries) as age is increased (Hall *et al*., 1980).

There was no significant gender variation in this study subjects. This finding is in line with study done in Debre markos town, northwest Ethiopia (Mihretie Kiber *et al*., 2019). On the contrary, this study is inconsistent with reports from many studies on hypertension in different parts of the world, like study done in Bahir dar town, northwest Ethiopia (Zelalem Alamrew *et al*., 2015) and in Vietnam (Van Minh *et al*., 2006). Van Minh *et al*., (2006) stated that that females are more likely for hypertension than males as females are more prone to accumulation of fat tissue than males because females have fat mass than lean mass. This variation might be due to the difference in the study area, study population and socio demographic status of the study participants.

In this study, urban residents and educational level were more likely to have HTN when compared with its counterparts, which is supported by the other large studies conducted in different countries (Ethiopia, Vietnam and Indonesia), Ghana, South Africa and Egypt as well (Ibrahim, 1996, Fikru Tesfaye *et al*., 2007). Increasing urbanization is one of the main reasons for the rise of prevalence of hypertension in Africa. The levels of hypertension are structurally higher in urban than in rural settings (Taylor *et al*., 2011). This similarity might be mainly because of contextual and behavioral factors associated with urban environments such as dietary changes and sedentary lifestyle that together form a complex system conducive for developing hypertension. Urbanization is associated with changes in dietary habits and with reduced physical activity that lead to obesity. Such changes of lifestyle and dietary habits contribute to

the excess prevalence of abdominal obesity in urban areas which eventually result in the increased prevalence of hypertension.

There was an association between marital status and hypertension which was found statistically significant in multivariate logistic regression analysis. This is in line with the study done in Tigray (Alemayehu Bayray *et al*., 2018), Addis Ababa (Kassamar Angaw *et al*., 2015). The possible reason for this may be study participants who are married, divorced or widowed are more stressful and have more loads than those study participants who are single in their life. But this finding did not match with findings of study in India by (Saxena *et al.,* 2011).

In our study having hypertensive family member does not have an association with hypertension. This finding coincides with results reported from northwestern Ethiopia (Mihretie Kiber *et al*., 2019). But the study from Addis Ababa states that Respondents with a positive family history were found to be almost two and half times at more likely to have hypertension, than those without such a history, adjusted for other variable which was studied by (Perpetua, 2003) and other studies in India, Saudi Arabia, Tunisia and Kenya showed significant association between hypertensive family member and hypertension. This discrepancy may be the small sample size taken in this study.

5.3. Behavioral characteristics

In this study, Outpatient attendants those who have been drinking alcohol in the past were 2.39 times more likely to be hypertensive compared to those who have not been drinking alcohol in the past. It was similar with the studies conducted in Gonder and Other parts of Ethiopia (Fikru Tesfaye *et al*., 2008; Takele Tadesse and Henok Alemu, 2014; Kassamar Angaw *et al*., 2015).The possible reasons for this fact is that alcohol can produce Central Nervous System (CNS) imbalance (initiates both central and peripheral reactions which lead to hypertension),sympathetic out flow(causes secretion of corticotrophin releasing hormone), shifts extracellular calcium to intracellular space increase sensitivity to vasoconstriction or epinephrine. These all lead to vasoconstriction and cause hypertension (Parry *et al*., 2011; Nuwaha and Musuinguzi, 2013; Husain *et al*., 2014; Colangelo *et al*., 2015). The other reason is that alcohol consumption raises the amount of lipids in the blood stream, which can damage the arteries. This in turn leads to hardening of arteries that increases the risk of blood clots which can

raise blood pressure. Another reason might be that alcohol has high calories and sugar which can increase the risk of high blood pressure in the long term by adding to the body fat (WHO, 2018).

The odds of having hypertension that consumed animal source fat foods were, almost two times more likely than who did not consume animal source fat. This finding was in agreement with study in Debre markos, Northwest Ethiopia (Mihretie Kiber *et al*., 2019) and with the finding of (Sabour *et al*., 2016) in Iran. This was due to the fact that those foods which contain animal fat source foods have saturated fats, which lead excess accumulation of fat in the blood vessels, and it leads to atherosclerosis. Additionally, saturated fat obtained commonly found from animals, is the risk of cardiovascular disorders.

Even if coffee drinking, smoking cigarette and khat chewing were associated factors of hypertension, no such association were found in this study. This finding was inconsistent with many studies done in Ethiopia, Egypt, Tunisia, China and India. The possible reason for this deviation may be due to the problem of hiding socially unacceptable behaviors by the participant in this study.

In the present study, physical activity was significantly associated with hypertension in Dangila and Durbete Hospitals, northwest Ethiopia and this was in line with many community-based cross sectional studies done in different part of Ethiopia; Jimma (Feyie Bonsa *et al*., 2014), Addis Ababa (Fikru Tesfaye *et al*., 2009), Bahir dar (Zelalem Alamrew *et al*., 2015), Debre Markos (Mihretie Kiber *et al*., 2019), Gonder (Akilew Awoke *et al*., 2012), Tigray (Alemayehu Bayray *et al*., 2018). Other studies in Eritrea (Mufunda *et al.*, 2006), Egypt (Ibrahim, 1996), Cameron (Mbanya *et al*., 1998) also have the same findings. This is expected because exercise may reverse the adverse effects of lack of activity. Exercise lowers systolic/diastolic blood pressure by at least 10/5 mmHg. Exercise not only improves blood pressure, but also attenuates other risk factors for cardiovascular complications. So exercise is a very healthy habit and patients with hypertension should consider exercising frequently (Brooks and Ferro, 2012).

6. CONCLUSION AND RECOMMENDATION

6.1. Conclusion

Age, marital status, residence, educational level, Alcoholic Habit, animal source food consumption and physical inactivity were significantly associated with hypertension in our study. But sex, blood group, Rh factor, coffee drinking, cigarette smoking, khat chewing and hypertensive family member were non-significantly associated with hypertension in this study. Hence the study leads to establish the fact that these socio-demographic factors and life style factors are important risk factors for hypertension. In health care delivery system it is important to screen people for hypertension and counsel them about lifestyle modification and risk reduction. Hence, healthy lifestyle changes like change alcoholic use, promotion of balanced diet complemented by regular physical activity should be promoted through behavior change communications to reduce the risk of hypertension.

The burden of risk factors for Hypertension is increasing in developing countries including Ethiopia. Simple and client-friendly intervention programs aimed at preventing and modifying the risk factors in persons prone to Hypertension should be incorporated into the healthcare system of Ethiopia to recognize them early and effectively manage them. Moreover, behavior change communication focusing on modifiable predictors of Hypertension should be preached for the general community. This study highlighted the need for further study to determine the burden and associated risk factors of Hypertension at Hospital level.

6.2. Limitation of the study

This case-control study, even if it identified the common factors of HTN, it has a number of limitations. Since it is a facility-based study, the cases and controls may not be the representative of the catchment population. As the study was carried out in a very difficult time because of the world's pandemic disease COVID-19, most of the participants are not willing for anthropometric measurements like height, weight, BMI, waist circumference and others. The other limitation during our study was, study participants are hiding socially unacceptable habits in the questionnaire.

6.3. Recommendations

From the findings of this study, it was recommended that:

- ✓ Policy makers should make this chronic illness a part of the public health agenda and they should plan timely interventions.
- ✓ Intervention measures at the community level should be undertaken using health education and other measures by providing an emphasis on the prevention, early detection, and treatment of hypertension.
- ✓ A broader population-based survey is needed to adequately assess the national prevalence and burden of hypertensions as well as explore the interactions of the various risk factors observed in this study.
- ✓ Health risk behaviors such as drinking alcohol, smoking cigarette, chewing khat should be avoided by the society to stay healthy from risk of hypertension.
- ✓ Furthermore, researchers and health care providers should work to uncover the burden of hypertension.

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8. APPENDICES

Appendix 1: Subject Information sheet (English version)

Principal Investigator: Abebe Kindie

Bahir Dar University

College of science

Department of Biology (Genetics)

Dear participant! Here, I the undersigned is MSc student at Bahir Dar University, College of Science, Department of Biology (Genetics). Currently I will be undertaking research on a topic entitled as Association of blood groups and other factors with hypertension In Dangila and Durbete hospitals, Northwest Ethiopia.For this study, you will be selected as a participant and before getting your consent, you need to know all necessary information related to the study which will be detailed as follows.

Introduction

Privacy is the state of being free from intrusion, and in the context of health care it concerns the responsibility of a care provider to protect a clients from any disclosure (i.e., discovery by others), even unintentional, of personal health data, by providing security to the patient and the patient's records. Confidentiality, in contrast, is the limiting of information to only those for whom it is appropriate. Therefore this information sheet briefly provides the necessary guide to be considered during the study.

Objective: - The main objective of this study is to assess the association between ABO blood groups and other factors with the risk of hypertension in Dangila and durbete primary hospital.

Participants to be included: all hypertensive and non-hypertensive people visiting the hospital who are volunteer to participate are sampled by simple random sampling will be included in the study.

Risks and discomfort: Participant in this research will not cause more discomfort and no need of extra sample other than sample taken for diagnostic purpose. But, there could be minor pain and challenge in your finger during the blood drawing. The amount of blood taken from each volunteer throughout the study period is 0.5ml which will not affect your health. There is no major risk in participating in this research, as the whole procedure is carried out by health professionals following the standard good clinical practice.

Benefits:-The immediate benefit in participating in this study is you will have the chance to know your blood group and the healthy study groups are able to know whether they hypertensive or not. In addition you are able to know the risk factors of hypertension.

Incentive:-There is no financial or material incentive in participating in this study.

Confidentiality: The information that we will collect from this research project will be kept confidential. Information about you that will be collected from the study will be stored in a file, which will not have your name on it, but a code number assigned to it. Which number belongs to which name will be kept under lock and key, and it will not be revealed to anyone except the principal investigator.

Participant Rights

Your participation is entirely voluntary and up to you to decide. There is no penalty if you do not agree to participate. Also you have the right not to answer any questions you do not want to. You may also withdraw from the study at any time. If in the middle you decide to stop filling questions and no longer participate, you can stop without worry.

Persons to contact:

If you have additional questions about the study, you can contact:

Abebe kindie (Investigator) phone: 09 21 28 07 20 Email: aberealman19@gmail.com

Dr. Ziyin Mihretie(Advisor) phone: 09 11 47 89 72 Email: ziyinm@gmail.com

Thank you for your cooperation.

Appendix 2: Consent form (English Version)

Department of Biology, School of graduate studies, College of Sciences, Bahir Dar University, Consent form for the participation of the study participants in the research.

Code number……

I have clearly been informed about the research project that aims to assess the association between ABO blood groups and other factors with the risk of hypertension.I have understood that participation in this study is entirely voluntarily. I have been told that my answers to the questions will not be given to anyone else and no reports of this study ever identify me in any way. I have also been informed that my participation or non-participation or my refusal to answer questions will have no effect on me. I understood that participation in this study does not involve risks. I understood that Abebe Kindie is the contact person if I have questions about the study or about my rights as a study participant, I will be contacting the principal investigator.

Respondent's signature_____

Interviewer name_____ signature _____ date_____

Data collection sheet for all study subjects:

Appendix 3: English version of Questionnaire for study participants

Date of data collection ----------

Study site----------

Sample code / patient ID ----------

Clinical data

1. Blood pressure ---------mmHg
2. Blood group type A. A B. B C. AB D. O
3. Rh factor A. Positive B. Negative

Socio-demographic data

1. Sex ----------
2. Age ----------
3. Residence A. urban B. Rural
4. Marital states A. single B. married C. divorced D. widow
5. Educational status:
   1. Cannot read and write 4. Grade 9 to 12
   2. Only read and write 5. Diploma and above
   3. Grade 1 to 8
6. Is there a person who has hypertension in your family?
   1/ yes 2/ no
7. If your answer for question 6 is yes. Specify your family name
   1/ mother 2/ father 3/ sister 4/ brother 5/ other----------

Behavioral characteristics data

1. Do you drink coffee?
   1/ past drinker 2/ current drinker 3/ never drink
2. If the answer for question 1 is 1 and 2, how often?
   A. 3 times a day C. 3 times a week
   B. Once a day D. once a week
3. Do you drink alcohol?

1/ past drinker 2/ current drinker 3/ never drink

4. If the answer for question number 3 is 1 and 2, how frequently?

   1/ daily 2/ weekly 3/ sometimes 4/ other (specify) ----------

5. Do you smoke cigarette?

   1/ past smoker 2/ current smoker 3/ never smoke

6. If your answer for question number 5 is 1 and 2, in what amount?

   1/ three times per week 4/ twice a day

   2/ Once per month 5/ other----------

   3/ once a day

7. Do you chew khat?

   1/ past chewer 2/ current chewer 3/ never chew

8. If the answer for question number 7 is 1 and 2, how frequently?

   1/ once a week 2/ twice a week 3/ once a day

   4/ five times a week 5/ other----------

9. Have you been exercising regular physical activity?

   1/ Yes 2/No

10. If the answer for question number 9 is yes, what type of exercise and how long?

    1/ walking 3/Jogging

    2/ running 4/swimming 5/ other (specify) ----------

11. Do you eat animal source food?

    1/ yes, I was eating in the past

    2/ yes, currently I eat

    3/ never eat

12. If your answer for question 13 is 1 and 2, how frequent?

    1/ daily 2/ three times a week 3/ once a week 4/ once a month 5/ other-----

Appendix 4: Subject Information sheet (Amharic version):

የተሳታፊዎች የፈቃደኝነትና መተማመኛ መረጃ መስጫ ቅፅ

በባህር ዳር ዩኒቨርስቲ ሳይንስ ኮሌጅ የባዮሎጂ ትምሕርት ክፍል፡

እኔ አበበ ክንዴ በባህር ዳር ዩኒቨርስቲ ሳይንስ ኮሌጅ የባዮሎጂ ትምሕርት ክፍል የድህረ ምረቃ ተማሪ ስሆን የመመረቂያ ጽሁፌን በዳንግላ እና በዱርቤቱ የመጀመሪያ ደረጃ ሆስፒታልየደም ግፊት በሽታ ከደም አይነት ጋር እና የደም ግፊት እንዲፈጠር ተፅዕኖ ሊያደርጉ ከሚችሉ ምክንያቶች ጋር ሊኖራቸዉ የሚችለው ተዛምዶ በሚል ርዕሰ-ጉዳይ በመስራት ላይ ነኝ፡፡ለዚህ ጥናት ደግሞ እርስዎ የተመረጡ ስለሆነ ከዚህ ቀጥሎ የሚገኘዉን መረጃ አንብበዉ በጥናቱ ላይ መስማማትዎን ወይም አለመስማማትዎን እንዲያረጋግጡ በትህትና እጠይቃለሁ፡፡

መግቢያ፡- ጥናቱ ከእርሶ የሚወስዳቸዉ ማንኛዉም መረጃወች ሚስጥራዊነት ሙሉ በሙሉ የተጠበቀ ሲሆን እርሶ በጥናቱ አለመሳተፍም ሆነ በማንኛዉም ሰዓት ተሳትፎዎን ማቋረጥ ይችላሉ፡፡

ይህ የጥናት መጠይቅ ጠቀሜታው ከእርሶ አልፎ ለሀገሪቱ ከፍተኛ ግብዓት ሆኖ የሚያገለግል ሲሆን የእርሶ ግልፅ ተሳትፎ ወሳኝነት አለው፡፡በዚህ ተሳትፎፍ ቃደኛ ቢሆኑ ደስ ይለኛል፡፡ ፍላጎትዎ ካልሆነ ያለመሳተፍ ሙሉ መብት አለህ/አለሽ፡፡

<u>የዚህጥናትዓላማ</u>

- **የዚህ ሆስፒታል የደም ግፊት ታማሚዎች ሊኖራቸዉ የሚችለዉን የደም ወገን መለየት፡፡**
- **የደም ግፊት እንዲፈጠር ተፅዕኖ ሊያደርጉ የሚችሉ ምክንያቶች ማወቅ፡፡**

<u>ይህም ጥናት ሁለት ክፍል አለው</u>

- **የተወሰነ የደም ግፊት እንዲፈጠር ተፅዕኖ ሊያደርጉ የሚችሉ ምክንያቶችን በመጠይቆችን መጠየቅ**
- **በባለሙያ የደም ወገን ዓይነትን ፣ክብደትን፣ ቁመትን መለካት**

በጥናቱ ለመሳተፍ ፍቃደኛ ሲሆኑ ለናሙና ይሆን ዘንድ 0.5 ሚሊ ሊትር ደም በሆስፒታሉ የጤና ባለሙያዎች አማካኝነት የሚሰጡ ሲሆን ናሙና በሚሰጡበት ጊዜ ሁልጊዜ ለምርመራ ከሚሰጡት የተለየ ህመም እና አለመመቸት የለዉም፡፡

ከጥናቱ ጋር በተያያዘ ጥያቄ ቢኖርዎ ወይም ችግር ቢያጋጥሞ እነዚህን አድራሻዎች መጠቀም ይቻላል፡፡

አበበ ክንዴ(ጥናቱን የሚሰራዉ) ስልክ: 09 21 28 07 20

Email: aberealman19@gmail.com

ዶ/ር ዝይን ምህረቱ (አማካሪ) ስልክ፡ 09 11 47 89 72

Email: ziyinm@gmail.com

በጥናቱ ለመሳተፍ ፈቃደኛ ከሆኑ እባክዎ ከዚህ ቀጥሎ ባለዉ የስምምነት ቅጽ ላይ በመፈረም ይተባበሩ፡፡

እናመሰግናለን!!!

Appendix 5: Informed consent (Amharic version)

የተሳታፊዎች ስምምነት ማረጋገጫ ቅጽ

የሚስጥር ቁጥር ----------

እኔ ---------- የደም ግፊት በሽታ ከደም አይነት ጋር እና የደም ግፊት እንዲፈጠር ተፅዕኖ ሊያደርጉ የሚችሉ ምክንያቶች ጋር ሊኖራቸዉ የሚችለው ተዛምዶ ለማወቅ ስለሚሰራዉ ጥናት እና ለጥናቱ በሚኖረኝ ተሳትፎ ዙሪያ በቂ መረጃ ያገኘሁ ሲሆን በጥናቱ ለመሳተፍ ያለኝን ፈቃደኝነት በፊርማዬ አረጋግጣለሁ፡፡

የተሳታፊ ፊርማ ---------- ቀን ------- ----- ------

Appendix 6: የጥናቱ ተሳታፊ ግለሰቦች መረጃ መሰብሰቢያ ቅፅ፡

መረጃው የተሰበሰበበት ቀን ----------
የጥናቱ ቦታ----------
የበሽተኛው መለያ ቁጥር----------

<u>የህክምና መረጃዎች</u>

1. የደም ግፊት መጠን -------ሚሊ. ሜትር ሜርኩሪ
2. የደም አይነት ሀ,A ለ, B ሐ, AB መ, O
3. አር-ኤች አይነት ሀ አዎንታዊ ለ አሉታዊ

የማህበራዊ እና የስነ-ህዝብ መረጃዎች

1. ፆታ ሀ, ሴት ለ, ወንድ
2. እድሜ----------
3. መኖሪያ ቦታ ሀ, ከተማ ለ, ገጠር
4. የጋብቻ ሁኔታ ሀ,ያላገባ ለ, ያገባ ሐ,የፈታ/ች መ. የሞተበ/ባት
5. የትህምርት ደረጃ
    1. ማንበብ ና መፃፍ የማይችል/ትችል
    2. ማንበብና መፃፍ ብቻ የሚችል/ትችል
    3. 1ኛ ክፍል- 8 ኛ ክፍል የተማረ/ች
    4. 9 ኛ ክፍል- 12 ኛ ክፍል
    5. ዲፕሎማና ከዚያ በላይ
6. ከቤተሰብዎ የደም ግፊት ታማሚ አለ?
   1/ አዎ 2/ አይ
7. ለጥያቄ ቁጥር 6 መልስዎት አዎ ከሆነ, ያለዎትን ዝምድና ይግለፁ፡፡
   1/ እናት 2/ አባት 3/ እህት 4/ ወንድም 5/ ሌላ (ይገለፅ)---------

<u>ከአኗኗር ዘዴ ጋር የተገናኙ መረጃዎች</u>

1. እርሶ ቡና ይጠጣሉ?
   1/ በፊት እጠጣ ነበር 2/ አሁን እጠጣለሁ 3/ በጭራሽ አልጠጣም
2. ለጥያቄቁጥር 1 መልስዎት 1 እና 2 ከሆነ, ምን ያህል?
   ሀ. ቢያንስ በቀን ሶስቴ ሐ. በሳምንት ሶስቴ
   ለ. ቢያንስ በቀን አንዴ መ. በሳምንት አንዴ

3. እርስዎ የአልኮል መጠጥ ይጠጣሉ?

1/ በፊት እጠጣ ነበር 2/ አሁን እጠጣለሁ 3/ በጭራሽ አልጠጣም

4. ለጥያቄ ቁጥር 3 መልስዎት 1 እና 2 ከሆነ መቼ መቼ ነዉ የሚጠጡት?

1/ በየ ቀኑ 2/ በየ ሳምንት 3/ አንዳንዴ 4/ ሌላ(ይገለፅ)

5. እርሶ ሲጋራ ያጨሳሉ?

1/ በፊት አጨስ ነበር 2/ አሁን አጨሳለሁ 3/ በጭራሽ አላጨስም

6. ለጥያቄ ቁጥር 5 መልስዎት 1 እና 2 ከሆነ መቼ መቼ ና በምን መጠን ያጨሳሉ?

1/ በሳምንት ሶስቴ 4/ በቀን ሁለት ጊዜ

2/ በወር አንዴ 5/ ሌላ (ይገለፅ)----------

3/ በቀን አንዴ

7. እርሶ ጫት ይቅማሉ?

1/ በፊት እቅም ነበር 2/ አሁን እቅማለሁ 3/ በጭራሽ አልቀምም

8. ለጥያቄ ቁጥር 7 መልስዎት 1 እና 2 ከሆነ መቼ ና ምን ያህል ይቅማሉ?

1/ በሳምንት አንዴ4/ በሳምንት አምስት ጊዜ

2/ በሳምንት ሁለት ጊዜ 5/ ሌላ (ይገለፅ)----------

3/ በቀን አንዴ

9. እርሶ ያልተቋረጠ የአካል እንቅስቃሴ ያደርጋሉ?

1/ አዎ 2/ አይ

10.ለጥያቄ ቁጥር 9 መልስዎት አዎ ከሆነ ምን ዓይነት የአካል እንቅስቃሴ ነዉ የሚያደርጉት፣ለምን ያህል ጊዜ?

1/ በእግር መራመድ 2/ መሮጥ 3/ ዱብዱብ ማለት 4/ መዋኘት 5/ ሌላ--------

11.የእንስሳት ተዋፅኦ ምርት የሆኑ ምግቦችን ይመገባሉ?

1/ በፊት እበላ ነበር 2/ አሁን እበላለሁ 3/ በጭራሽ በልቸ አላዉቅም

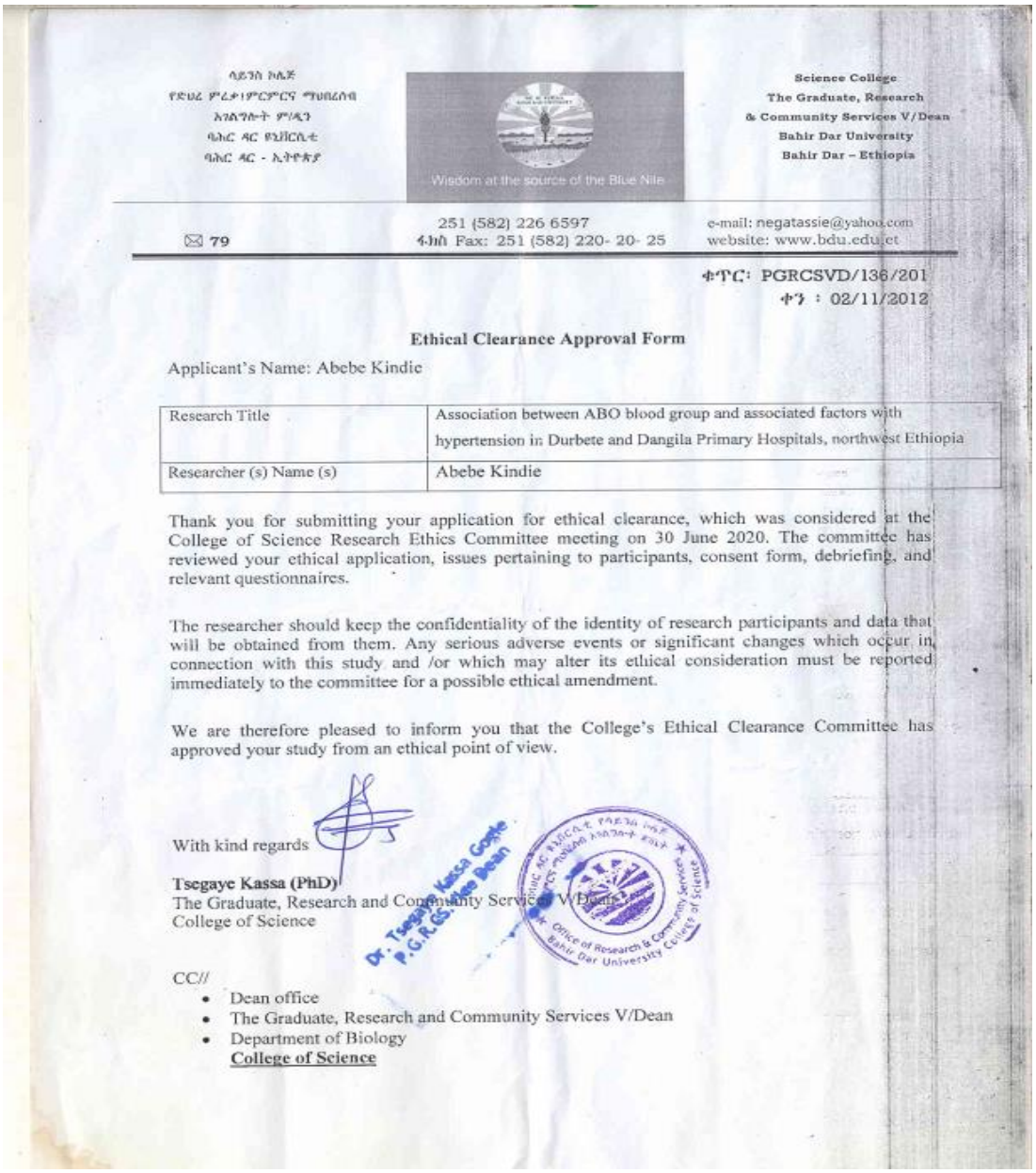
12.ለጥያቄ ቁጥር 13 መልስዎት 1 እና 2 ከሆነ, ምን ያህል?

ሀ. በየቀኑ መ. በወር አንዴ

ለ. በሳምንት ሶስቴ ሠ. ሌላ----------

ሐ. በሳምንት አንዴ

Appendix 7: Ethical Clearance Approval Form



ሳይንስ ኮሌጅ
የድህረ ምረቃ ምርምርና ማህበረሰብ
አገልግሎት ምክትል
ባሕር ዳር ዩኒቨርሲቲ
ባሕር ዳር - ኢትዮጵያ

Science College
The Graduate, Research
& Community Services V/Dean
Bahir Dar University
Bahir Dar – Ethiopia

Wisdom at the source of the Blue Nile

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e-mail: negatassie@yahoo.com
website: www.bdu.edu.et

ቁጥር: PGRCSVD/136/201
ቀን : 02/11/2012

Ethical Clearance Approval Form

Applicant's Name: Abebe Kindie

Research Title	Association between ABO blood group and associated factors with hypertension in Durbete and Dangila Primary Hospitals, northwest Ethiopia
Researcher (s) Name (s)	Abebe Kindie

Thank you for submitting your application for ethical clearance, which was considered at the College of Science Research Ethics Committee meeting on 30 June 2020. The committee has reviewed your ethical application, issues pertaining to participants, consent form, debriefing, and relevant questionnaires.

The researcher should keep the confidentiality of the identity of research participants and data that will be obtained from them. Any serious adverse events or significant changes which occur in connection with this study and /or which may alter its ethical consideration must be reported immediately to the committee for a possible ethical amendment.

We are therefore pleased to inform you that the College's Ethical Clearance Committee has approved your study from an ethical point of view.

With kind regards

Tsegaye Kassa (PhD)
The Graduate, Research and Community Services V/Dean
College of Science

Dr. Tsegaye Kassa Gashe
P.G.R.C.S. Vice Dean

Office of Research & Community Service
Bahir Dar University College of Science

CC//
- Dean office
- The Graduate, Research and Community Services V/Dean
- Department of Biology
  College of Science

Appendix 8: Sample Photos during Data Collection

ABO/Rh blood aggulitination

Disposing blood sample after identification.

Blood pressure measuring

Blood sample taking