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United Arab Emirates University

College of Science

Department of Biology

STUDY THE QUALITY OF GROUNDWATER OF AL-ZOROUP AREA IN MAHDAH STATE, THE SULTANATE OF OMAN

Fahad Khalifa Rashid Al-Shidi

This thesis is submitted in partial fulfillment of the requirements for the degree of Master of Science in Environmental Sciences

Under the Supervision of Dr. Ruwaya Alkendi

November 2014

Declaration of Original Work

I, Fahad Khalifa Rashid Al-Shidi, the undersigned, a graduate student at the United Arab Emirates University (UAEU), and the author of this thesis, entitled "*Study the Quality of Groundwater of Al-Zoroup Area in Mahdah State, the Sultanate of Oman*", hereby, solemnly declare that this thesis is an original research work which has been done and prepared by me under the supervision of Dr. Ruwaya AlKendi, in the College of Science at the UAEU. This work has not been previously formed as the basis for the award of any academic degree, diploma or similar title at this or any other university. The materials borrowed from other sources and included in my thesis have been properly cited and acknowledged.

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Abstract

The groundwater is the most important source of water in the Sultanate of Oman. Accordingly, the Oman Government has paid great attention commensurate with the importance of water resource in terms of their sources, quality, and ways to rationalize consumption to remain under the term of sustainability and validity of present and future generations. This study was conducted in Al Buraymi at Mahdah State in Al-Zoroub area targete a groundwater quality for 20 wells in private farms. The water samples were taken from these wells and subjected to analysis for microbial and geochemical aspects. For this analysis, the assorted devices and instruments were implemented. Additionally, to focus on the possible sources of pollution and the range of competence for the human user or livestock and agriculture of this water, the international guideline measures were used for compression.

Based on investigations from a group of chemical and biological analysis; it turns out that this water contained varying proportions of salts and dissolved minerals. These results also build on the proportions of dissolved salts which showed different types of water in these wells which are mostly fresh (magnesium bicarbonate) in ten wells, brackish (magnesium chloride) in six wells and saline (sodium chloride) in four wells.

The dominant water type (magnesium bicarbonate) resulted from the fresh water in the upper aquifer, which recharged from northeastern catchment areas of Mahadah, Wadi Al Jizi, and Wadi Hamad. Moreover, it was found that the abundance of the major cation ions in groundwater is in the following order: $Na^+ > Mg^{2+} > Ca^{2+} > K^+$, where as the abundance of the major anion ions in groundwater is in the following order: $Cl^- > HCO_3^- > SO_4^- > NO_{3.}^-$ These distribution is ffected by salinization phenomena (ion exchange), mixing of saline water in the deep aquifer with fresh water and by anthropogenic sources.

The building at the expense of sodium adsorption ratio (SAR) found that 65% of the wells are located within the category of hazardous water at least for sodium ion and medium salinity rate. So they are safe for agricultural use except for well no (8) located in the category of high salinity.

On the other hand, the results showed microbial water containing high levels of coliform bacteria and *E.coli* species in nine wells. Compared to WHO guidelines, not all 20 wells that I have investigated are suitable for human use without treatment, and 70% of them are not suitable for animal use, but in contrast, all wells are suitable for agricultural use.

During the term of microbial source tracking method (MST) that applied on 9 *Escherichia coli* (*E.coli*) samples used 16SrRNA, which blasted in the NCBI; sequences of these samples were identified by comparison with the GenBank database used BLAST searches. It was found that all isolated *E.coli* were homologous with isolated single *E coli* species (*E.coli* K-12 substr - MG 1655 strain) which first identified in the faeces of human. According to filed survey and investigation, the main cause of *E. Cole* in the groundwater is may be septic tank and animal manure, which is very close to these wells, or the well heads are not completely sealed, which would allow contaminants to enter the well.

Keywords: Groundwater, water quality, hydrogeochemical properties, microbial profile, *E.coli*, Al-Zoroub area, Sultanate of Oman

Title and Abstract in Arabic

دراسة جودة المياه الجوفية في منطقة الزروب في ولاية محضة، سلطنة عمان

الخلاصة

تعد المياه الجوفية من أهم مصادر المياه في سلطنة عمان إن لم تكن أهمها على الإطلاق، فهي الركيزة الرئيسة لجميع الاستخدامات الحياتية سواء الاستهلاك الآدمي المباشر أو في الأنشطة الزراعية والحيوانية. و بناء على ذلك، فقد أولت حكومة السلطنة اهتماما كبيرا بالثروة المائية بما يتناسب وهذه الأهمية من حيث مصادرها، وجودتها، وطرق ترشيد استهلاكها لتبقى تحت مظلة الاستدامة والصلاحية للأجيال الحاضرة والمستقبلة. وبناء على ذلك، أجريت هذه الدراسة في محافظة البريمي بولاية محضة في منطقة الزروب مستهدفة جودة المياه الجوفية لعدد ٢٠ بئرا في مزارع خاصة للمواطنين. حيث أخذت عينات المياه من هذه الآبار وأخضعت للتحليل والدراسة من نواحي كيميائية و أحيائية (كائنات دقيقة) . ولتحقيق ذلك استخدمت أجهزة وأدوات متنوعة لتسليط الضوء على المصادر المحتملة لتلويث هذه المياه ومدى صلاحيتها للاستخدام البشري، والحيواني، أو الزراعي مقارنة بمقاييس عالمية. واستنادا إلى النتائج المستخلصة من مجموعة التحاليل الكيميائية والبيولوجية، اتضح أن هذه المياه احتوت على نسب متفاوتة من تراكيز الأملاح والمعادن المذابة.

كما أن هذه النتائج وبناء على نسب الأملاح الذائبة أظهرت أنواع مختلفة للمياه في هذه الآبار كان أغلبها مياه مختلطة لعدد ١٠ آبار (بيكربونات الماغنيسيوم) ناتجة من تداخل المياه العذبة القادمة من شمال شرق الزروب من أودية ولاية محضة ووادي الجزي ووادي حمد مع المياه المالحة في الخزان الجوفي العميق، مع وجود مياه مالحة (كلوريد الصوديوم) لعدد ٤ آبار و ٦ آبار قليلة الملوحة (كلوريد الماغنيسيوم). كما أظهرت النتائج أن الأيونات السائدة في مياه الآبار تتخذ الترتيب التالي : (صوديوم > ماغنيسيوم > كالسيوم > بوتاسيوم) والأيونات السالبة تتخذ الترتيب التالي: (كلور > بيكربونات > كبريتات > نيترات). و بناء على حساب معامل امتصاص الصوديوم (SAR) (تبين أن٦٥% من الآبار يقع ضمن فئة المياه ذات الخطورة الأقل لأيون الصوديوم والمتوسطة لمعدل الملوحة لذلك فهي آمنة للاستخدام الزراعي باستثناء بئر واحد يقع في فئة الملوحة العالية. أما من حيث التحاليل الميكروبية ومقارنة بمقياس منظمة الصحة العالمية فقد أظهرت النتائج احتواء المياه على نسب عالية من بكتيريا الكوليفورم

(Coliform) وظهور بكتيريا الإيكولاي (*E. coli*) في تسع عينات. و بذلك، فإن جميع الآبار (١٠٠%) لا تصلح للاستخدام البشري كمياه شرب مباشرة بدون معالجة، كما أن نسبة ٧٠% منها لا تصلح للاستخدام الحيواني ولكنها من جانب آخر تصلح جميعها للاستخدام الزراعي. اما من حيث عملية تتبع مصادر وجود بكتيريا الإيكولاي *(E. coli)* في تسع عينات باستخدام تقنية (MST) 16SrRNA– فإن النتائج المستخلصة من البيانات العالمية (NCBI) المسجلة أظهرت أن هذه البكتيريا تتشابه بنسبة ١٠٠% مع بكتيريا (*E. coli* K–12) التي تم عزلها من مصدر بشري وبذلك يحتمل أن يكون مصدر تلوث الآبار المحتوية على الإيكولاي *(E. coli)* هو مصدر بشري أو حيواني بسبب وجود خزانات التصريف وأسمدة الحيوانات بقرب هذه الآبار، كما أن بعض هذه الآبار غير محكمة الإغلاق ربما تسمح بدخول الملوثات مباشرة إلى فتحة البئر.

كلمات مفتاحية: مياه جوفية، جودة المياه، عناصر كيمائية، أحياء دقيقة، بكتيريا كولاي، منطقة الزروب، سلطنة عمان.

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Dedication

To those pure spirits which are derived from pure memory of hope, strength and inspiration and from their mentions in my prayer got a challenge of the difficulties: my Compassionate Mother, my dear father, and my lovely son (Faham). I hope Allah covers them by his mercy and His pleasure. Then, to my dear wife, my sons, daughter, my brothers and sisters who joined this journey with me to complete this work.

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List of Abbreviations and Acronyms

µS/cm	Micro Siemens per centimeter
BST	Bacterial Source Tracking
cfu	Colony Forming Unit
dNTPs	Deoxynucleotide Triphosphates
dS/m	deci Siemens per meter
EC	Electrical Conductivity
EDTA	Ethylene Diamine Tetra acetic Acid
EMB	Eosin-Methylene Blue
EXO-SAP	Exonuclease Shrimp Alkaline Phosphatase
FIB	Faecal Indicator Bacteria
FISH	Fluorescent in Situ Hybridization
GW	Groundwater
Hi Di	Highly Deionized
LDM	Library-Dependent Methods
Mg/l	Milligram per liter
mM	MilliMolar
MRMWR	Ministry of Regional Municipalities, and Water Resources
MST	Microbial Source Tracking
NCBI	National Center for Biotechnology Information
PACADGMAN	Public Authority for Civil Aviation Directorate, General of Metrology & Air Navigation.
PCR	Polymerase Chain Reaction
PFGE	Pulsed-Field Gel Electrophoresis
pH	Hydrogen Ion Concentration
BLAST	Basic Local Alignment Sequencing Tool
ppm	Parts Per Million
R	Milli Equivalent Per Liter (meq/l)
R	Regression

SAR	Sodium Adsorption Ratio
TBE	Tris Borate EDTA
TDS	Total Dissolved Solids
TGE	Tryptone Glucose Extract
WHO	World Health Organization

Chapter 1 : Introduction

1.1. Overview

It is clear that increasing consumption and uses of water in different sectors have caused insufficiency of water resources, lack of balance between supply and demand, loss of quality, depletion of water resource, and absence of prompt and integrated water policies. These factores have cumulatively led to critical problem. The Middle East in general, and the Gulf Region in particular, are considered amongst the areas that suffer from scarcity of water resources, especially the countries that lie in the arid and semi-arid zones like the Sultanate of Oman.

This situation has led the concerned countries to enhance the standard of water management, quality and conservation, to look for new water resources and to implement intensive new programs and technologies in order to emphasize the importance of conservation of this precious resource (MRMWR, 2008). Accordingly, as the Sultanate has a deep-rooted history of water use in various aspects of living, it has dealt with the challenges imposed by the water situation and quality. Plans used to impose rules and perfectly implemented standard. These standards made a balance between development and quantity of efficient available water from different resources.

In this chapter, the overview of Oman topography, water management and use and various aspects of groundwater of Al-Buraymi governorate and specifically thes study area Al-Zoroup will be thoroughly discussed.

1.2. Statement of the problem

Water quality affects life activities, whether for human or animal use, as well as agricultural. Hence, the interest in the study of water quality and impact assessment of its use is very important. However, many natural factors such as weathering and dissolving of soil or rocks salts and minerals in addition to human activities direct and indirect impacts that reflecte in the quality of water and thus lead to contamination and lack of suitability for use.

Oman is one of the arid and semi-arid countries, where climate is characterized by high temperature, low rainfall, high evaporation rates and limited groundwater recharge. Hence, presence of agricultural activities and growth in the number of people led to withdrawal high quantity of groundwater. Furthermore, the using of many types of fertilizers and pesticides are which in turn linked to concentrations of pollutants in the aquifer. This particular study is conducted in the Al - Zoroup area which is situated in the Al-Buraymi governorate. The purpose of this study is to identify the quality of the groundwater in private farms both chemically and microbiologically and keep track of potential fecal contamination sources.

Chapter 2 : The Environmental and Geographical Properties

2.1. Geography and Location of Oman

The Sultanate of Oman is located in the Southeastern corner of the Arabian Peninsula. The United Arab Emirates (UAE) lies to the northwest, the Republic of Yemen to the southwest, and the Kingdom of Saudi Arabia (KSA) to the west. The climate of Oman is hot, dry in summer and mild in winter. The average temperature is 27-37 °C in Asee, 17-23 °C in Saiq on Al-Jabal Al-Akhadar, and 24-30°C in Salalah.

The main water resources are wadis flow (rainfall), groundwater, springs, desalination plants and treated wastewater (MRMWR, 2008). The mean annual rainfall is low, but could be highly variable: it can exceed 350 mm in the mountains of Northern Oman and Dhofar, but be as little as one hundred millimetres in the foothills and fifty millimetres on the coast and desert interior (Nassereldeen, 2007). Administratively, the Sultanate, is divided into eleven governorates (Figure 1) and this particular study was performed in Al-Buraymi Governorate.

2.2. Physical Setting of Al- Buraymi Governorate

The Al-Buraymi governorate is located in the northwestern area of the Sultanate, close to the border with the United Arab Emirates (UAE) and approximately three hundred kilometers away from Muscat. The provinces within the Al-Buraymi governorate are; Al-Buraymi, Mahdah, and Al- Sininah. The regional center of the governorate is the Wilayat of Al-Buraymi. The Al-Buraymi covers an area of the central and northern parts of the Oman Mountains (about 11,000 km^2) between the border with the UAE to the west and the north and coast of the Oman Sea to the east.

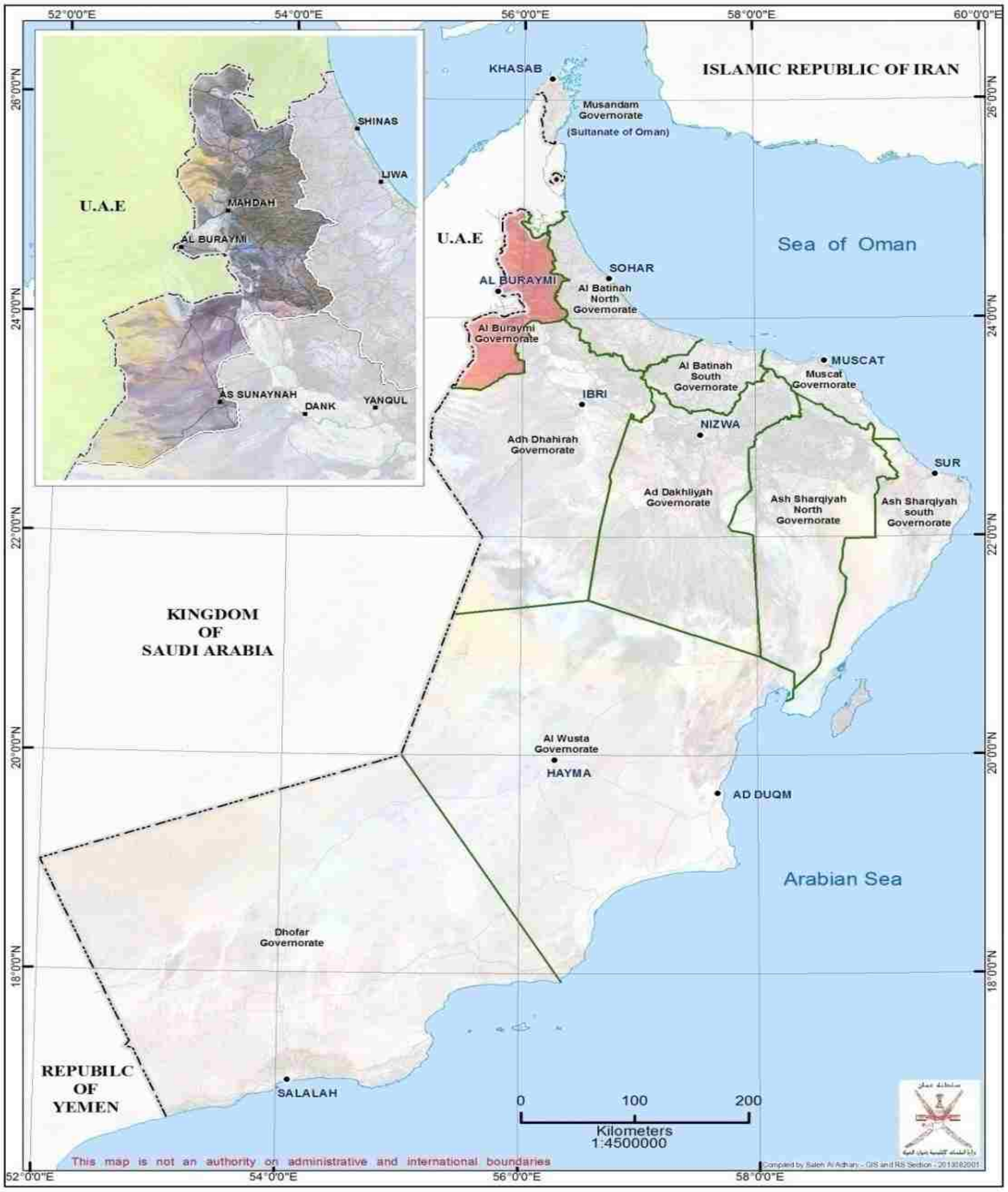


Figure 1: Map shows Oman's 11 Governorates & Al-Buraymi study area.

(Source: GIS, Remote Sensing Section office in MRMWR, 2013).

Al-Buraymi Governorate comprises of three main physiographic regions:

1) An extensive mountain range running NNW-SSE, known as Al Hajar Al Garbi. The highest peaks along the axis of the range are more than 1000 m above sea – level, and reach 1619 m in the southernmost part. The mountains cross west to east by the Wadi Hatta in the north and Wadi Al Jizi in the south.
2) An eastern piedmont zone running NNW to SSE, which forms the Al Batinah coastal plain. This plan is 10-25 km wide, slightly concave toward the northeast, and dissected by a dense network of wadis running down from the piedmont of the mountain range to the flat coastal Khabra, a cultivated and inhabited areas bordering the coast.
3) A discontinuous western piedmont zone that forms an irregular boundary with the western flank of the mountain range. Its forms are comprising of gravelly plains and sand dunes at the edge of the Rub' al Khali.

The coastal highway, along with the highways following the Wadi Hatta and Wadi Al Jizi, from the main lines of communication between the capital Muscat and settlements, such as Sohar, Liwa and Shinas (Figure 2) which lay along the Al Batinah coast, and Mahdah and Al-Buraymi which lay inland. In addition to the highways, a number of graded roads and tracks link villages that are scattered across the mountain and piedmont terrains.

Figure 2: Google Earth map shows highways linking Al-Buraymi to other cities.

2.3. Study Area

This chapter aims to discuss the characteristics of Al - Zoroup areas within its location, geology, lithology, topography, climate prosperities and other related aspects.

2.3.1. Location of Al-Zoroup Area

Al-Zoroup lays south of Mahdah city southeast of Al-Buraymi and east of the UAE border close to Khotom Ashiklla. The coordinates are 395000N and 268000 E.

Administratively, the area belongs to the city, and is of significant importance because it lies between four major highways: the main road from Albatina to Muscat (Wadi Al Jizi), the road from Al-Dahira (Wadi Saa), the road from the UAE (Khotom Ashiklah), and the road to the center of Mahdah. These road configurations ensures the continuous movement of people in and out of the Al-Zoroup area.

2.3.2. General Aspects

The main activity in the Al-Zoroup area is agriculture and many of the private farms are situated beside the highways and in the geographical interior of the area. Most farms produce palm dates, vegetables and fruit, although some areas rais cattle and goats. Each farm has a private well, from which water is pumped and collected in a cement pool (Figure 3). This water is then used to irrigate crops and gardens. Each private well is assigned an admission number, which is printed on an aluminium label fixed to the well’s wall (Figure 4)

The local population and household composition in the Al - Zoroup area are not crowded because it is located in the countryside of the Al - Buraymi city. Table.1 gives more details about the number of total houses and total population depending on National Centre for Statistical and Information data.

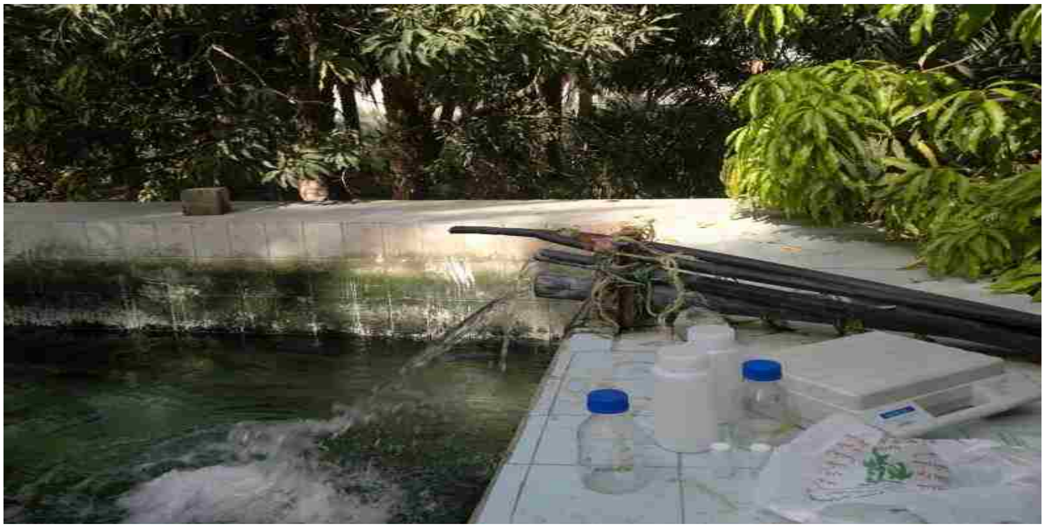


Figure 3: Cement pool used to store well water

Figure 4: Well ID printed on an aluminum label.

Table 1: The Al-Zoroup demographic profile & total population

Location	Houses		Institutional households			Population				
Al-'Zourop	Occupied	Total	Expatriate	Omani	Total	Expatriate		Omani		Total
						Female	Male	Female	Male	
	63	105	56	6	62	9	195	16	29	249

2.3.3. Physiographic Framework of Al-Zoroup Area

Al-Zoroup has a varying physiography that can be generalized as follows:

2.3.3.1 Mountainous Zone

A mountainous area composed of outcrops of hard rock comprises ophiolite, limestone, and marls. Runoff is highly efficient, resulting in marked erosion (Regional Development Committee, 1984).

2.3.3.2 Wadi deposits and catchment areas

These areas consist of stratified alluvial silts and gravels. In place, the alluvium has cemented resulting in gentle slopes that are more conductive to infiltration. The direction of runoff is roughly NE to SW. Al-Zoroup area in southern gap regions (Wadi Hamad & Al-Jizi System) as shown in (Figure 5) appears much greater catchment area than northern (Wadi Musaydirah-Kahl & Wadi Al-Wadiiyain Systems) (Regional Development, 1984).

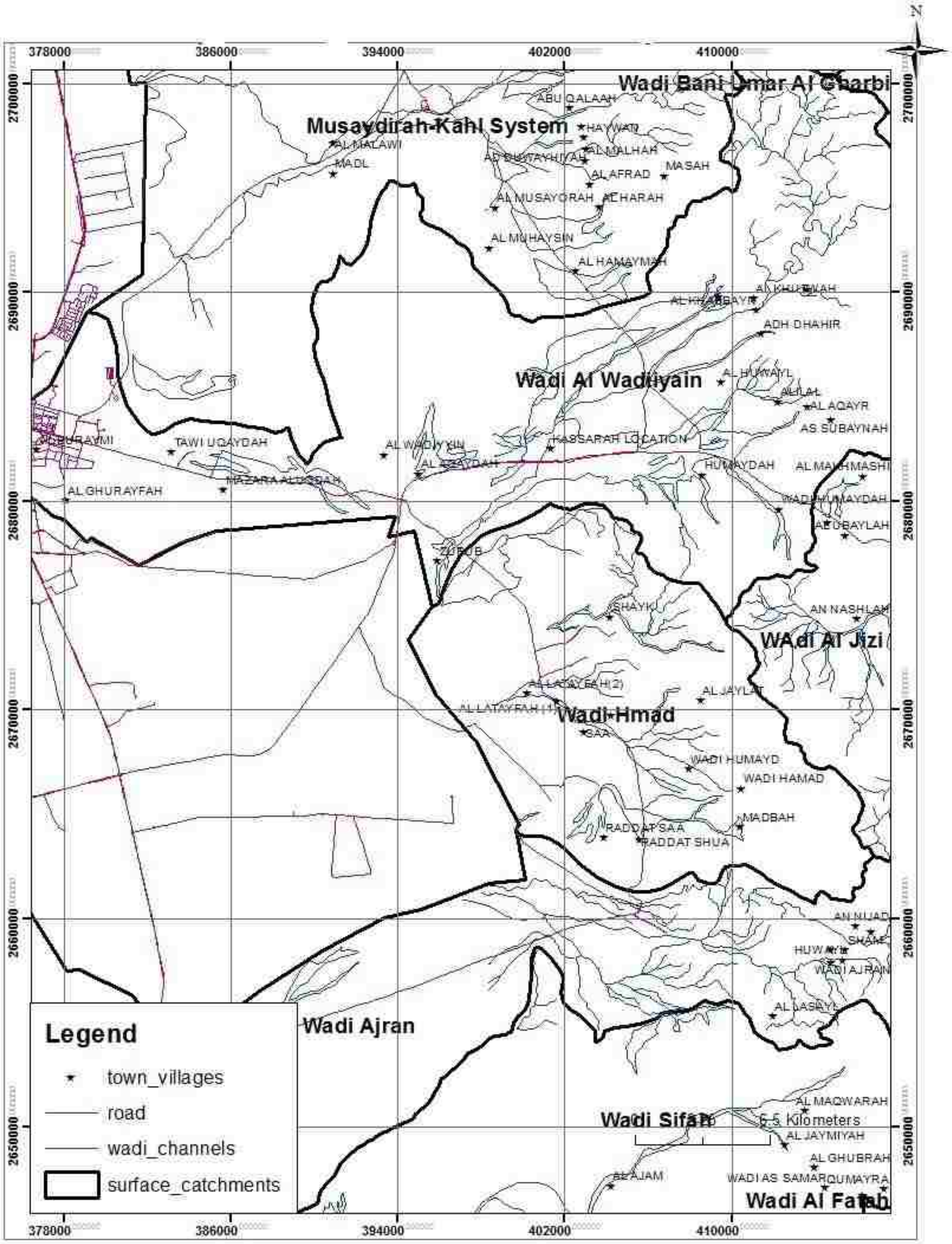


Figure 5: The surface catchments and wadi channels surrounding the Al-Zoroup area.

2.3.4. Hydrogeological and Vegetation Profile

A. Piedmont and Alluvial Fan Zones

The Al-Zoroup area within the piedmont zone contains alluvial fans, in which drainage changes from a tributary system to a distributed system that spreads over large areas and ultimately disappears into the sand dunes at the UAE border (MRMWR, 2003).

B. Historic Water Table

The water table in the study area indicted less than 260 m above sea level. The water table is measured regularly in dedicated wells or boreholes to check the position of the water table in the area and to observe the effect of climatic conditions or increase in domestic, agricultural or industrial demands for water extraction.

Historically, MRMWR monitored water levels on existing boreholes in all the catchments of the study area. Depth to water level in the study area depends on the aquifer; it fluctuates within 25m below ground level in the alluvium and up to 46m in the bedrock. Due to the large-scale groundwater pumping for Al-Buraymi town and Falaj support wells, water table declines are steeper in lower catchment whereas in the upper and middle catchment, the water table is more stable as illustrated in the (Figure 6) (MRMWR, 2003).

C. Natural Vegetation

The semi-arid climate causes natural sparse vegetation such as acacias, spiny bushes, and juju biers, which grow in the wadi beds. However, winter rain is relatively common in the mountains, which maintains the water table below the alluvial sediments in the piedmont and allows the cultivation of gardens as mentioned above (MRMWR, 2003).

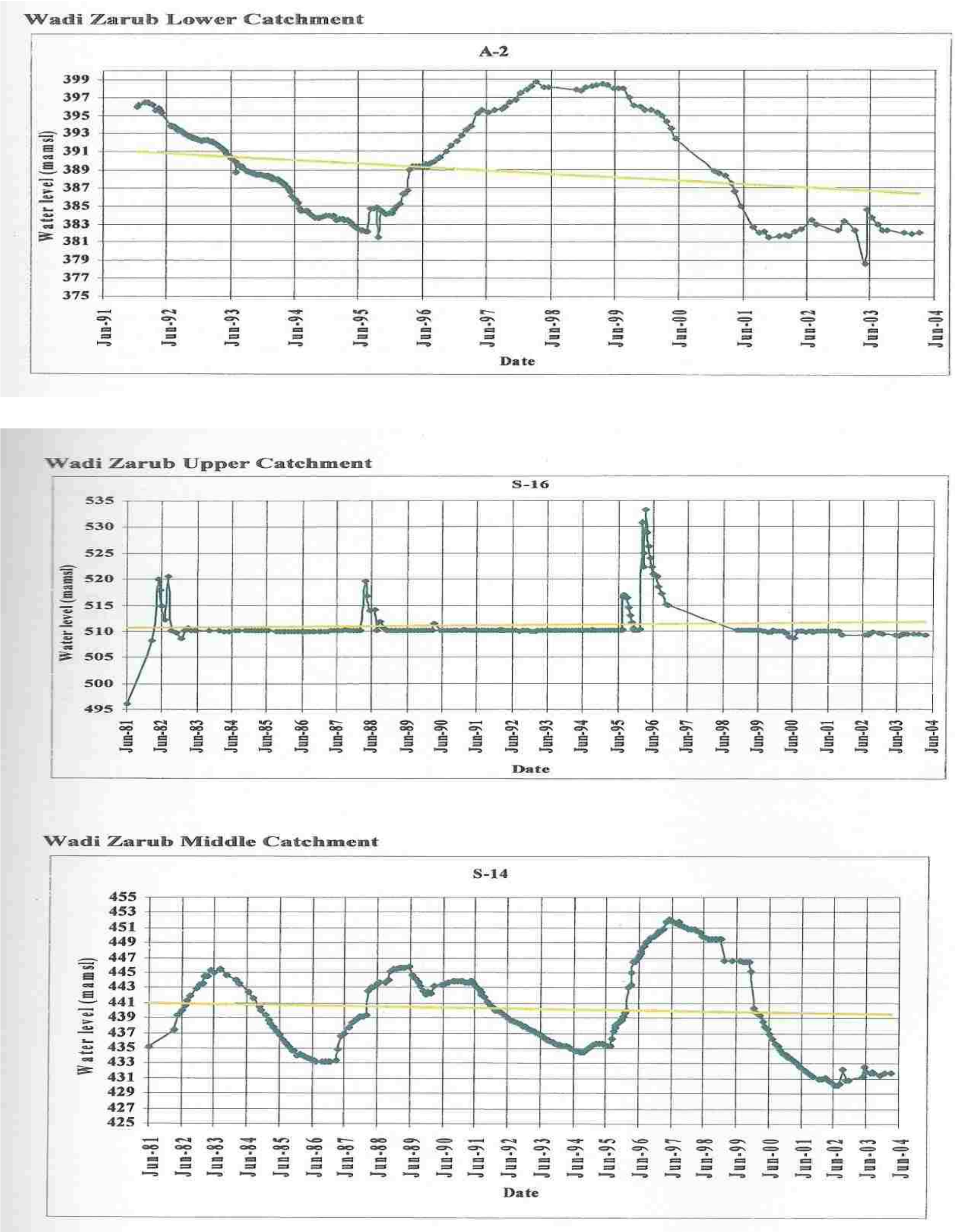


Figure 6: Water table record (Jun 1981-Jun 2004) in catchment of Al-Zoroup

2.4. Climate

2.4.1. Temperature

The air temperature data obtained from the Public Authority for Civil Aviation Directorate, General of Metrology & Air Navigation (PACADGMAN) in Al-Buraymi varies widely throughout the year. Temperatures are highest (up to 50.8°C) during the summer (May to September), whereas it is dry and cool (minimum 2.6°C) during the winter (November to March).

The annual average minimum temperature ranges from 11°C to 18.3°C, and the annual average maximum temperature ranges from 38.4°C to 41.5°C. The hottest months are June, July, August, and September, with average maximum temperatures of 47.1°C, 47.1°C, 46.6°C, and 44.2°C, respectively. By contrast, the coolest months are December, January, February, and March, with average minimum temperatures of 8.3°C, 6.3°C, 7.7°C, and 10.3°C, respectively. Table 2 shows the mean temperatures recorded by the Al-Buraymi meteorological station from 1977-2013.

Table 2: The mean temperatures of Al-Buraymi between 1977-2013.

Degree	Temperature (°C)
Maximum	50.8
Mean average maximum	41.5
Minimum	2.5
Mean average minimum	11
Mean	26.45

2.4.2. Relative Humidity

The relative humidity in the Al-Buraymi governorate is an important factor affecting hydrological conditions and groundwater levels. The increase in the relative humidity along the Al Batina coast during the summer months helps local cloud formation over the Mahdah Mountains, which provides much-needed rain. Table 3 shows the monthly, annual relative humidity levels recorded by the Al-Buraymi meteorological station from 1986 to 2013.

Table 3: The Al-Buraymi monthly, annual relative humidity levels during 1986-2013.

Month	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Mean
Relative Humidity	60.6	55.2	44.5	32.4	25.8	29	32.8	33.3	35.2	37.1	49.4	59.8	41.8%

2.4.3. Rainfall

There are two meteorological stations in the Al-Zoroup area that measure rainfall: the first meteorological station is located in the city of Al-Buraymi, and the second in Sharam near Nuway in Mahdah city. The data from these meteorological stations are being taken from MRMWR and PACADGMAN.

The data cover the period from 1986 to 2011. All measurements are in millimeters (mm). The Al-Buraimi governorate is an arid area that only receives intense rainfall in a short period, sometimes for a few hours or less. Rainfall is much higher (maximum rainfall is 200 mm per year) in the mountains (not monitored) than on the plains (MRMWR, 2003). During 1986–2011, rainfall in the Al-Zoroup area was higher in January, February, and March (the average rainfall recorded in Al-Buraymi and Mahadah during these months was 375.9 mm, 429.4 mm, and 401.5mm, respectively). However, little rain falls in the summer (although storms can occur). For example, the Al-Buraymi and Mahdah meteorological stations recorded 174mm and 117mm, respectively, of rainfall in July 1995.

The lowest average recorded from 1986 to 2011 was 6.8 mm in both cities. The mean annual rainfall for each climatic station varied widely. The Sharam gauge in Mahdah recorded zero annual rainfall in 1999, 2002, and 2003, whereas the maximum rainfall (284.5mm) was recorded in 1988. The lowest average levels (6.45mm) were recorded by Al-Buraymi station in 1997 and 2001, with the maximum level (423mm) being recorded during the rainy season in 1995. The average recordings from the two stations in the study area illustrate the variation in rainfall during 1986–2011. Overall, the records were very low, in keeping with the arid climate in the region as shown in (Figure7).

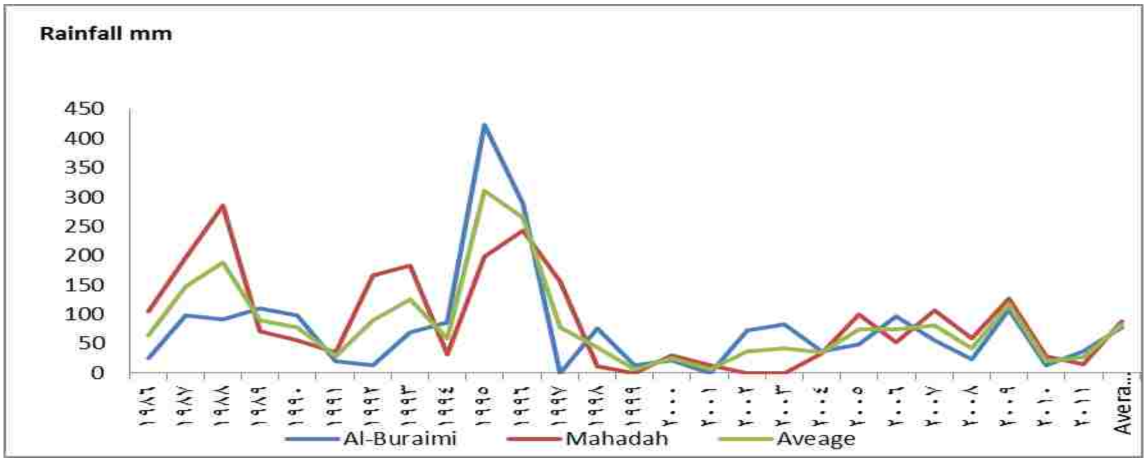


Figure 7: Average annual rainfall records in Al-Zoroup area, 1986-2011.

2.4.4. Wadis Flow

The runoff from the wadis that flow through the catchments and the volume of water discharged near the study area affect the level of water in the wells and the levels of chemicals and biological contaminants that infiltrate into the aquifer.

Two wadi gauges located around Al-Zoroup are equipped with water stage gauges. These gauges enable peak discharges by estimating the slope area. The two stations are located at Wadi Salmh Al-Ain near Al-Buraymi (site ID, CB973852AD; 393600 E, 2678200 N) and Wadi Ma'Shiq at Sharam (site ID, CC907987AD; 397800 E, 2710200 N) (MRMWR 2008).

The average annual flow recorded at Wadi Salmh from 1984 to 2011 was 135725.9526 cm^3 whereas that recorded at Wadi Sharam was 391948.1527 cm^3. The highest annual flow volume recorded by the Wadi Sharam gauges in 1996 was 11145342 cm^3 and that recorded by the Wadi Salmh gauge (2006) was 7603200 cm^3. As shown in (Figure 8) several years yielded a volume of zero. The runoff recorded by Wadi Sharam was high for 8 months in both 1996 and 1997; however, the runoff recorded by Wadi Salmh in 1996 was high for only 4 months (MRMWR, 2008).

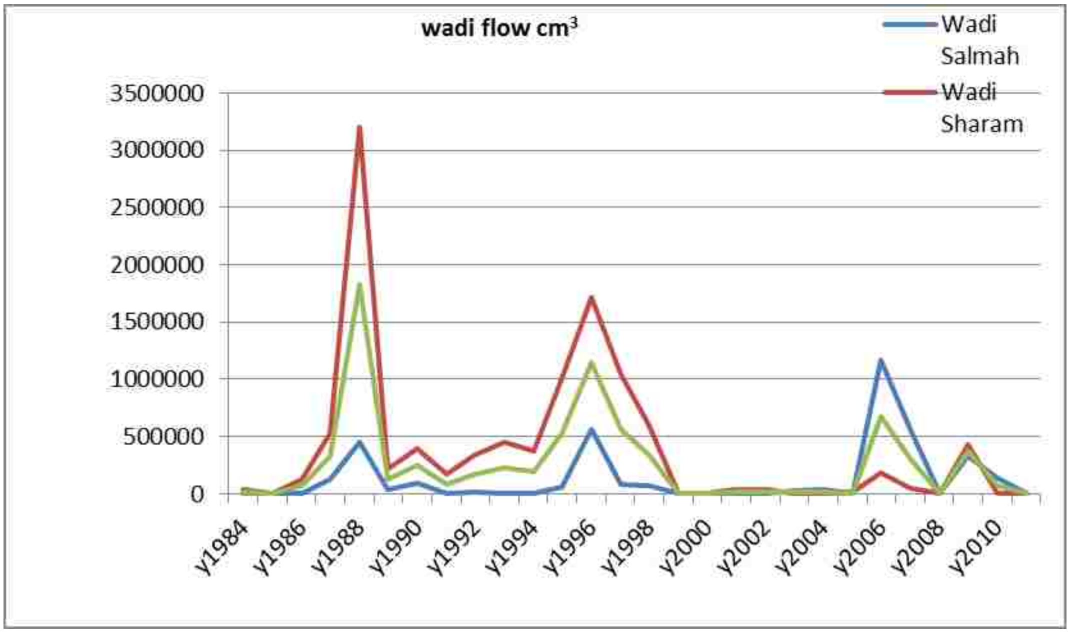


Figure 8: Total & average wadi flow records in 1984-2011, Mahadah city.

2.4.5. Geology

The geologic features of the Al - Zoroup area, illustrated in the map as shown in (Figure 9) are characterized with ophiolite in eastern side where limestones and marls are shown in the western part. The dip is roughly sub-vertical, and the strike of the strata and the general structure is NW to SE.

Tectonic activity had caused fracturing in all units. Weathering of the marls and ophiolites may generate a clay-like material (Regional Development Committee, 1984). Further to the south, the Hawasina (oceanic) nappe exposures in the piedmont zone occur mainly as broken hills running in N to S direction. They display extensive folding, faulting, and thrusting, making hydrogeological interpretation very difficult.

In addition, there are many faults in the ophiolite rocks in the main mountain range, which run NNW to SSE and ENE to WSW, and control the course of the wadis.

These faults may act as conduits for groundwater flow, as found in the Samail catchment in Northern Oman (Century Architects Consulting Engineers, 2004). The major part of the Al - Zoroup area is covered by the hard rock, which does not contain primary porosity, the alluvium that is considered the most prolific aquifer and main source of groundwater. However, the potential of the aquifer is limited due to the restricted saturated thickness, although it has a high transmissivity.

The Tertiary limestone may contain locally significant supplies of groundwater and show limited fractured characteristics. They are generally tight and elsewhere have not been shown contain water (MRMWR, 2003). The resource increases immediately after major recharge events and not cemented Wadi alluvium, associated with active Wadi channels, is typically coarse grained and allows quick recharge to the aquifer.

2.4.6. Lithology

Boreholes (depth 300 m) were drilled in the main Wadi located in the Al-Zoroup area about 800 m east of the UAE border (0393883 E, 2678968 N). The borehole summary report as appeared in (Figure 10) characterizes the lithology of the area as follows: Alluvium (0–17.5 m) overlying a clay/limestone bed to a depth of 16 m. This bed overlies a thick sequence of clay stone extending down to the total drilled depth. Minor seepages (total 0.2 L/s) were encountered at 35, 92, and 135 m (MRMWR, 2003).

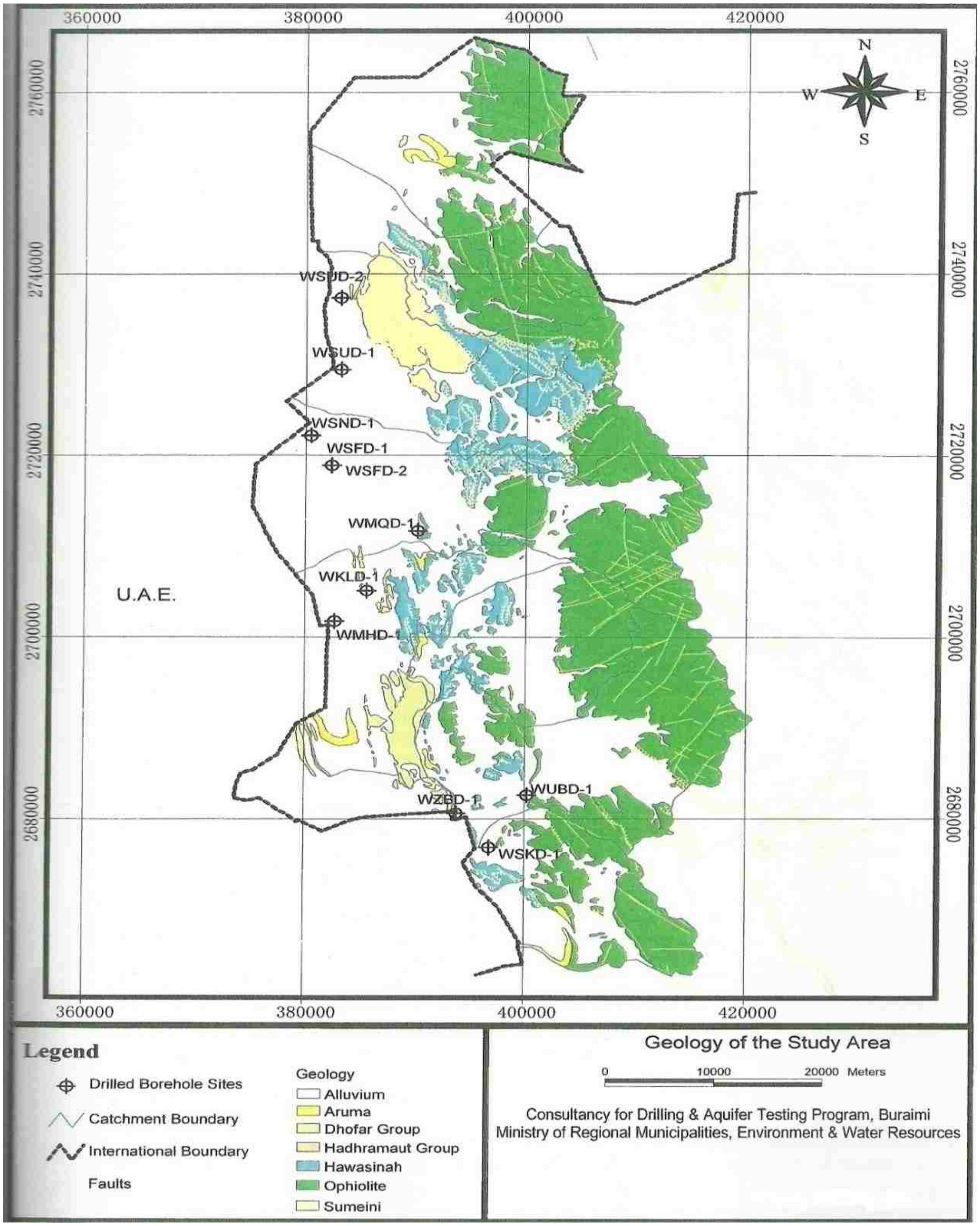
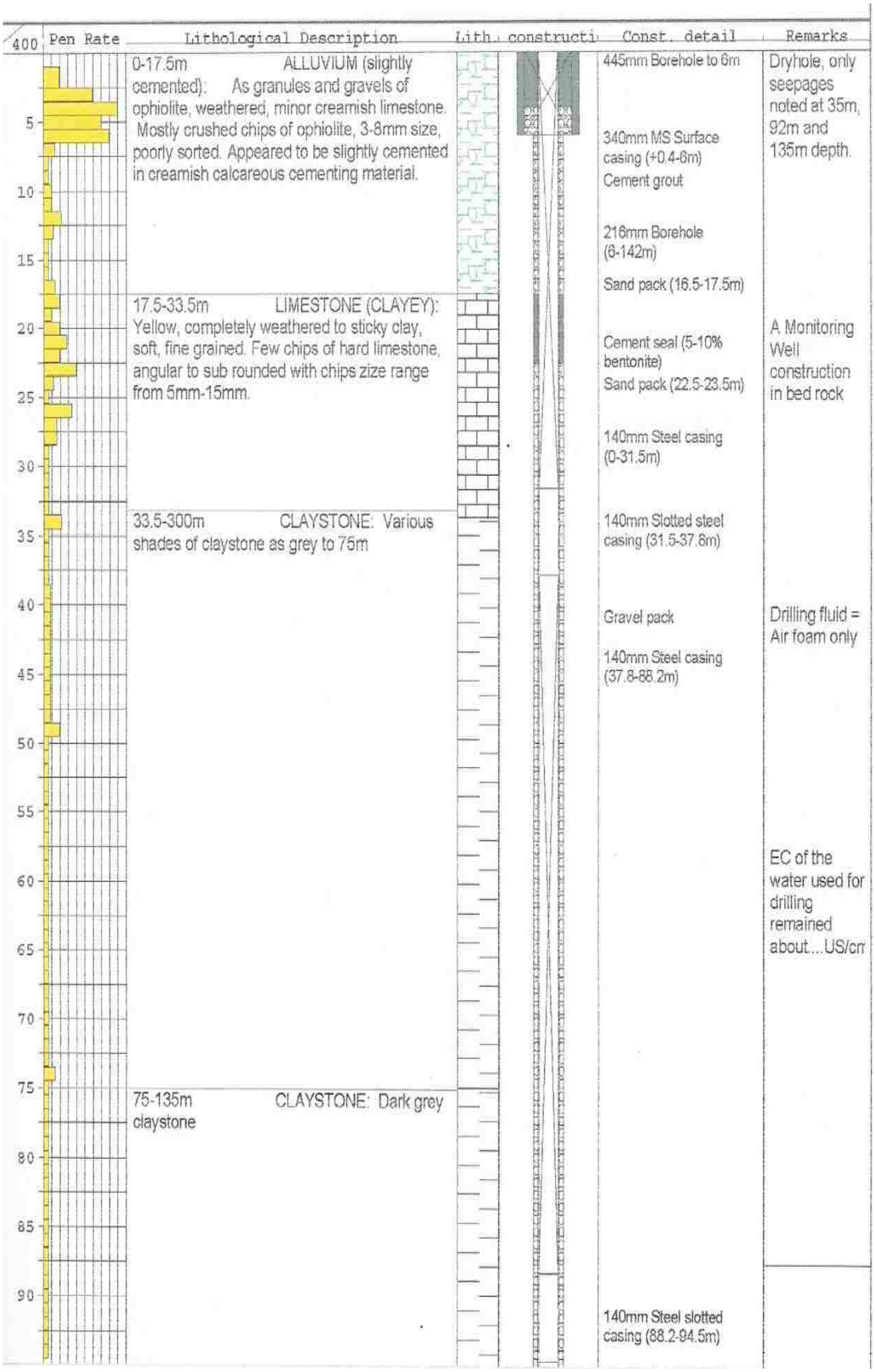
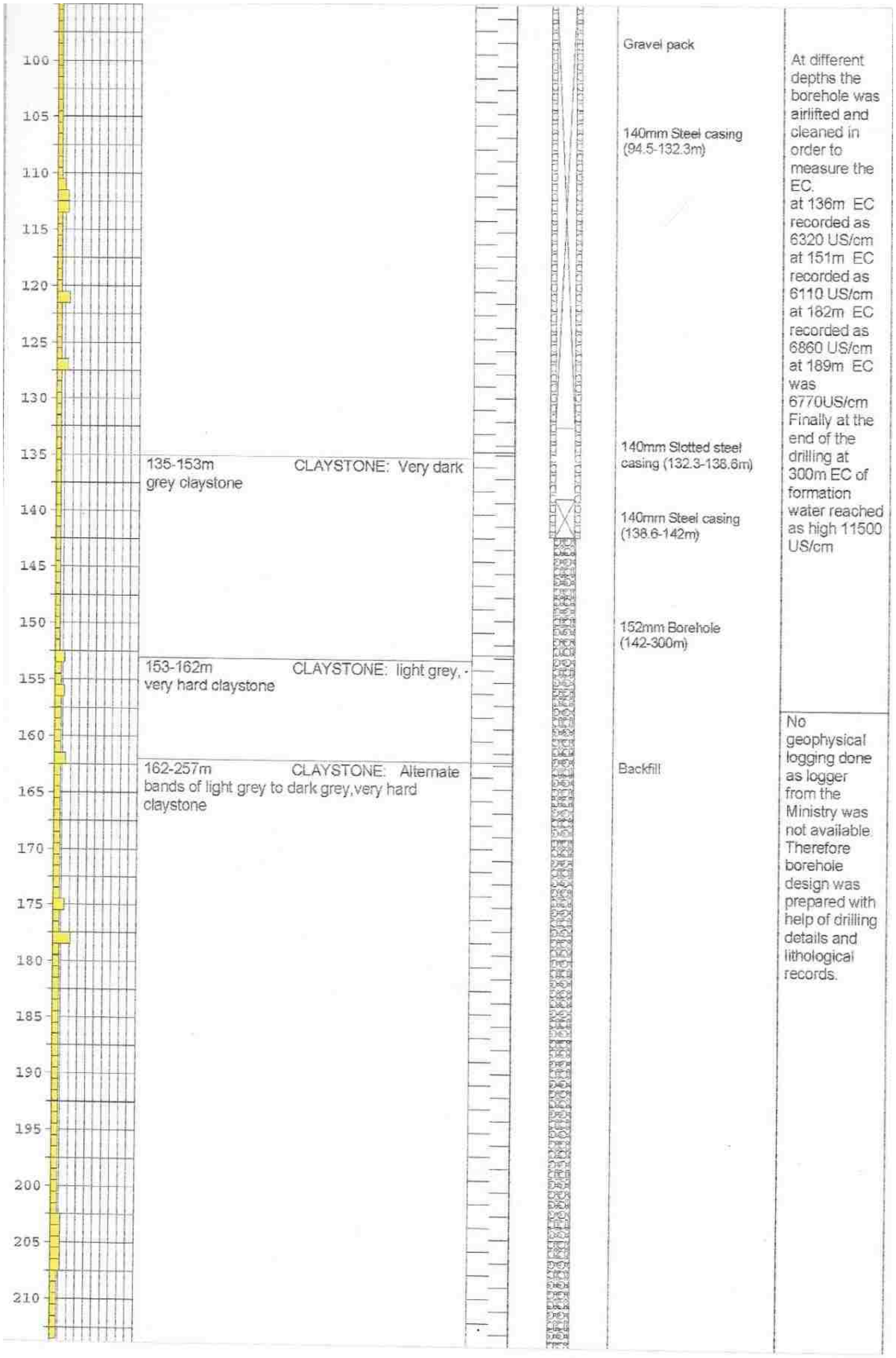


Figure 9: Geology of the study area



Lithological Description	Const. detail	Remarks
0-17.5m ALLUVIUM (slightly cemented): As granules and gravels of ophiolite, weathered, minor creamish limestone. Mostly crushed chips of ophiolite, 3-8mm size, poorly sorted. Appeared to be slightly cemented in creamish calcareous cementing material.	445mm Borehole to 6m 340mm MS Surface casing (+0.4-6m) Cement grout 216mm Borehole (6-142m) Sand pack (16.5-17.5m)	Dryhole, only seepages noted at 35m, 92m and 135m depth.
17.5-33.5m LIMESTONE (CLAYEY): Yellow, completely weathered to sticky clay, soft, fine grained. Few chips of hard limestone, angular to sub rounded with chips zize range from 5mm-15mm.	Cement seal (5-10% bentonite) Sand pack (22.5-23.5m) 140mm Steel casing (0-31.5m)	A Monitoring Well construction in bed rock
33.5-300m CLAYSTONE: Various shades of claystone as grey to 75m	140mm Slotted steel casing (31.5-37.8m) Gravel pack 140mm Steel casing (37.8-88.2m)	Drilling fluid = Air foam only EC of the water used for drilling remained about....US/cm
75-135m CLAYSTONE: Dark grey claystone	140mm Steel slotted casing (88.2-94.5m)	



135-153m CLAYSTONE: Very dark grey claystone

153-162m CLAYSTONE: light grey, - very hard claystone

162-257m CLAYSTONE: Alternate bands of light grey to dark grey,very hard claystone

Gravel pack

140mm Steel casing (94.5-132.3m)

140mm Slotted steel casing (132.3-138.6m)

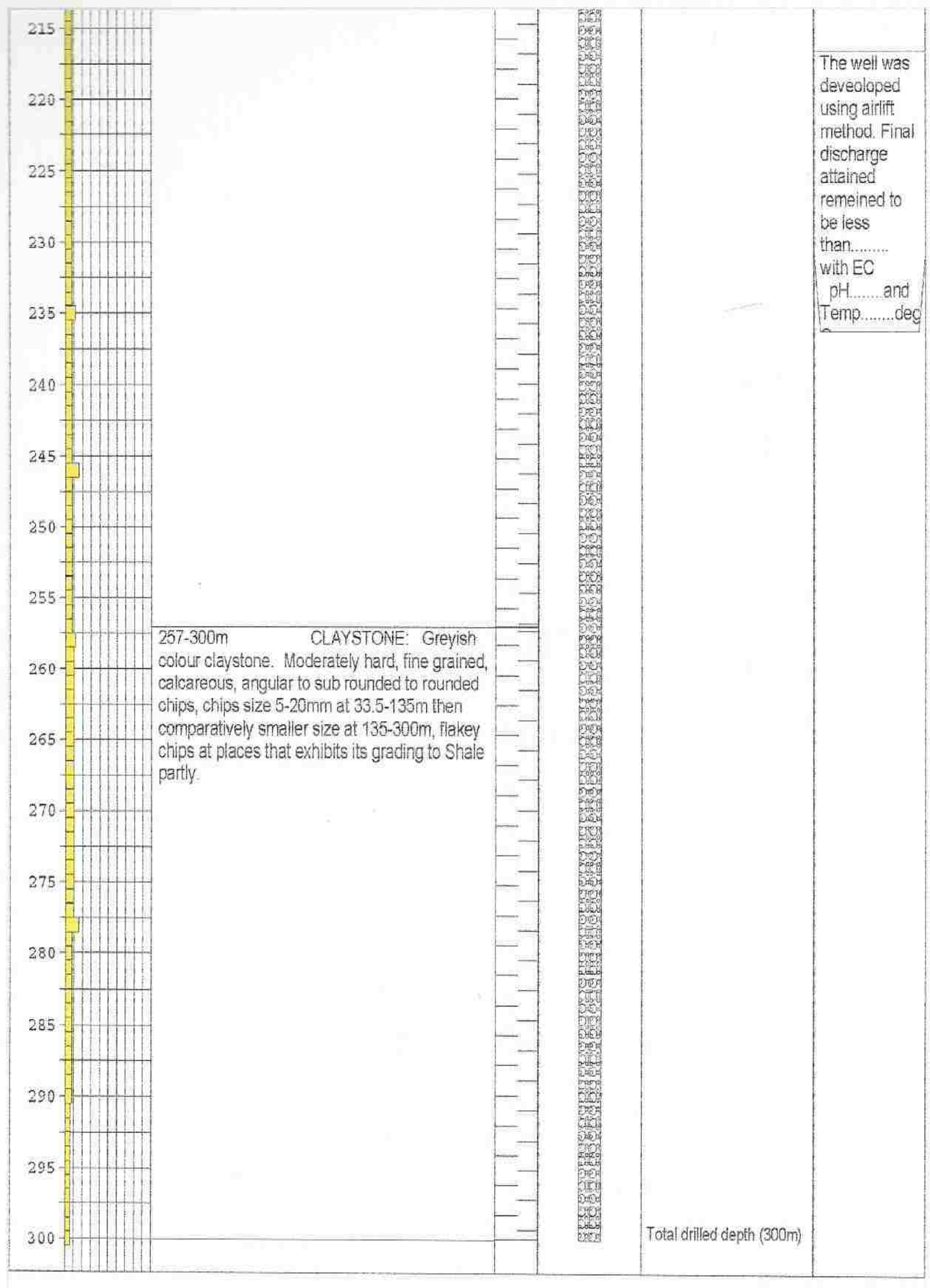
140mm Steel casing (138.6-142m)

152mm Borehole (142-300m)

Backfill

At different depths the borehole was airlifted and cleaned in order to measure the EC.
at 136m EC recorded as 6320 US/cm
at 151m EC recorded as 6110 US/cm
at 182m EC recorded as 6860 US/cm
at 189m EC was 6770US/cm
Finally at the end of the drilling at 300m EC of formation water reached as high 11500 US/cm

No geophysical logging done as logger from the Ministry was not available. Therefore borehole design was prepared with help of drilling details and lithological records.



257-300m CLAYSTONE: Greyish colour claystone. Moderately hard, fine grained, calcareous, angular to sub rounded to rounded chips, chips size 5-20mm at 33.5-135m then comparatively smaller size at 135-300m, flakey chips at places that exhibits its grading to Shale partly.

Total drilled depth (300m)

The well was deveoloped using airlift method. Final discharge attained remeined to be less than........ with EC pH.......and Temp........deg

Figure 10: Lithological description of the Al-Zoroup area

Chapter 3: Literature Review

3.1. Groundwater Hydrogeochemisty

The function of solution kinetics, rock-water interactions and geology should be clarified before we can define the term hydrochemcial. In addition, studying the contamination sources used to determine the quantities of water that differ in their chemical composition which got it from geological formations in the catchment areas (Srinivasamoorthy et al., 2014).

On the other hand, the study of quantity of water alone is not sufficient to solve the water management problems because it is used for various purposes depending on its quality. Furthermore, a number of other researchers show that the hydrogeochemical characteristics of groundwater and groundwater quality in different aquifers over space and time are important parameters in solving groundwater management issues (Panigrahy et al., 1996).

Accordingly, there are many factors affecting the groundwater in the aquifer such as; amount, duration, intensity of precipitation, and depth of weathering, specific yield and general slope of formation toward drainage channels. It occurs under unconfined conditions as well as in the fractured rocks under semi-confined conditions. The thickness of weathered layer irrespective of rock type ranges from 2.2 m to 50 m (Srinivasamoorthy et al., 2014)**.** The other studies presented other factors including dissolution, precipitation, absorption and desorption, ion exchange reactions, and the residence time along the flow path, all of which affect the types and concentrations of chemical composition of groundwater (Currell et al., 2011).

However, lithological characteristics and geo-climatic conditions are initial factors that affect the groundwater quality and its dynamic change. Hydrogeochemical analysis of groundwater can provide insights into the origin and history of the materials and conditions with which water has been in contact (Prasanna et al., 2011). Hence, it can classify the factors which directly affect the quality as; the natural processes (lithology, groundwater velocity, quality of recharge water, interaction of water with soil and rock, and interaction with other types of aquifers), anthropogenic activities (agriculture, industry, urban development, and increasing exploitation of water resources) and atmospheric input (Helena et al., 2000). Additionally, saline water intrusions in coastal area, climate properties, topography of the area and soil characteristics are fundamental factors to changing groundwater quality (Reghunath et al., 2002).

The result of that is, sustainable use of groundwater is threatened by the quality of the groundwater and lack of insufficient management. Therefore, it is important to evaluate the geochemical conditions of groundwater and to assess the impact of anthropogenic contamination. Consequently, to reach these purposes, regional- or national-scale groundwater quality monitoring is established in most countries (Jousma & Roelofsen, 2004).

The factors (natural, anthropogenic or atmospheric) that mentioned above drive to existence different groundwater types reflect the existence of various hydrogeochemical zones. For example, Carbonic acid forms as rainwater dissolves carbon dioxide gas in the atmosphere and unsaturated zone. The incongruent dissolution of albite mineral increases Na+ and HCO_3^- ions in the groundwater thus giving rise to a Na^++HCO_3^- hydrochemical water type in the background aquifer. During incongruent dissolution, some of the mineral components go into a solution at a different or to a different extent than others (Alley, 1993).

Nevertheless, quality of groundwater changes and is modified when it moves along its flow path from recharge to discharge areas through the processes like: evaporation, transpiration, selective uptake by vegetation, oxidation/reduction, cation exchange, dissociation of minerals, precipitation of secondary minerals, mixing of water, leaching of fertilizers, manure and biological processes (Appelo & Postma, 2005). In order, rock dissolution is the dominant process effecting groundwater hydrochemistry. This is consistent with observations about the general hydrochemistry of groundwater from the major hydrogeological terrains. Where silicate mineral weathering is the major controlling process, concentrations of the major physicochemical parameters are relatively low (Gill, 1969).

Although the groundwater contains many chemical constituents that are present at different concentrations, most of the soluble components within groundwater are derived from soluble minerals present in soil and sedimentary rocks (Sharma & Sharma, 2005). Overall, 95% of the ions present in groundwater are sodium (Na^+), potassium (K^+), calcium (Ca^{2+}), magnesium (Mg^{2+}), chloride (Cl^-), sulphate (SO_4^{2-}), bicarbonate (HCO_3^-), and nitrate (NO_3^-). These ionic species account for most of the salinity and are referred to as “total mineralization” or; “total dissolved solids” (TDS) (Sundaram et al., 2009).

There are natural and anthropogenic sources of these constituents in the groundwater. For example, the source of potassium in groundwater is likely to be the weathering of areas of higher agricultural activities. Especially, during the farming seasons, the synthetic fertilizers are applied and the application of liquid fertilizer N-32% through fertilization is a common practice in the agricultural areas. Even though, sulphate is unstable if it exceeds the maximum allowable limit of 400 mg/ l. It is causing a laxative effect on the human system with the excess magnesium in groundwater.

The main sources of additional sulphate in groundwater are due to the dissolution of filtering water, leaching from fertilizers, septic tank and municipal waste. However, the sodium values in higher concentrations are due to contributions from silicate weathering processes and dissolution of Clay, Gravel, Kanker and Feldspar (Na+ Plagioclase) and by agricultural sources (Singh & Hasnain, 1999). However, some groundwater samples have Na/Cl ratios lower than those either from halite dissolution or from evaporation of rainfall. This is most likely due to the absorption of Na^+ by clays, which is a common process in salinized areas (Ghassemi et al., 1995).

Nitrate can be a natural constituent but high concentrations often suggest a source of pollution. In certain regions, increased nitrate concentrations were observed due to intense use of fertilizers (Protano et al., 2000). However, the most influential parameter of groundwater quality is electrical conductivity (EC). According to (World Health Organization, 2006) guidelines; the permissible limit of EC for drinking water is 1400 µs/cm. A semi-arid climate, high evaporation rate and nutrient enrichment may be responsible for the enrichment of EC. During infiltration or along the flow, groundwater may dissolve the $CaCO_3$, and $CaMg(CO_3)_2$ existence in the rocks by increasing calcium and magnesium ions in groundwater. The intensive agricultural activities in the study area may also directly or indirectly influence mineral dissolution in groundwater (Böhlke, 2002).

In several hydrochemical studies the sodium adsorption ratio (SAR) is also used. It is important for determination of suitability of irrigation water. SAR is an index of the sodium hazard of water and is determined on the ratio of sodium to calcium and magnesium. Sodium is one of the most studied elements because of its toxic effects to crops and detrimental effect to the soil texture. If the Na+ concentration is high it gets the soil hard and resistant to water entrance.

Moreover, the build-up of osmotic pressure in soil due to high sodium concentration causes difficulty in water absorption by plant roots. Plants are sensitive in varying degrees to soil salinity and when this exceeds a permissible limit, their growth is very weak, which lowers their productivity. Sodium adsorption ratio (SAR), is an easily measured property that gives information on the comparative concentrations of Na^+, $Ca^{2+,}$ and Mg^{2+} in soil solutions where $[Na^+]$, $[Ca^{2+}]$, and $[Mg^{2+}]$ are the mill equivalent weight of these ions in the soil solution the permissible limit of SAR is below 12. If it exceedes this limit, serious physical soil problems arise and plants have difficulty absorbing water (Sherif et al., 2012).

Based on much researche and groundwater investigations, the Middle East is affected by dry land salinity growing problem like many semi-arid parts of the world, such as Australia, Argentina, China, western USA, and India (Ghassemi et al., 1995).

For example, Psychoyou et al., (2007) used a network of 56 observation boreholes to monitor water levels and assess the quality of groundwater in the region of Marathon in Attica, Greece. They recoded the values of electrical conductivity (EC), hydrogen ion concentration (pH), total hardness (TH) and other related parameters as well as main anions and cations in rainy and dry periods. Severe deterioration of water quality recorded in the areas very close to the sea due to high salt loads. On the other hand and far way of coastal areas, a major problem in drinking water quality and management of domestic water supply is salinization of groundwater in dug wells and in deep boreholes (Gill, 1969).

The evaporation and recycling of saline groundwater due to land irrigation in clay-rich sediments at the breaks of slope and on the plains are the dominant controls on salinity. There is also a systematic variation with depth. The total dissolved solids (TDS) and EC values commonly increase dramatically between the upper slope bores and those on the mid slopes to the plains over horizontal distances (Cartwright et al., 2004). Regarding on the quality of groundwater and various factors affecting its quality, poor water quality adversely affects human healthy, and plant growth.

Furthermore, the effects of dry land salinity on agriculture and natural ecosystems are significant and include different disadvantages such as dieback of deep-rooted trees due to water logging of their root systems, replacement of local and benefiual grasses by salt-tolerant flora, formation of salt scalds, destruction of soil structure, increased saline water discharge into rivers and lakes, and damage to buildings and infrastructure (Karanth, 1987).

3.2. Groundwater Microbiology

There are several sources of water pollution. Point sources (e.g. household and industrial discharges), non-point pollution (e.g. agricultural and urban runoff), line source (saltwater intrusion, seepage from canals carrying waste water, etc.), physical (heat, suspended solids, etc.), chemical (inorganic chemical contaminations, brines, acid, NO_3-N, sulphate, petroleum products, etc.), biological (bacteria, virus, and protozoa derived mainly from sewage and decomposable materials),and nuclear (radioactive materials (Bhatnagar & Sharma, 2002).

In the case of groundwater, the biological source contains many different types of bacteria, fungi, and protozoa. In some way this pathogenic organism may infiltrate through soil, sediment and rock, and contaminate the underlying groundwater (Plazinska, 2000). There are many ways for the contaminants to enter to the groundwater such as;

- Missing or damaging well cap seals that are around the wires, pipes, or where the caps meeting the casing might be cracked.
- Cracks or holes in the well casing allow unfiltered water (through the soil) to enter the well especially in wells made of concrete, clay tile, or brick.
- Many older wells were not sealed with grout when constructed which allows contaminant to seep through the pore hole.
- Well flooding is a common problem for wellheads below ground in the farms that flood during wet weather or where flooding irrigation is used.
- When the well is next to septic tanks facility, drain fields, sewers, and animal feedlots. The surface water contamination can enter the well.

- Cross connections with wastewater plumbing. This water can mix with the well water (Gaffield et al., 2003).

In addition, groundwater contamination often occurs with the intrusion of surface water through poorly constructed wellheads, where the cover is not properly sealed to the bore hole around the metal annulus (Chauret et al., 1999). In areas where wells are used for the purpose of drinking water, there is a potential risk of faecal contaminants and agricultural wastes to leak into the groundwater and this risk exists even where wells are not used or it is very close to contaminants such as springs and base flow to streams.

During the last few decades, disposal of effluents from industries, municipal sewage and their percolation to groundwater, adversely affected the quality of the groundwater. For example, during 1981 the generation rate of wastewaters in India was estimated between 74,529 million liter/day and about 27 km^3 annually. The increased use of chemical fertilizers and pesticides further complicated the situation (Bhatnagar & Sharma, 2002). Moreover, quantity and type of chemical fertilizers, organic fertilizers, pesticides, poor water quality, storage of animal waste and feedlots, etc. are some sources of chemical / biological contamination which can be life threatening, if crossing the permissible limits (Lawrence & Foster, 1987). Thus, surface and groundwater supplies for drinking and other purposes can easily become contaminated with pathogens by the lack of management and efficient monitoring (Grossman et al., 1992).

3.2.1. Faecal Indicator Bacteria (FIB)

Faecal contamination of water bodies poses a definite risk to human health via waterborne pathogens. Monitoring for all waterborne pathogens in environmental waters is currently unrealistic due to many reasons including the great diversity of pathogens (including viruses, bacteria, and protozoa), and the disparate methods required for concentrating and analyzing them. Furthermore, many pathogens are difficult and costly to culture, and have low concentrations in environmental waters (Field & Samadpour, 2007: Stoeckel & Harwood, 2007). FIB have been selected for monitoring faecal pathogens due to their low pathogenic potential, high levels in sewage and feces, and relationship to pathogen presence (Plazinska, 2000).

Faecal Indicator Bacteria are present in the feces and intestinal tracts of humans and animals (Literak et al., 2010). FIB are present in high numbers relative to pathogens and are often easier to identify using guideline standard culture methods (Simpson, Santo Domingo, & Reasoner, 2002). Faecal contaminants in the groundwater averaged 106 cfu/100 ml, and did not meet current standards for irrigation (0 to 200 cfu/100 ml) in most western countries (Halperin & Aloni, 2003).

The presence of faecal coliforms in well water may indicate the presence of other bacteria, viruses, or disease-causing organisms. Shallow wells located in intensive agricultural areas serviced by septic fieldS are at the greatest risk of contamination (Galanis et al., 2010). The major FIB used worldwide include faecal coliforms, Escherichia coli, and enterococci (Tallon et al., 2005:Leclerc et al., 2001)

3.2.2. Escherichia Coli

One of the greatest members of faecal coliform is *E.coli* bacteria. It is one of the most common inhabitants of the human intestinal tract and is probably the most familiar organism in microbiology. Its presence in water or food is an indication of faecal contamination, although the bacterium is not usually pathogenic.

However, it can cause urinary tract infections, and certain strains produce toxins that cause travellers' diarrhoea and in some cases cause serious food-borne diseases (Tortora et al., 2007). The optimum temperature for *E. coli* growth in a rich complex medium is 39°C (maximum, 48°C; minimum, 8°C), although the exact temperature differs from strain to strain (Madigan et al., 1997). The National Health and Medical Research Council (2003) recommend the use of *E.coli* as a primary indicator of faecal contamination of drinking water.

E.coli has long been used as an indicator of faecal pollution due to its characteristics including not being normally pathogenic to humans, present at concentrations much higher than the pathogens it predicts, easier and less costly to detect than the pathogens themselves (Desmarais et al., 2002).

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3.2.3. Health Impact of Faecal Contamination

Faecal contamination is a human health risk. Humans can get water borne diseases such as typhoid fever, cholera, dysentery and enteritis (bacterial infection), hepatitis (type B) (viral infection), and parasitic protozoa can cause diseases including dysentery (*Ameba*), Giardiasis (*Giardia Lamblia*), and diarrhea (*Cryptosporidium*). The world health organization (WHO) estimates that 3.2 million people die prematurely every year from infectious diseases conducted through contaminated drinking water. This amounts to an average of almost 8700 premature deaths per day. The WHO also estimates that each year, diarrhea alone kills about 1.9 million people: 90% of deaths in developing countries occur in children less than 5 years of age. This means that diarrhoea caused by exposure to polluted water kills one young child every 18 seconds (Miller & Spoolman, 2011).

On the other hand, using contaminated water for landscape irrigation (in particular for household gardening) causes two major hazards. One hazard is the harmful environmental effect and pollution caused by elevated levels of salinity, which can alter the soil properties, damage plants and contaminate groundwater (Garland et al., 2000). The other hazard is related to potential health risks associated with the spread of pathogenic organisms (Dixon, Butler, & Fewkes, 1999). Recent outbreaks of gastrointestinal diseases focused public attention on one of the more widely known members, *Escherichia coli*. Most of these bacteria are harmless but some strains may cause serious illness. Its precence in drinking water sample indicates recent faecal contamination, meaning other pathogens are likely to be present.

Most outbreaks of *E.coli* are due to food contamination caused by a specific strain known as *E.coli 0157:H7*, which can cause serious illness and death (Domek et al., 1984). One of the history events in 2000 was the supply of water to Walkerton in Canada. *E. coli O157*:*H7* and *Campylobacter* spp contaminated the water originating from local farms. A Health Unit investigation reported an increase in diarrheal illness in elementary schools, long-term care facilities, and emergency departments in the Walkerton area (Häfliger et al., 2000).

Moreover, sixty-five patients were admitted to hospital, twenty seven of whom developed haemolytic uremic syndrome (rapid loss of renal function and a low platelet count) and six people died. An investigation followed and concluded that after the heavy rain in the area, the well water became contaminated with surface water carrying livestock waste. Sampling of animals kept on 11 local farms identified the *C. jejuni* and, *E. coli* responsible for the outbreak (Clark et al., 2003).

3.2.4. Bacterial Source Tracking

Recently, many types of microorganisms are used as indicators to predict the presence of and/or minimize the potential risk associated with pathogenic microbes (Scott et al., 2002).Therefore, understanding the origin of faecal pollution is fundamental in assessing associated health risks and implementing the best management practices.

Traditional and alternative indicator microorganisms have been used for many years to predict the presence of faecal pollution in water; however, it is well established that the majority of these organisms are not limited to humans but also exist in the intestines of many other warm-blooded animals (Murray et al., 1990). The following is microbial categories used as indicators for faecal contamination.

Bacterial indicators

Four bacterial genera are used as target organisms in MST studies: *Escherichia*, *Enterococcus*, *Bacteroides*, and *Bifidobacterium*. Species belonging to these bacterial groups are normally present in the feces of higher mammals and birds. One advantage of using *E. coli* and faecal enterococci is that they are relatively easy to grow (Simpson et al., 2002).

Protozoan indicators

Research with *Cryptosporidium spp.* oocysts has shown that there are genotypes specific to different animals (e.g., cow, dogs, and humans) and for this reason it is used to identify the origin of faecal pollution in storm waters. Using this approach indicates that humans and cattle feces were primarily responsible for the contamination of the surface water samples (Xiao et al., 2000).

Viral indicators

Studies using coliphages, which are viruses that infect *E. coli*, have shown that there are four different groups of male specific (or F+) RNA coliphages. Group I coliphages are found in both human and animal feces, group II and III are associated with human faecal contamination, while group IV is associated with animal faecal waste (Havelaar et al., 1990).

Chemical indicators

It is based on the premise that certain chemicals are only found in faecal samples. Different chemical compounds have been recently evaluated as tools to predict sources of faecal pollution. Human contamination was mostly traced by using chemical markers. For example, caffeine, fragrance materials, and fluorescent whitening agents have been under investigation due to their exclusive use by humans (Bull et al., 2002).

Immunological indicators

It is focused on the identification of species-specific secretory immunoglobulin a (sIgA) in faecally impacted waters. (sIgA) is normally present in the feces of mammals, and it has been implicated in the protection of intestinal mucosa against enteric infections (Simpson et al., 2002).

With this in mind, it is important to identify the different point and non-point sources and practices that contribute to the faecal contamination of surface and groundwater systems. Bacterial Source Tracking (BST) is a relatively new method that is increasingly being used to identify sources of faecal contamination (Scott et al., 2002).

Recently, various (BST) methods have been developed and these methods are either on library-dependent or library-independent of analysing phenotypic and/or genomic characteristics (Meays et al., 2004). This section will provide an overview of microbial source tracking methods that are currently being used to predict and identify sources of faecal pollution in the environment as well as provide insight into future directions in the microbial pollution field (Scott et al., 2002).

Several methods were developed and used in microbial source tracking.
The methods were divided into two groups: molecular methods and biochemical methods (Simpson, et al., 2002).

Phenotypic Methods used for microbial source tracking

This method provides an indication of the faecal pollution levels in surface waters.
The following are the types of phenotypic methods used for microbial source tracking:

Faecal Coliform/Faecal Streptococci (FC/FS) Ratio

According to many studies, human feces normally have a ratio of faecal coliform to faecal streptococci greater than or equal to 4.0, whereas ratios below 0.7 are associated with animal feces. Based on this criterion, several researchers have used the

FC/FS ratio to differentiate between human and animal faecal pollution sources.(Feachem et al., 1983)

Antibiotic Resistance Analysis (ARA)

The method uses antibiotic resistance patterns for faecal streptococci, enterococci, or *E. coli* to identify the faecal sources. The human faecal bacteria have greater resistance to antibiotics than those from animal sources. The faecal microbiota of animals should have significantly less and different resistance to the various antibiotics and concentrations used commercially (Kaspar et al., 1990).

Carbon Utilization Profile (CUP).

Another phenotype technique, CUP is based on differences between bacteria in their use of a wide range of carbon and nitrogen sources for energy and growth. The BIOLOG system allows the user to rapidly perform, score, and tabulate 96 carbon source utilization tests per isolate and is widely used in the medical field for identification of clinical isolates (Hagedorn et al., 2003).

Molecular Typing Methods used for microbial source tracking Ribotyping

Ribotyping consists of fingerprint pattern generation for genomic 16SrDNA restriction fragment length polymorphisms (RFLP). Essentially, patterns are based upon size differences of fragments of DNA related to the location of specific target sequences. In this technique, total genomic DNA is extracted from pure cultures after which the DNA is enzymatically treated. The digested DNA fragments are first separated via agarose gel electrophoresis and transferred to nylon membranes. Southern blot hybridization analysis is then performed using rDNA probes, which results in a pattern composed of four to twelve bands (Grimont & Grimont, 1986).

Length Heterogeneity-PCR (LH-PCR) and Terminal Restriction Fragment Length Polymorphism (T-RFLP)

LHPCR and T-RFLP are two other techniques which have recently been proposed and are based on the detection of fluorescently labeled 16S rDNA PCR products using an automated DNA sequence. These methods are used to analyze differences in lengths of gene fragments due to insertions and deletions, and once a suitable target sequence is identified, it can be followed easily through the automated process (Bernhard & Field, 2000).

Repetitive PCR (rep-PCR)

Hundreds of studies have used conserved sequences in bacterial repetitive elements as PCR primers to distinguish among different strains of the same bacterial species. This typing approach, also known as rep-PCR, has been used to study many types of bacteria, including phyto-pathogenic bacteria, human pathogens, and animal pathogens. (Rep-PCR has also been used to examine faecal bacterial strains isolated from different sources of faecal pollution (Simpson et al., 2002).

Denaturing Gradient Gel Electrophoresis (DGGE)

The DGGE technique is capable of discriminating between different PCR products of similar size based on changes in electrophoretic mobility which are influenced by the melting properties of the DNA fragments. DGGE has been used to characterize and compare fecal and gastrointestinal bacterial populations of both humans and animals (Muyzer & Smalla, 1998).

Pulsed-Field Gel Electrophoresis (PFGE)

For PFGE analysis, pure culture bacterial cells are placed in agarose plugs where the DNA is digested using a series of restriction enzymes. These digested plugs are then imbedded into specialized electrophoresis gels and electrophoresed for an extended period of time (avg. 30 to 50 h) with alternating currents from different directions using specialized equipment (Dicuonzo et al., 2001).

Amplified Fragment Length Polymorphism (AFLP)

The AFLP technique has two main advantages over other fingerprinting methods: First, it has the ability to inspect an entire genome for polymorphisms. Second, it is highly reproducible. AFLP analysis has been applied to many different types of DNA samples (human, animal, plant, and microbial) thereby demonstrating its potential to become a universal DNA fingerprinting method (Savelkoul et al., 1999).

Chapter 4 : Materials and Methods

4.1. Water Sampling Locations

The study area at Al-Zoroup contains a number of private farms that obtain water from wells. First, 20 such wells within an area of about 6 km from northeast toward southwest were randomly selected for sampling (Figure 11). A GPS device was used to identify the UTM coordinates of each well.

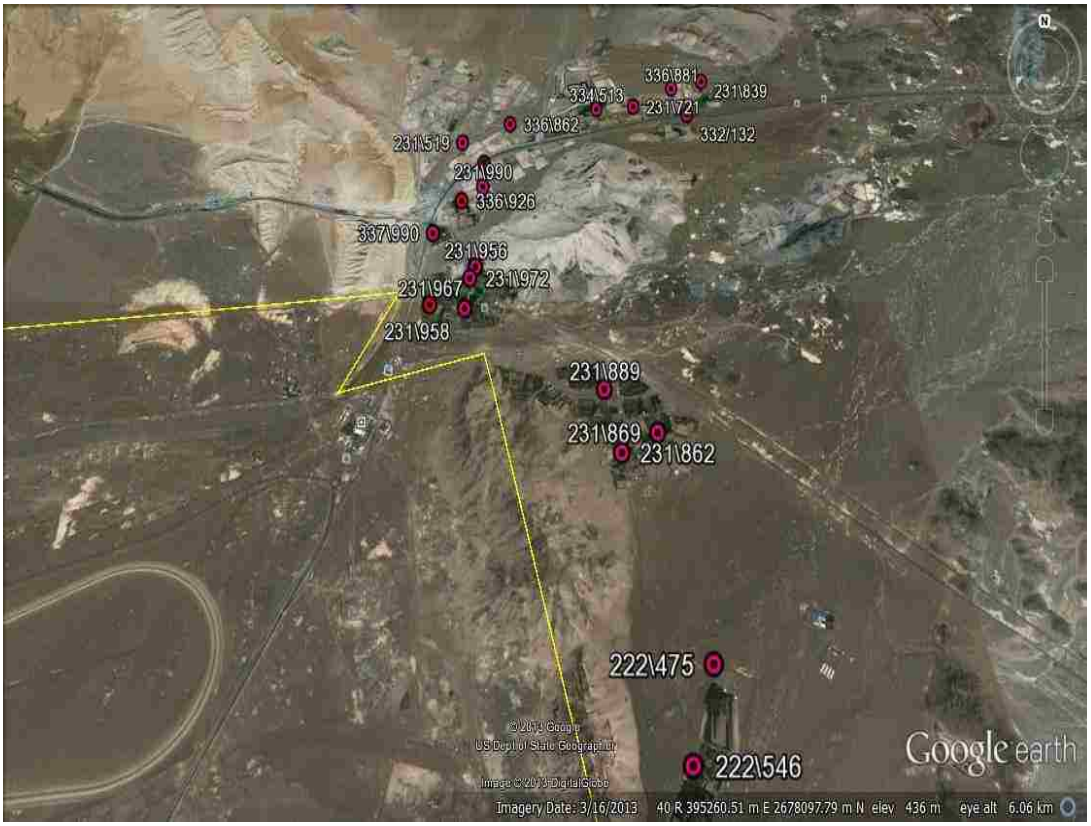


Figure 11: Map showing the 20 sampled wells in the Al-Zoroup area

4.2. Sampling Method

The water ran for three to five minutes before the collection of the samples. The water samples were collected for biological analysis (collected in duplicate, 500 ml samples) and shipped back to the laboratory using an ice box (temperature of the ice box less than 4°C).

Water samples used for the chemical analysis (cations & anions) were collected in two (1000 ml) polyethylene bottles. One of the sample bottles used for cation analysis, was acidified by adding a few drops of nitric acid (HNO3, 65%), while the second sample bottle was shipped back to the laboratory using an ice box (temperature of the ice box 4°C).

Field measurements; water temperature, pH, total dissolved solids (TDS) and electrical conductivity (EC) were measured by geologic laboratory instruments (Hanna, USA). Collected groundwater samples underwent laboratory testing for major cations, anions, and trace elements at the Geology Department of UAE University using different advanced instruments.

Ionic Chromatography (IC) was used to analyze groundwater samples for major cations and anions (K^+, Na^+, Ca^{2+}, Mg^{2+}), (Cl^-, SO_4^{2-}, NO_3^-). To detect trace elements, such as Al, BA, Cd, Cr, Cu, Mn, Pb, Zn, N, Fe, Sr, P, Mo, Co, and V an Inductively Coupled Plasma Mass Spectrometry (ICP-MS) was used. The (848 titrino plus) was used for determination of HCO_3^-. The researcher used a Piper’s (1944) diagram, contour maps, and a Wilcox salinity diagram to interpret the collected data.

4.3. **Biological Analysis**

4.3.1 Membrane Filtration and bacterial counts

The researcher completed a biological analysis within twenty-four hours of sample collection and the method to the data was membrane filtration and performed under the laminar flow cabinet. The researcher placed the membranes on the plates to test for total bacteria, total Coliforms, and total *E.coli*.

The Millipore device (USA) was prepared firstly, sterilized, and fitted with three sterilized S-Pak membrane filters (white gridded, 0.45 ×47 mm). An appropriate culture medium was selected for the three bacterial groups (Tryptone Glucose Extract (TGE) Broth (Total Bacteria), M-End Total Coliform Broth (Total Coliforms), M-coli Blue 24 Broth (*E.coli*)), then the broth was dispeuse into a sterile Petri dish, evenly saturating the absorbent pad. Using the vacuum, a 100 ml water sample was allowed to draw completely through the filter. Then, with flamed forceps the membrane filter was removed from the funnel and placed into the prepared Petri dish. All plates were incubated at 37°C for total coliforms and total bacteria, and 44°C for *E.coli* for 24 hours. The colonies were counted under a zoom scope (Leica ZOOM 2000) (Germany), confirmed and reported.

4.3.2. **Growth and Isolation of *E.coli***

Firstly, a single *E.coli* bacterial colony was selected and cultured on Eosin-Methylene Blue (EMB) agar plate (0.375g/ ml). The plates were incubated for 24 hours at 44°C. The *E.coli* colonies (metallic green) grown on the plates were then placed in liquid growth medium, and incubated for 24 hours at 44°C.

4.3.3. Extraction of *E.coli* DNA

One milliliter of the liquid broth was collected and centrifuged to form a pellet and the supernatant was removed. Using the QIAmp DNA Mini Kit (Qiagen, USA) the following reagents were added in the following order: Buffers ATL (100 µl), AL (200µl), AE (200 µl), AW1 (500 µl) and AW2 (500 µl), proteinase K (20 µl), and ethanol (200 µl, 96–100%). The mixture was vortexed for five minutes prior to centrifugation at 3500 xg. In a heat block, the researcher incubated the cell mixture for 30 minutes at 56°C. The extracted DNA was stored in the refrigerator at −20°C. The DNA concentration was measured by using NanoDrop 2000, (Thermo Scientific Company). Table 4 illustrates the extracted DNA concentration (µg/µl) for the nine *E.coli* isolates.

Table 4: The extracted DNA concentration for the *E.coli* isolates (µg/µl).

Well ID	332/132	231/956	231/839	222/546	222/475	231/721	336/926	336/883	231/519
DNA concentration (µg/µl)	27.2	28.3	27.2	37.9	29.7	29.3	34.5	49.6	48.3

4.3.4. **PCR Amplification of 16S rRNA**

Initially 16S rRNA was amplified from *E.coli* isolates using specific primers:

i. ECA75 F: 5'-GGAAGAAGCTTGCTTCTTTGCTGAC-3'(25) and

ii. ECR619 R: 5'-AGCCCGGGGATTTCACATCTGACTTA-3'(26).

The PCR kit Qiagen Master Mixes (Qiagen, USA) were used to amplify 16S rRNA gene in the Gene AMP PCR System 9700 (Applied Biosystems, USA).

The lyophilized oligonucleotides were prepared using sterile water into a stock of 100 picomole and then diluted into 5 picomole as working primers. The amplification mixture (25 µl) for each of the nine *E. coli* DNA samples contained 2 µl of target DNA, 2 µl primer (1 µl forward and1 µl reverse), 12 µl of Taq mix, and 9 µl of water. Annealing temperatures of the primers were determined from the Certificate of Analysis provided with the primers. The thermal profile of the PCR was as follows: 1 min of denaturation at 94°C, followed by 30 cycles at 95°C for 30 s, 60°C for 1 min, and 72°C for 1 min. A final extension was performed for 7 min at 72°C followed by a 4°C hold.

4.3.5. **Agarose Gel Electrophoresis**

The PCR product (5 µl) from the nine *E. coli* samples were run on 1.1% (w/v) agarose gel (Promega, USA) in 1× Tris Borate EDTA (TBE) buffer (50mM Tris, 100mM borate, and 10mM Ethylene Diamine Tetra acetic Acid (EDTA), pH 8.2) in a NanoPAC-300 electrophoresis device for 30 min at 70 V. The size of the DNA (800 bp) fragment was determined by UV spectrophotometry; the gel image is shown in (Figure 12).

Figure 12: Gel electrophoresis for the amplified DNA fragments from *E.coli* 16SrRNA.

4.3.6. PCR Purification and DNA Sequencing

The researcher confirmed the amplified product size during the early stages of this study using agarose gel electrophoresis. Then, a treatment with EXO SAP enzyme was performed to remove the excess of primers and Deoxynucleotide Triphosphates (dNTPS). This step was necessary to improve the sequencing. The researcher transferred five microliter of PCR product into a sterile 0.2 ml Eppendorf PCR tube and added 2 µl of EXO SAP enzyme to the mixture, vortexed and incubated in the PCR machine at 37°C for 15 minutes to degrade the excess primers, dNTPS followed by a final incubation at 80°C for 15 minutes to inactivate the EXO SAP enzyme.

4.3.7. Cycle Sequencing

Big Dye Terminator v3.1 cycle sequencing kit (Life Technologies, USA) was used with specific 3 µl of the forward or reverse primer with 5 µl of PCR product. The PCR cycle condition was as follows: 96°C for 1 min; followed by 25 cycles of 96°C for 10 seconds, 50°C for 5 seconds, 60°C for 4 min and 4°C hold until ready to purify.

4.4. Ethanol/ EDTA Precipitation

After the cycle, sequencing Ethanol EDTA precipitation was used to produce a consistent signal by minimizing the unincorporated dyes used to label the terminators. The 96 well reaction plate was removed from the thermal cycler (PCR) and subjected to a quick centrifuge spin (Sigma, Germany). Then 5 µl of 125mM EDTA and 60 µl of absolute ethanol were added to each well. The plate was sealed with aluminium foil and mixed by inverting 4 times. The plate was incubated at room temperature in the dark (stored in a drawer) for 15 minutes. The mixture was transferred into a properly labelled 1.5 ml Eppendorf tube and centrifuged (Sigma, Germany) at 12000 g for 30 minutes. Decante the supernatant carefully and added 70% Ethanol into all 9 tubes. Centrifuge again at 12000 g for 20 minutes. Carefully removed the liquid using a pipette and left the tubes on a heat block at 50°C for a maximum of 5 minutes to evaporate the remaining ethanol. Then 10µl of Hi Di Formamide were added into each tube, mixed and then transferred to the sequencing plate.

4.5. **Sequencing and identification**

The plates with samples were loaded into a 3500 Genetic Analyzer, (Life Technologies USA). The generated sequences shown in the appendices (Table 3) were blasted against the database of *E.coli* 16s rRAN of the National Centre for Biotechnology Information (NCBI). Sequences were identified by comparison with the GenBank database using Basic Local Alignment Sequencing Tool (BLAST).

Chapter 5 : Results and Discussions

5.1. Physical Properties

The physical parameters identified which affect the quality of water and its usage included:

a. Hydrogen ion concentration (pH),

b. Electrical conductivity (EC),

c. Total dissolved solids (TDS).

However, the values of these parameters are changing with time and directly measured in the field. The following is a further discussion of these parameters.

5.1.1. Temperature

The temperature of the collected groundwater samples in the study area ranged between 27°C to 39.8°C, with an average of 31.9°C. The variations of samples temperature (Figure 13) are mainly similar for most samples among the study area. This may be attributed to groundwater recharge with rainfall, which usually has a relatively lower temperature than deep groundwater. The highest temperature in well No.4 may be related to low amount of recharge.

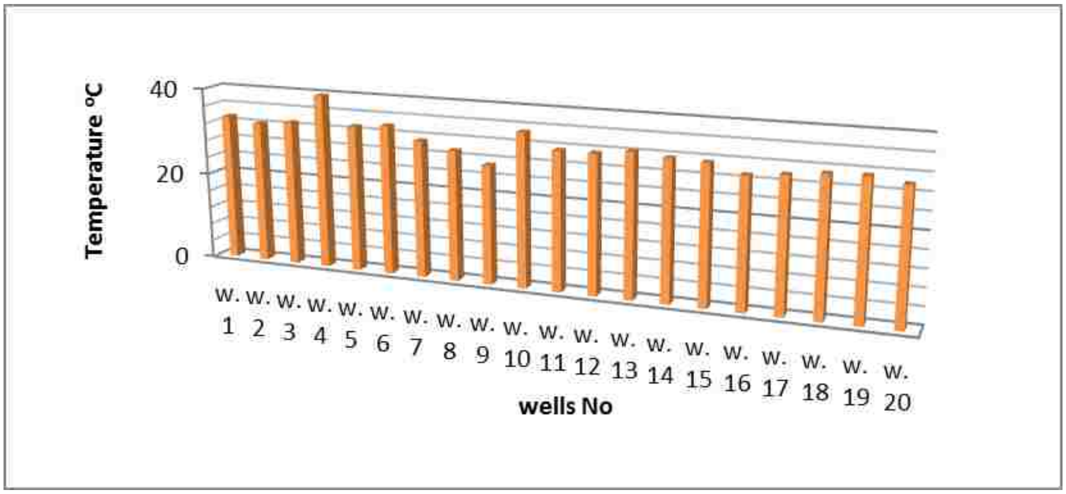


Figure 13: Temperature values of GW samples from the study area.

5.1.2 Hydrogen Ions Concentration (pH)

The pH is a measure of the acidity of groundwater. The lower the pH, the more acidic is the water. The pH is actually a measurement of the hydrogen ion (H^+) which is produced by the reaction, $H_2CO_3 = HCO_3^- + H^+$ (Nelson, 2002).

The pH values of the collected groundwater samples in the study area varied with a reading of 6.9 to 8.7 with an average of 8.34. This reading meets the WHO (World Health Organization) standard (6.5-8.5). WHO, 2011 for drinking water (Figure 14).

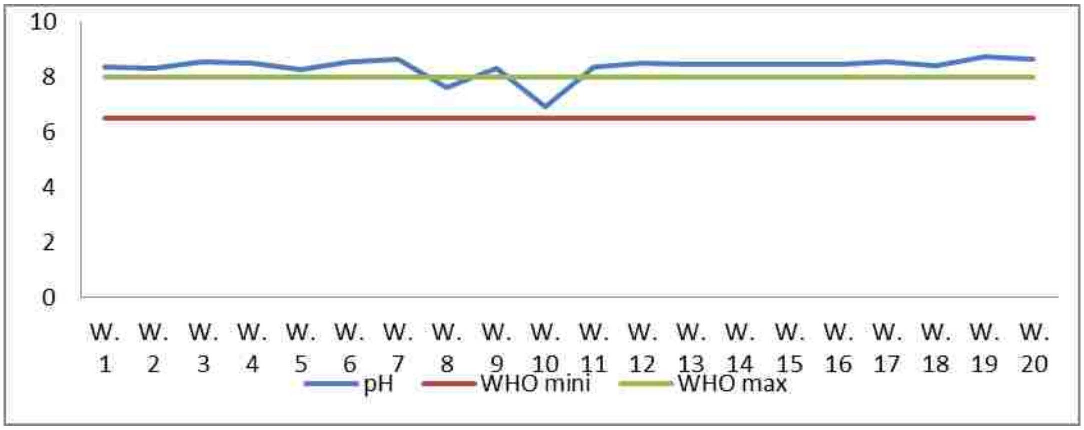


Figure 14: pH values of GW compared to WHO standards

The studied groundwater samples are mostly alkaline (up 7) except for well No.10 (pH = 6.92). The declining of pH in this well might be attributed to intensive fertilizer activity close to the well location, whereas the alkalinity could be linked to the dissolution of limestone, which is the main constituent of the aquifer matrix.The contour map (Figure 15) shows pH value variation among the study area.

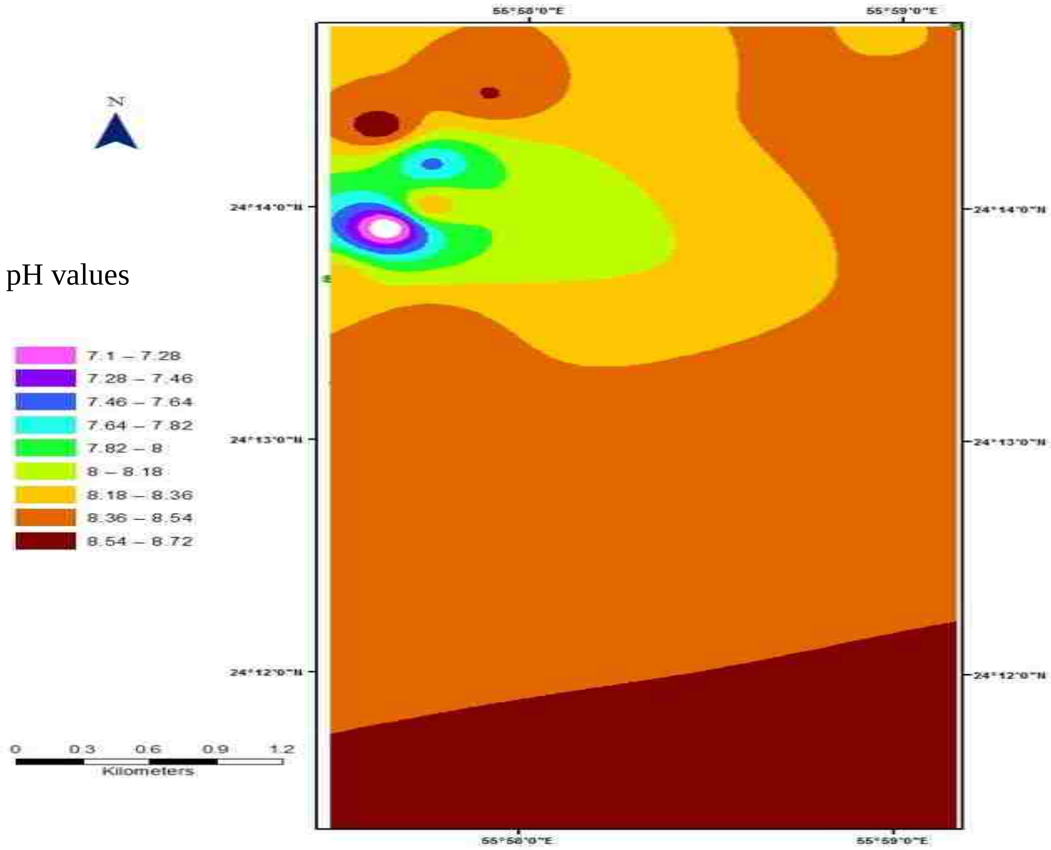


Figure 15: Distribution of pH in the study area.

5.1.3. Electrical Conductivity (EC)

The Electrical Conductivity (EC) of groundwater is a measure of its salinity and it is used as a parameter for the classification of drinking and irrigation waters (Erguvanli & Yüzer, 1987). In µS/cm unit, a measurement of electric current is reported.

The EC values of the collected groundwater samples ranged between 414 µS/cm of well No.3, and 4200 µS/cm of well No.8. The variations of EC in the study area were not significant except in samples No.8 (EC = 4200 µS/cm) and sample No.18 (EC= 2590 µS/cm). In this special condition, heavy pumping of the aquifer leads to migration of the brackish water due to up coning from the deep aquifer, resulting in high EC. Furthermore, higher values were identified and noticed along the southeast and northern parts of the study area as shown in the contour map (Figure 16) which might be attributed to the agricultural activities and intensive uses of fertilizers (Figure 17) such as cattle manure and Urea that will lead to increase EC.

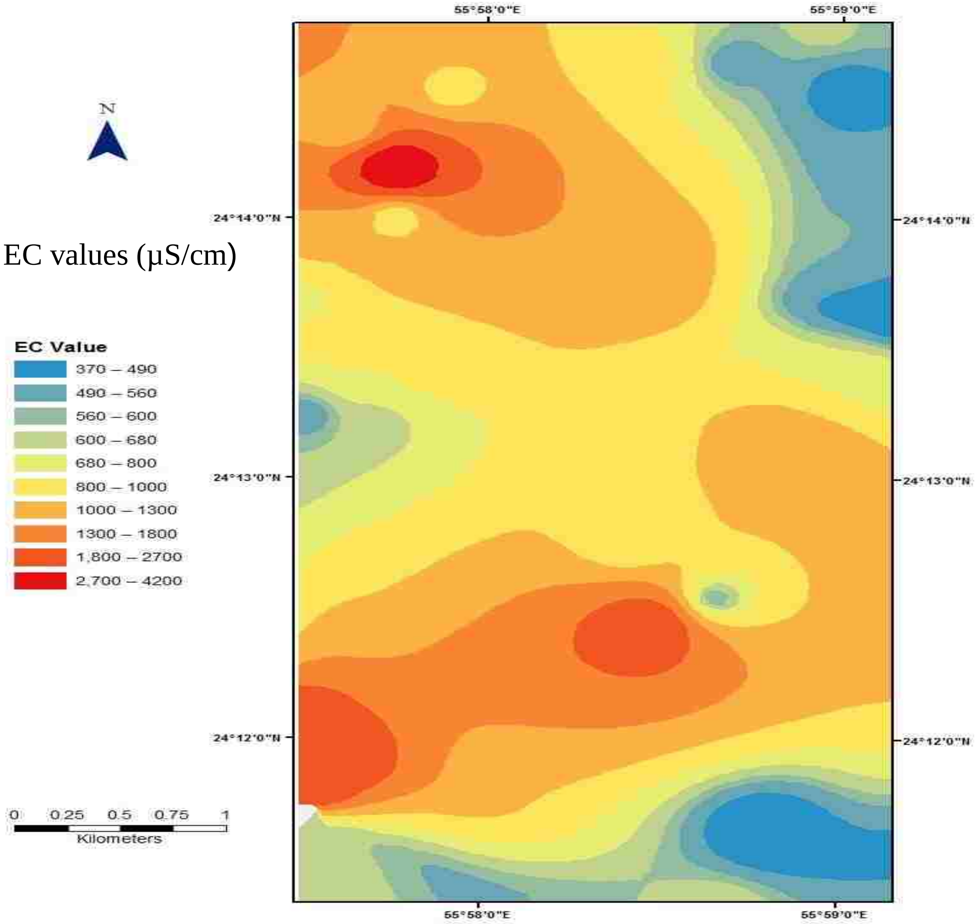


Figure 16: Contour map showing the EC distribution in the study area.

Figure 17: Organic fertilizer used in the study area.

5.1.4. Total Dissolved Solids (TDS)

Total dissolved solids (TDS) is a term referred to the sum of the concentrations of all dissolved solid chemicals in the water. How low a level of TDS is required or acceptable varies with the usage in domestic application. Standards might specify a maximum of 500 or 1000 (mg/l) as TDS for drinking water; 2000 (mg/l) as TDS might be acceptable for watering livestock (Kendall & McDonnell, 1999).

The recorded range of the TDS is between 152 mg/l to 4500 mg/l. It is noticed that the concentration of TDS increases to southwest of study area (Figure 18). Based on the WHO (2011) standard for drinking water and irrigation water as shown in the Table 3.1, the majority of wells (60%) are suitable for drinking and most of them (90%) are slight to moderate safety for irrigation, while 5% of the samples are lying within sever degree category.

Table 5: TDS (mg/l) in comparison with the WHO standard for drinking & irrigation.

	Drinking water	Irrigation water		
WHO standard	500	Degree of Restriction on Use		
		None	Slight to Moderate	Severe
		<450	450- 2000	>2000
# of wells not meeting the WHO standard	12(60%)	12(60%)	7(35%)	1(5%)

The highest value of TDS in well No.8 (4500 mg/l) may be ascribed to lowest depth of this well (200 m) as the farmer mentioned. In this case, heavy pumping of the aquifer leads to migration of the brackish water due to up-coning from the deep aquifer, leading to elevated TDS. Moreover, water interaction with rock and intensive dissolving constituents of return flow may help to elevate TDS in groundwater (Li, Zhang, & Hou, 2008).

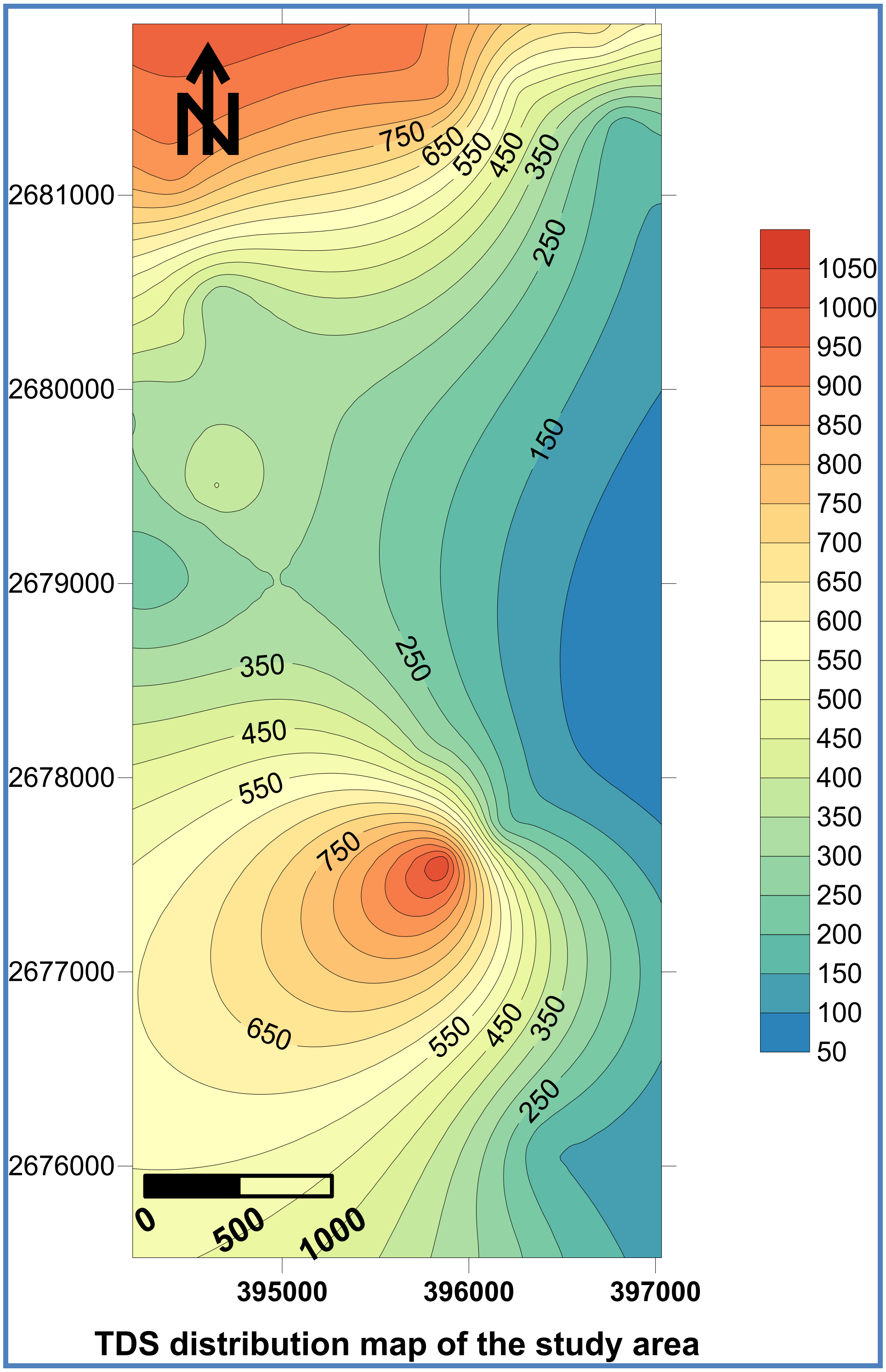


Figure 18: Distribution of TDS values in the study area, in mg/l.

5.2. Chemical Properties

Groundwater contains many chemical constituents that are present at different concentrations. The main source of soluble components within groundwater is derived from soluble minerals present in soil and sedimentary rocks besides leaching of dissolving chemicals from anthropogenic sources (Sundaram et al., 2009).

Overall, 95% of the ions present in groundwater are sodium (Na^+), potassium (K^+), calcium (Ca^{2+}), magnesium (Mg^{2+}), chloride (Cl^-), sulphate (SO_4^{2-}), bicarbonate (HCO_3^-), and nitrate (NO_3^-) (Sundaram et al., 2009). The following discussion is based on the chemical analyses of 20 groundwater samples collected from Al-Zoroup area. It includes discussion on the major ions concentration (Figure 19) hydrochemical water types, water–dissolved salts and hydrochemical ratios and all chemical results and analyses are illustrated in Appendix (Table 1).

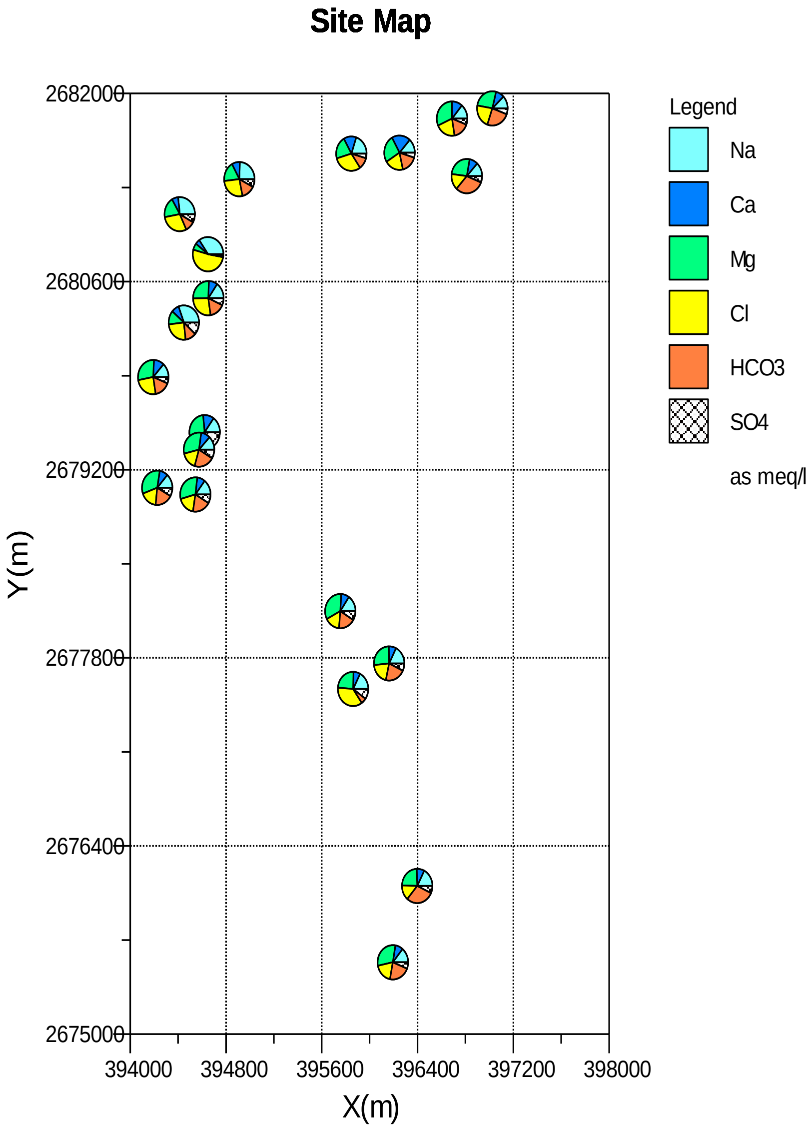


Figure 19: Concentration of major ions of GW samples

5.2.1. Major Cations

The order of the major cations of the clay limestone aquifer in the study area is Na^+ (56%) > Mg^{2+} (25%) > Ca^{2+} (17%) >K^+ (1%) as illustrated in (Figure 20). The following is a detailed discussion and comparison between groundwater samples with WHO (2011) standards of each element.

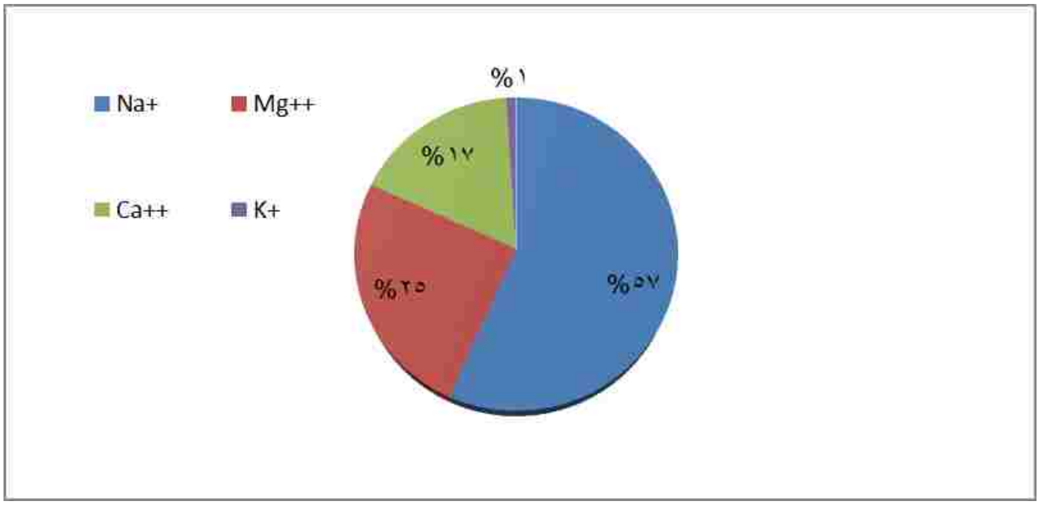


Figure 20: Abundance distribution of major cations (mg/l) in the study area.

5.2.2. Sodium (Na^+)

Sodium is a highly soluble chemical element in the water. It is often naturally found in groundwater and it has no smell but can be tasted by most people at concentrations of 200 (mg/l) or more. High concentrations of sodium in groundwater occur naturally in some areas. An increase in sodium in groundwater above natural levels may indicate pollution from point or non-point sources or salt water intrusion (Sapkota et al., 2007).

Davis and De Wiest (1966) stated that the most common sources of elevated sodium levels in groundwater are weathering of plagioclase feldspars mineral, argillaceous sediments, evaporation of water, irrigation and precipitation leaching through soils high in sodium, groundwater pollution by sewage effluent, and seeping from landfills or industrial sites. The Na^+ ion concentrations of groundwater samples ranged from 31 mg/l in well (No.3) to 1250 mg/l in the well (No.8) with an average of 133 mg/l. The variations of Na^+ concentrations of groundwater samples in the study area are presented in (Figure 21). It can be seen that the concentration of the Na+ increase toward the northwest of the study area. The overall trend of Na^+ values remains almost constant over the distance among the study area and within the WHO standards of drinking water (WHO standards of 2011 is 200 mg/l) and is illustrated clearly in (Figure 22).

However, well No.8, which lies in the center of the study area, has a concentration of 1250 mg/l. This case may be related to low depth of well (200 m) and to the irrigation process of using brackish groundwater from a deep saline aquifer. However, higher Na^+ in groundwater is related to silicate weathering (Singh & Hasnain, 1999), or it might be contributed from dissolution of clay, gravel, Kanker and Feldspar (Na^+ Plagioclase) and by agricultural sources (Srinivasamoorthy, Gopinath, Chidambaram, Vasanthavigar, & Sarma, 2014b). On the other hand, the relative decreasing of Na^+ concentrations of about 90% of water samples may be attributed to groundwater recharge events from different catchments of Mahadah and Wadi Al Jizi mountains (rainfall/recharge) and to the dilution by infiltrated water (Sherif et al., 2012).

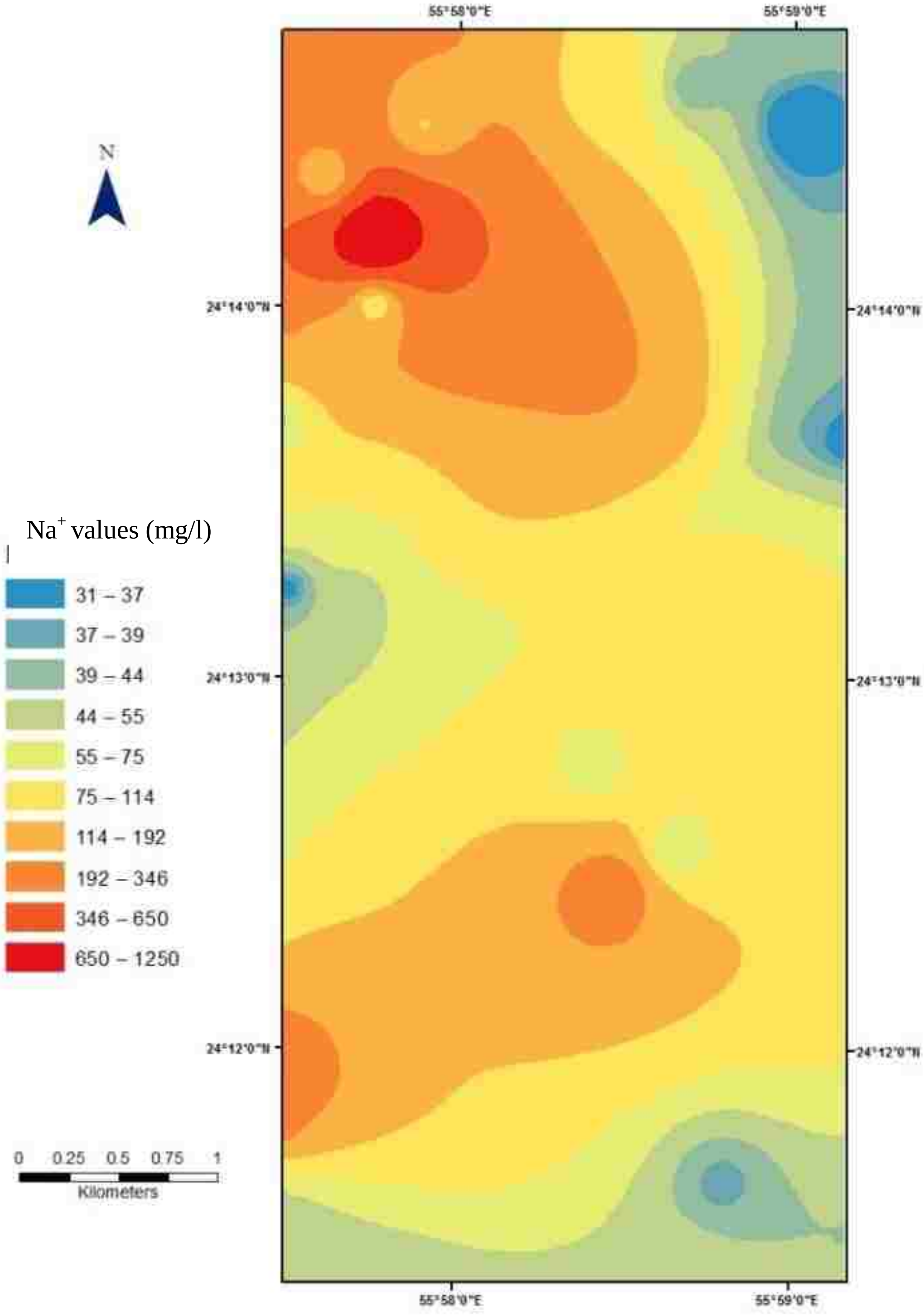


Figure 21: Distribution of Na^+ in the study area.

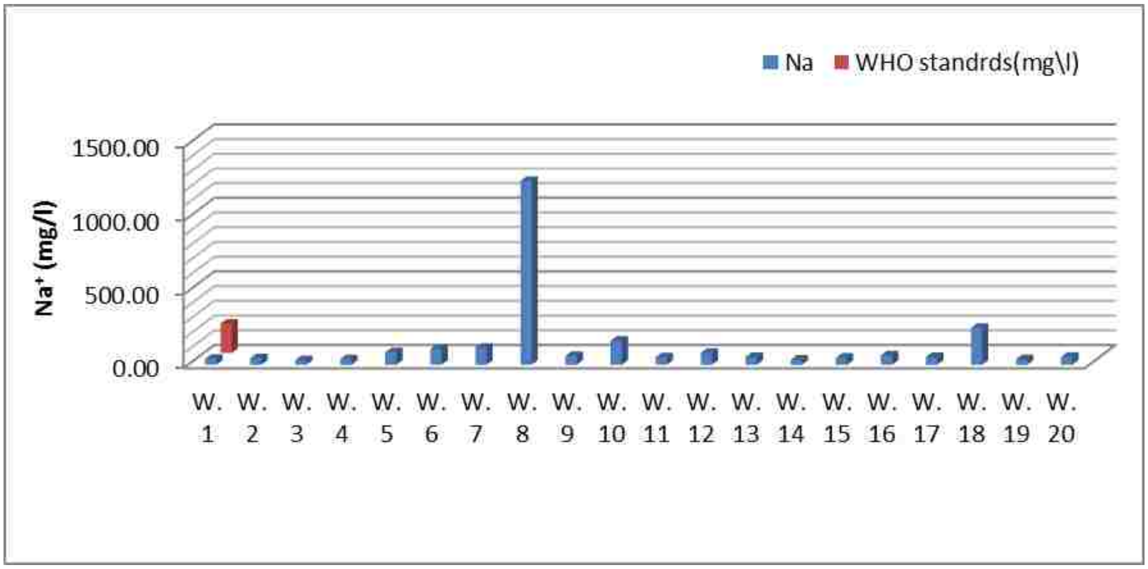


Figure 22: Na^+ concentration in GW samples compared to WHO standard.

5.2.3. Magnesium (Mg^{2+})

The Ca^{2+} and Mg^{2+} ions are aproximately similar state of equilibrium in most groundwater (Hem, 1985). However, Mg^{2+} ion in water affects the soil by making it alkaline and results in decreasing the crop yield (Kumar et al., 2007).

The overall (Mg^{2+}) concentrations in the study area are almost steady. However, the case of high concentration of Mg^{2+} in well (No.18) may be related to an increase in the recharge rate which could be an indication of weathering by hydrolysis of magnesium rich ophiolite from the mountain in northeast of the study area. Additionally, during infiltration or along the flow paths, groundwater may dissolve the $CaCO_3$, and $CaMg(CO_3)_2$which present in the rocks by increasing calcium and magnesium ions in groundwater (Srinivasamoorthy et al., 2014) . According to WHO (2011) standard guideline (50 mg/l) for drinking water, about 50% of water samples lay within the limit where high variation was observed in wells No. 8 and 18 as illustrated (Figure 23).

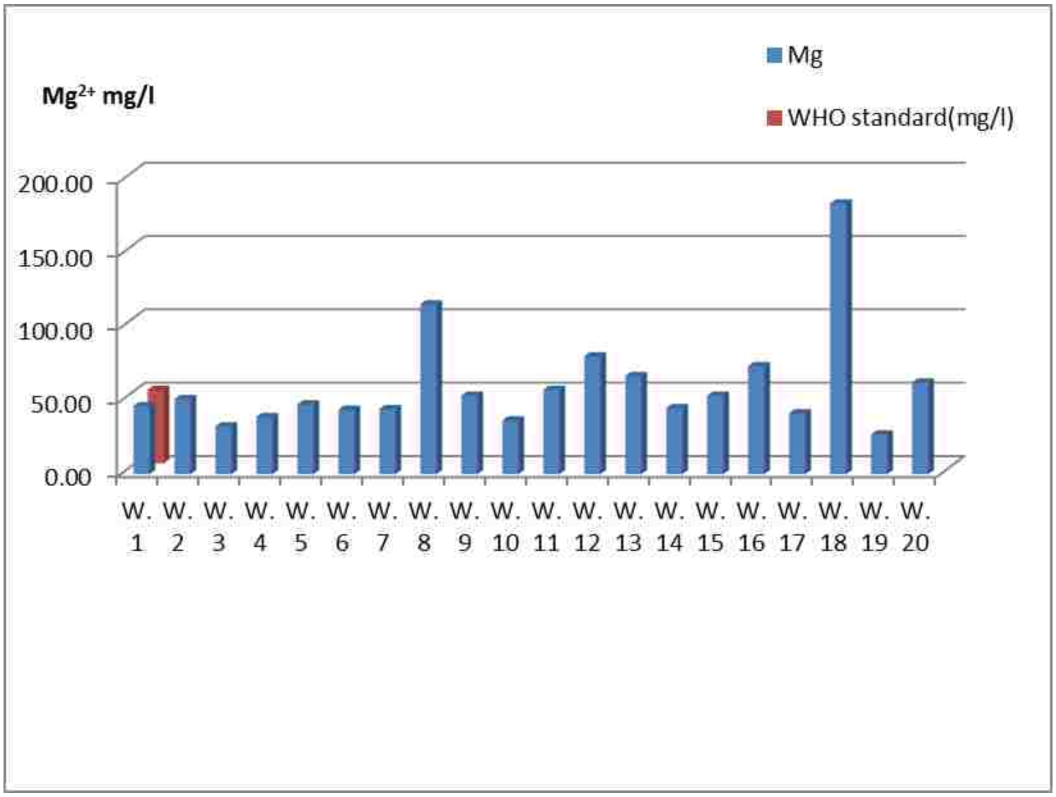


Figure 23: Mg^{2+} concentration in GW samples compared to WHO

The maximum permissible limit of magnesium in drinking water is 50 mg/l as suggested by WHO (2011). The high concentrations work as a laxative with an unpleasant taste and increases water hardness. On the contour map of this study area (Figure 24), the concentration variations of magnesium (Mg^{2+}) were plotted. The lowest value recorded in the well (No.19) was 27 mg/l and the biggest value recorded in well (No.18) was 184 mg/l. The average concentration of (Mg^{2+}) was 60 mg/l.

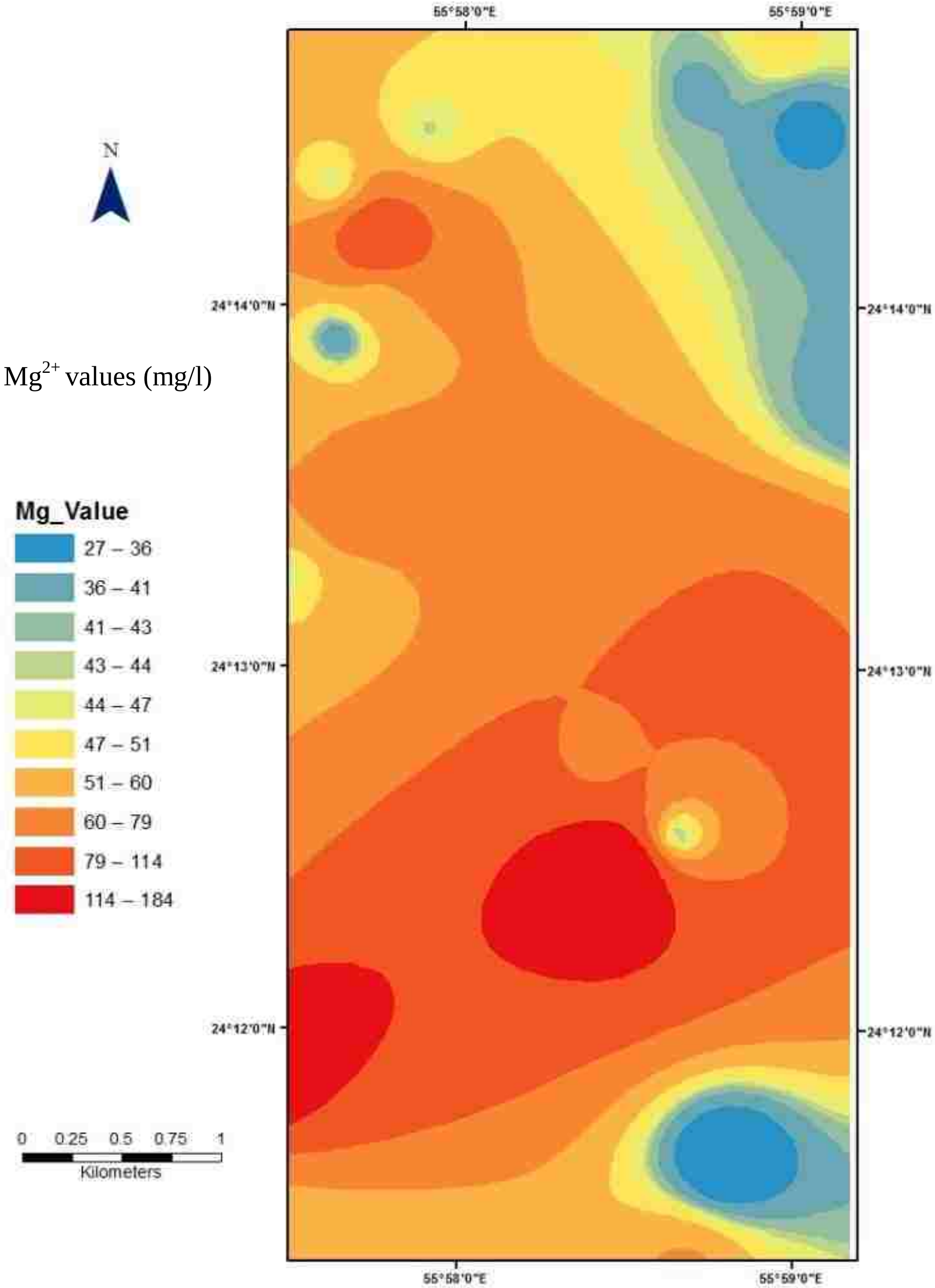


Figure 24: Distribution of Mg^{2}+ in the study area.

5.2.4. Calcium (Ca^{2+})

The most common form of calcium in sedimentary rocks is carbonates, particularly as limestone or dolomite, which are dominant in the study area. The dissolution of carbonate provided calcium to the groundwater (Hem, 1985). In addition, is derived from the weathering of silicate minerals (Cartwright et al., 2004).

In the study area, the lowest Ca^{2+} concentration is 15.091mg/l in sample no. 19, while the highest concentration is 149 mg/l in well No. 8 and the average of Ca^{2+} is 40 mg/l. Ca^{2+} values remain steady through the study area with little increase to northwest and southwest as illustrated (Figure 25).

This condition might be due to the flow of recharge during the rainy period, which dissolves $CaCO_3$ and $CaMg(CO_3)_2$, which present widely in abundant limestone rocks in the study area and therefore increase the Ca^{2+} in the groundwater. Furthermore, the intensive agricultural activities in this location may also indirectly influence Ca^{2+} dissolution in groundwater (Böhlke, 2002). Based on the WHO drinking water standards (WHO, 2011) (200 mg/l), all wells in the study area are within the range as plotted (Figure 26).

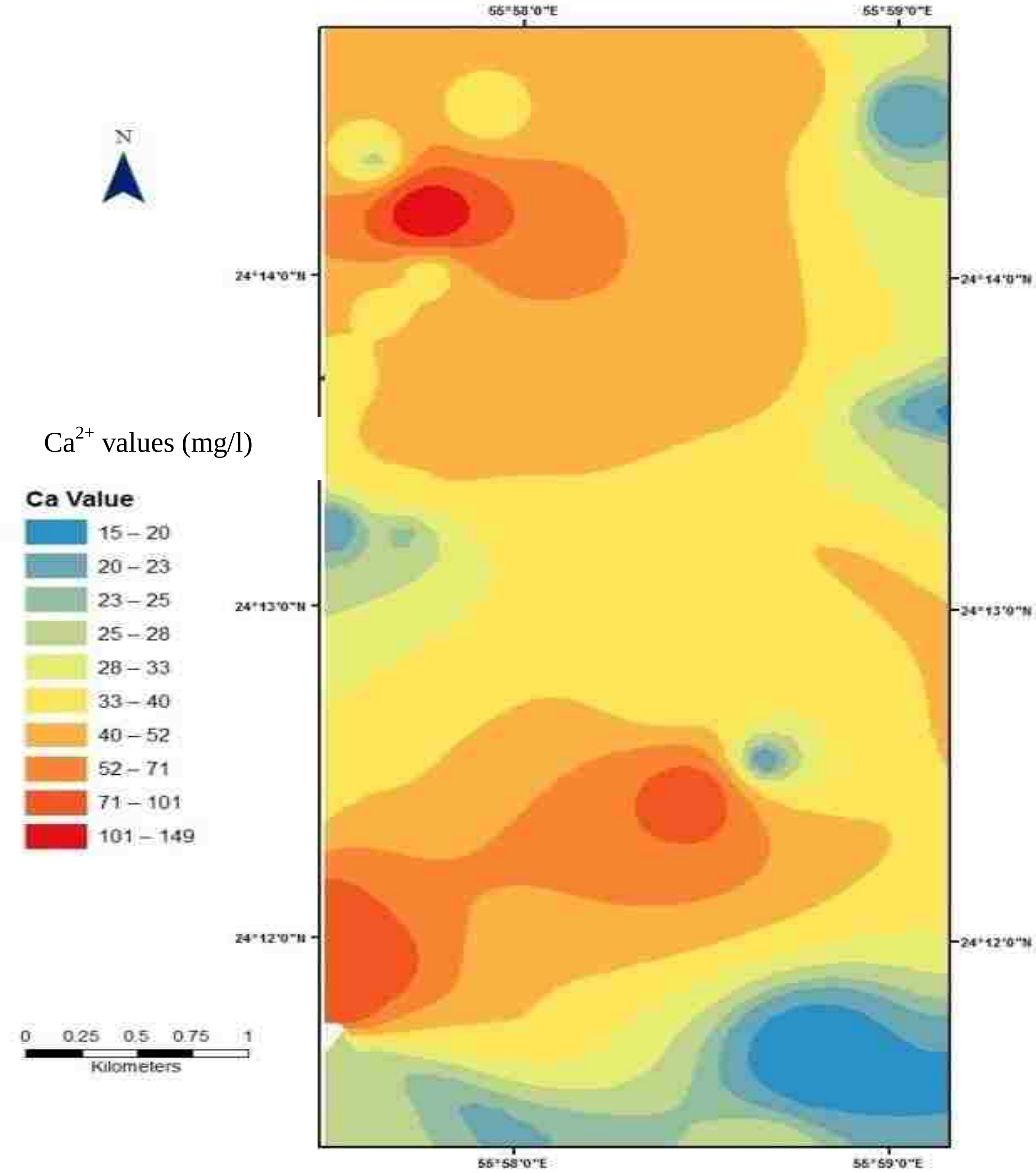


Figure 25: Distribution of Ca^{2+} in the study area.

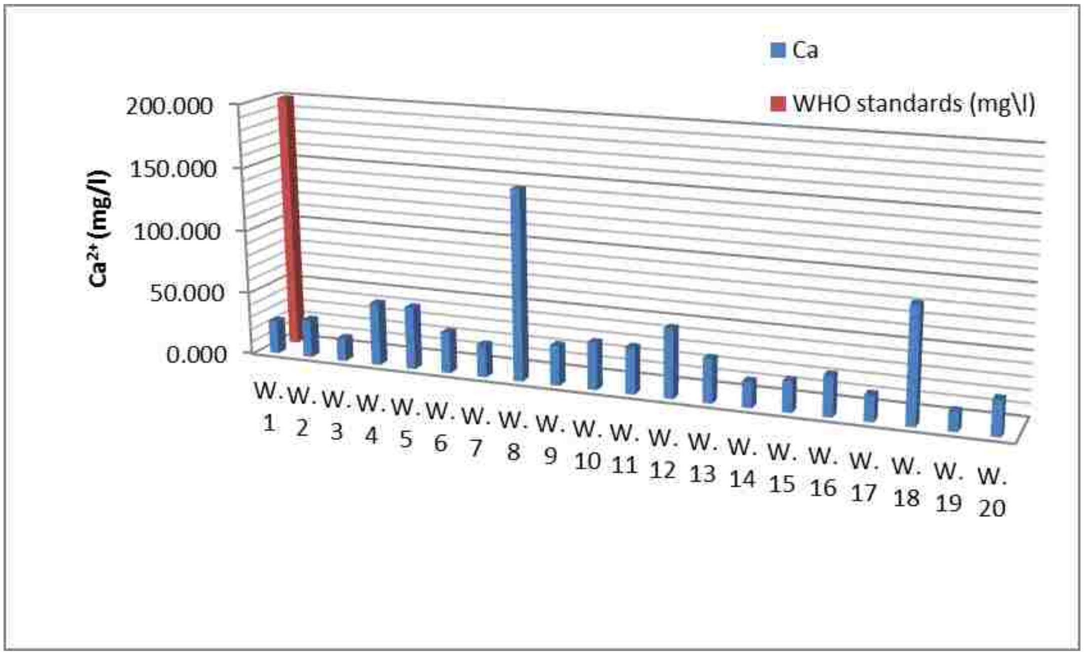


Figure 26: Ca^{2+} concentration in GW compared to WHO standard.

5.2.5. Potassium (K^+)

Potassium ion (K^+) is associated with Na^+ ion in the groundwater, but it usually has a much lower concentration compared with sodium ion. The natural common sources of potassium ions in groundwater are feldspars in igneous rocks, silicate, and clay minerals in sedimentary rocks whereas the anthropogenic source is synthetic fertilizers (Davis & De Wiest (1966).

K^+ concentrations range from 2 to 13 mg/l with a mean value of 3 mg/l and high K^+ were identified in the north as well as the northwest. Very little incensement to southern parts of the study area were observed as illustrated in (Figure 27). The spatial source of potassium in groundwater samples is likely to be the weathering of feldspar and the application of synthetic fertilizers. Potassium concentration of groundwater samples is within WHO (2011) value for standard drinking water (30 mg/l) as presented

(Figure 28). Lower potassium in groundwater is due to its greater resistance to weathering and fixation in the form of clay minerals that are dominant in the aquifer of the study area (Kolahchi & Jalali, 2007).

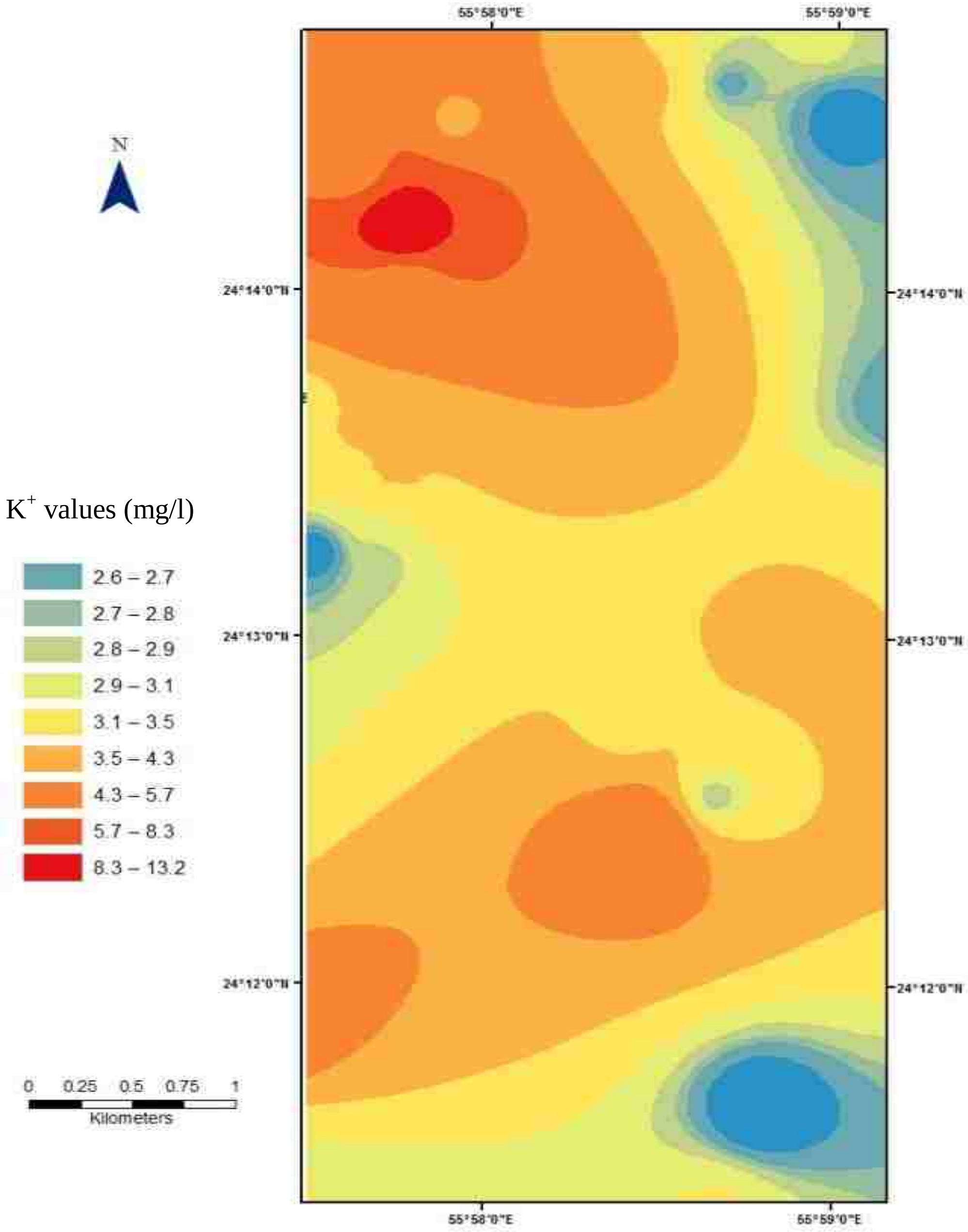


Figure 27: Distribution of K$^+$ in the study area.

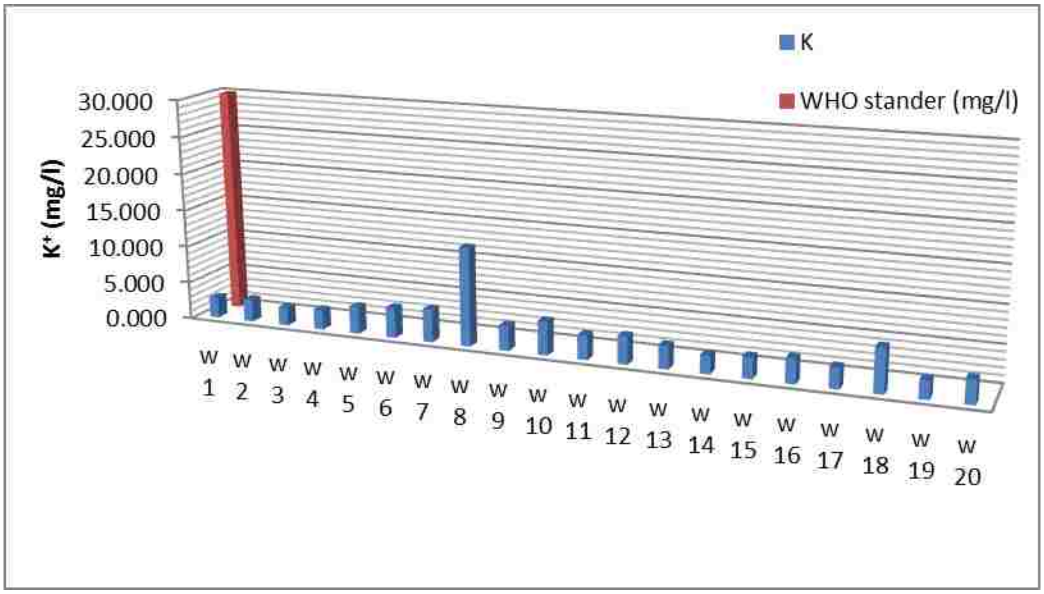


Figure 28: K^+ Concentration compared with WHO (2011) standard

5.3. Major Anions

In Figure 29, the order of major anions in the study area is illustrated as Cl^- (55%); HCO_3^- (31%); SO_4^{2-} (13%); NO_3^- (1%).

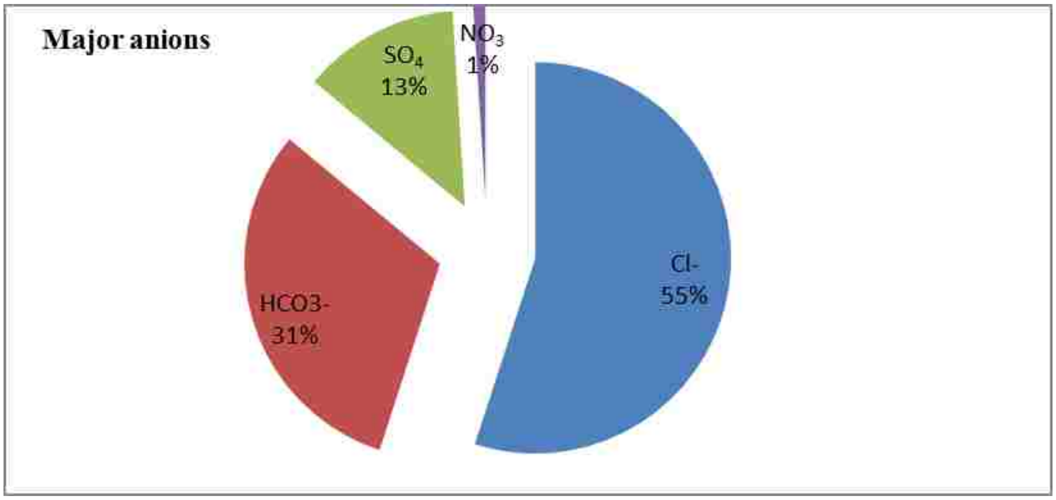


Figure 29: Distribution of major anions in GW of the study area

5.3.1. Chloride (Cl^-)

Sources of chloride in groundwater include rainwater, fertilizers and sewage water pollutants and it occurs naturally in all types of water. It could be related to human activities, which include road salt, effluent from industrial facilities, leachate from municipal landfills, effluent from private and municipal septic systems, and some agricultural chemicals. Natural sources include rock-water interactions, saline seeps, and minor atmospheric contributions (Srinivasamoorthy et al., 2014).

Chloride ranges in collected groundwater samples were from 46.9 mg/l to 2858 mg/l with an average of 299.8 mg/l. Figure (30) clearly shows that the variations of (Cl^-) in the study area are almost similar to most of the wells. The highest concentration is confined in the centre and northwest of the study area. The fluctuation in the measured values may be ascribed to the variation in the corresponding groundwater recharge events (Sherif et al., 2012). Chloride concentrations of 90% of groundwater samples are within the WHO (2011) permissible limits for drinking water (250-600 mg/l (Figure 31).

The highest concentration in well No.8 (2858 mg/l) and well No 18 (801mg/l) may mostly due to intrusion of deep saline water because of over pumping or due to agricultural return flow. Agricultural return flow water is characterized by higher concentrations of Cl^- attributed to the application of gypsum fertilizers (Vengosh et al., 2002).

According to Stuyfzand, (1993), the study area was divided into four regions based on Cl^- concentration (Table 6) as follows:

Table 6: Classification of GW samples according to Cl^- concentration

Classification	Cl^- concentration (mg/l)	No. of wells	Percentage
Fresh water	< 150	12	60 %
Fresh - brackish	$150 < Cl^- < 300$	6	30 %
Brackish	$300 < Cl^- < 1000$	1	5 %
Brackish - salt	$1000 < Cl^- < 10,000$	1	5%

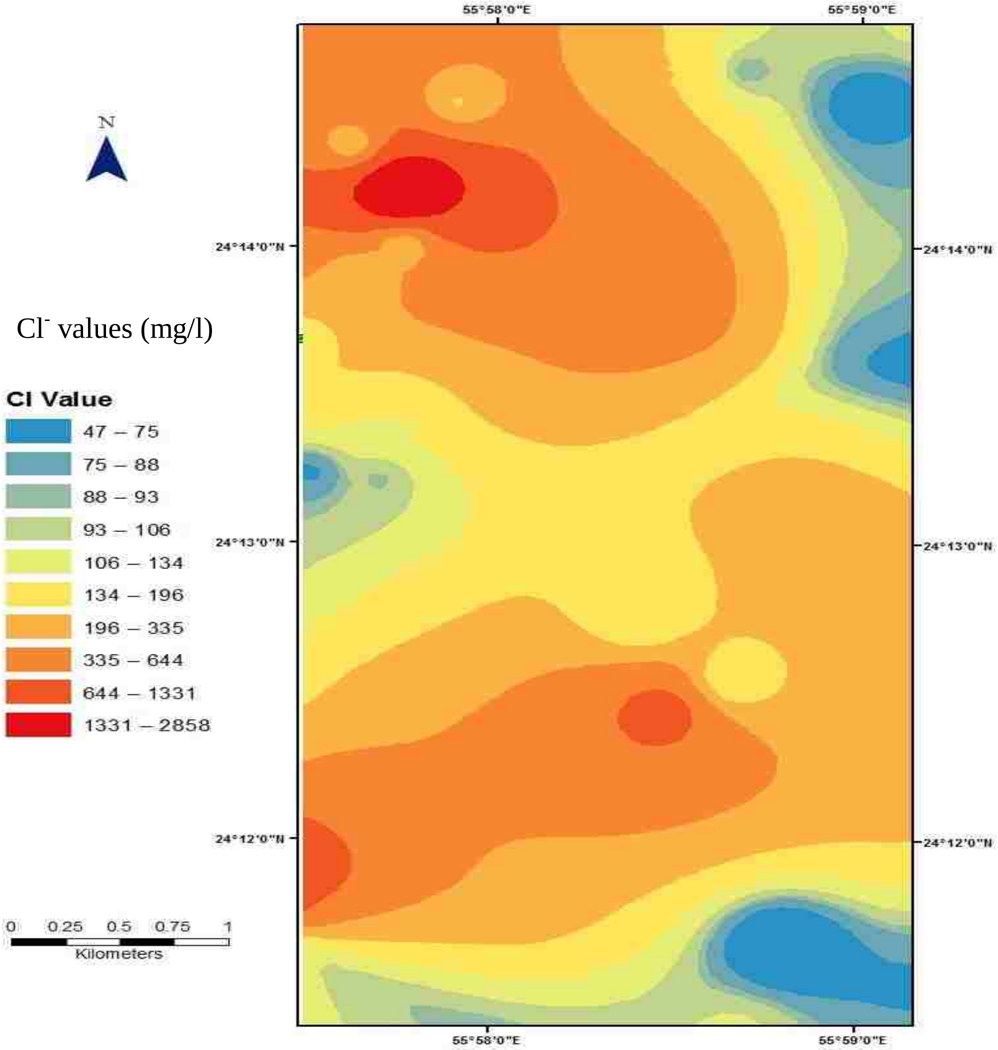


Figure 30: Distribution of Cl^- in the study area.

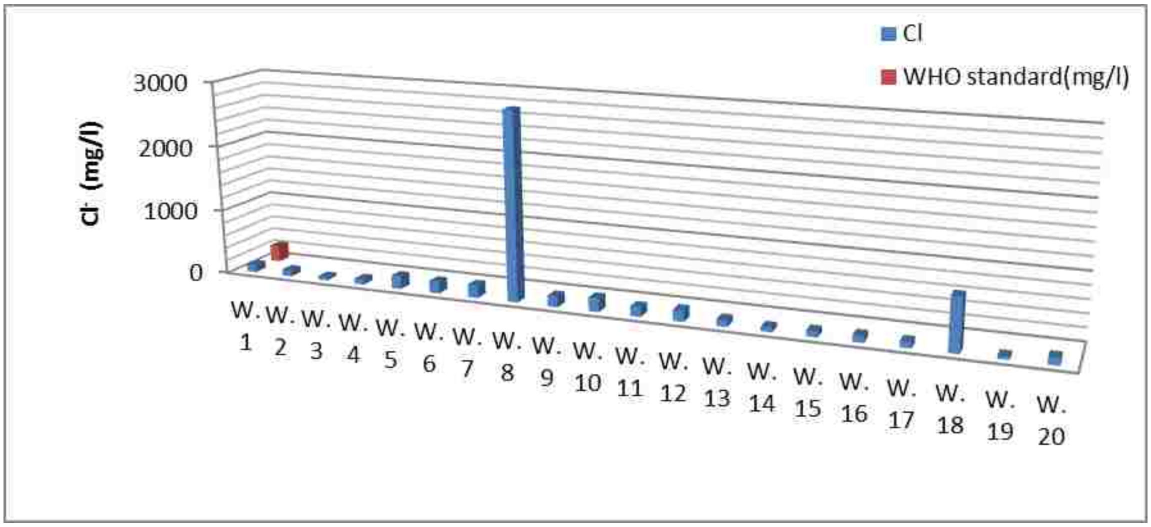


Figure 31: Cl^- concentration of GW compared with WHO (2011) standard

5.3.2. Bicarbonate (HCO_3^-)

Bicarbonate is abundant anion in the groundwater. The main source of bicarbonate in groundwater is carbon dioxide(CO_2) in the atmosphere, carbon dioxide in soil and by the weathering process of carbonate rocks which is mainly silicates (Subba Rao, 1998).

In addition, Lakshmanan et al., (2003) noted that the reaction of the feldspar minerals with carbonic acid in the presence of water (recharge events) releases HCO_3^-. The bicarbonate concentration in the study area ranged from 127 mg/l to 229 mg/l with an average of 61 mg/l. The increasing of bicarbonate to the southwest in the study area might be related to the dissolution of carbonate values along the mountain zone in Al-Zoroup area as shown in (Figure 32). This dissolution may be due to recharging flow from Wadi Hamad in the west and Wadi Al Jizi in the northeast catchment zone. The bicarbonate values compared with WHO drinking water standard (2011) (300 mg/l) is presented in (Figure 33). The data illustrated that there is a steady variation between most of the wells because all of the groundwater samples are laying within these limits.

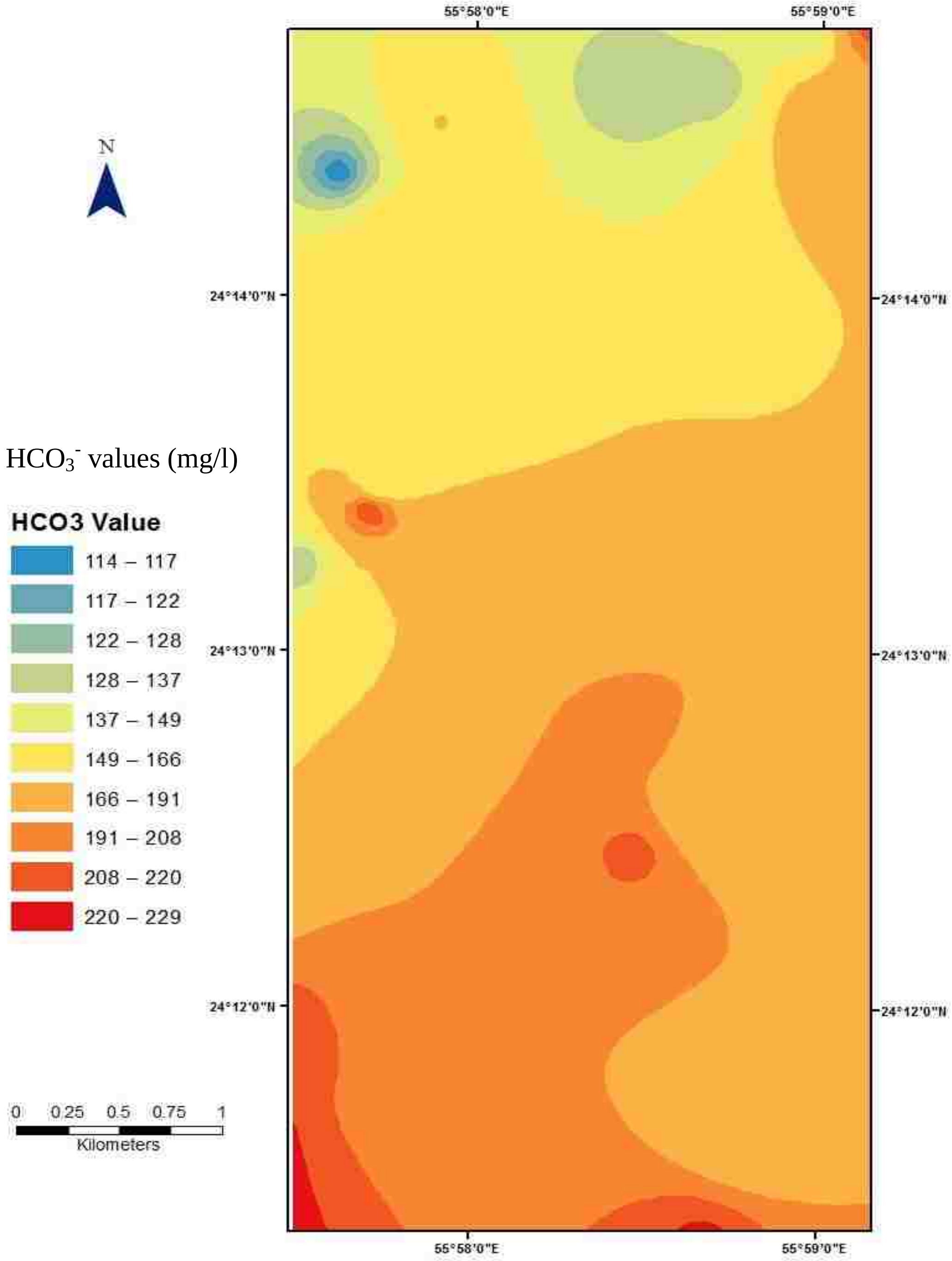

HCO$_3^-$ values (mg/l)

Figure 32: Distribution of HCO$_3$- in the study area.

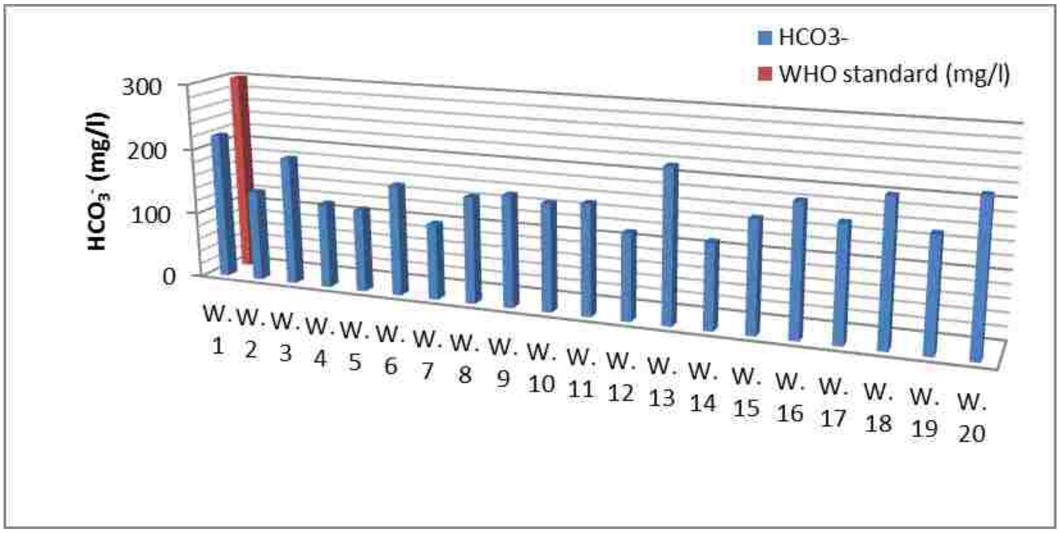


Figure 33: HCO_3^- Concentration of GW compared with WHO (2011).

5.3.3. Sulphate (SO_4^{2-})

The igneous rocks and sedimentary rocks are a source of metallic sulphate (Hem, 1985). Gypsum ($CaSO_42H_2O$) and anhydrite ($CaSO_4$) are examples of sedimentary rocks, which provide groundwater with sulphate (SO_4^{2-}).When the sulphide minerals interact with water, it is oxidized and yieldes groundwater with sulphate concentration (Todd, 1980).

The sulphate (SO_4^{2-}) ions in groundwater of the study area ranged between 27 mg/l to 291 mg/l with an average of 72.5 mg/l. The map of sulphate ion (SO_4^{2-}) distribution (Figure 34) shows that the highest values are to the northeast and central parts of the study area. This increas may be due to dissolution of gypsum and anhydrite within a limestone sequence of Al-Zoroup Mountains. In addition, it might be attributed to the dissolution of infiltrated waters, leaching from fertilizers and agriculture wastes, which are abundant in this zone of the study area. All the samples are within the permissible limit of the WHO drinking water standards (250mg/l) except well No.18 (29mg/l) as shown in (Figure 35). This is may be due to intensive fertilizer on this farm, which leads to leaching to the groundwater.

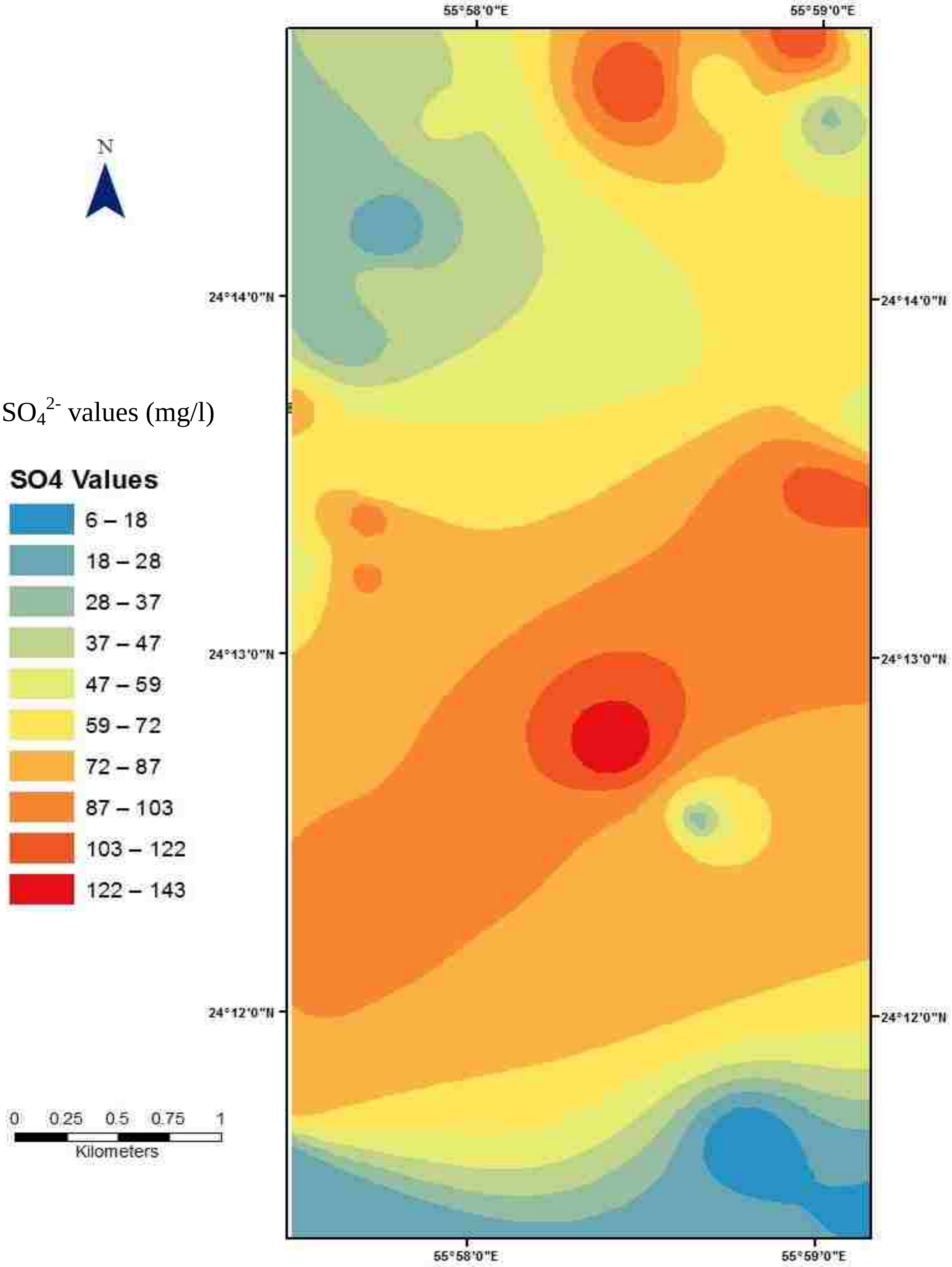


Figure 34: Distribution of SO_4^{2-}in the study area.

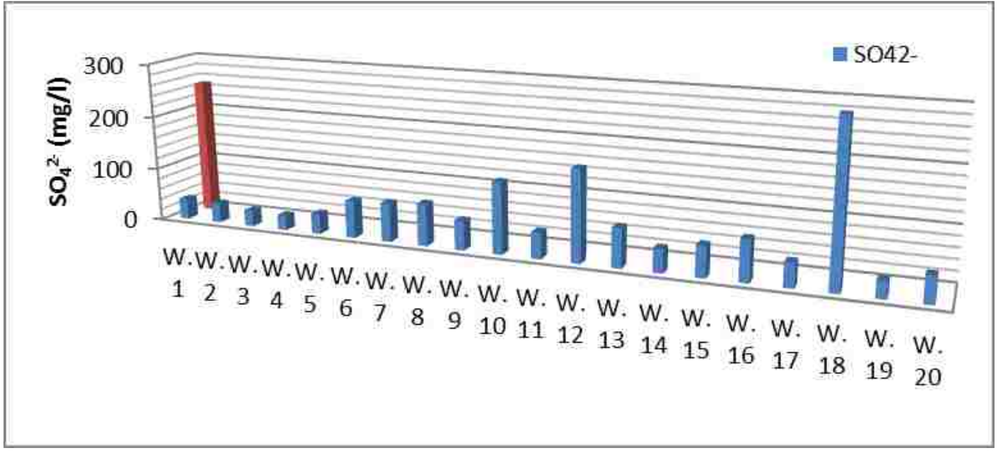


Figure 35: SO_4^{2-} concentration of GW compared with WHO standard.

5.3.4. Nitrate (NO_3^-)

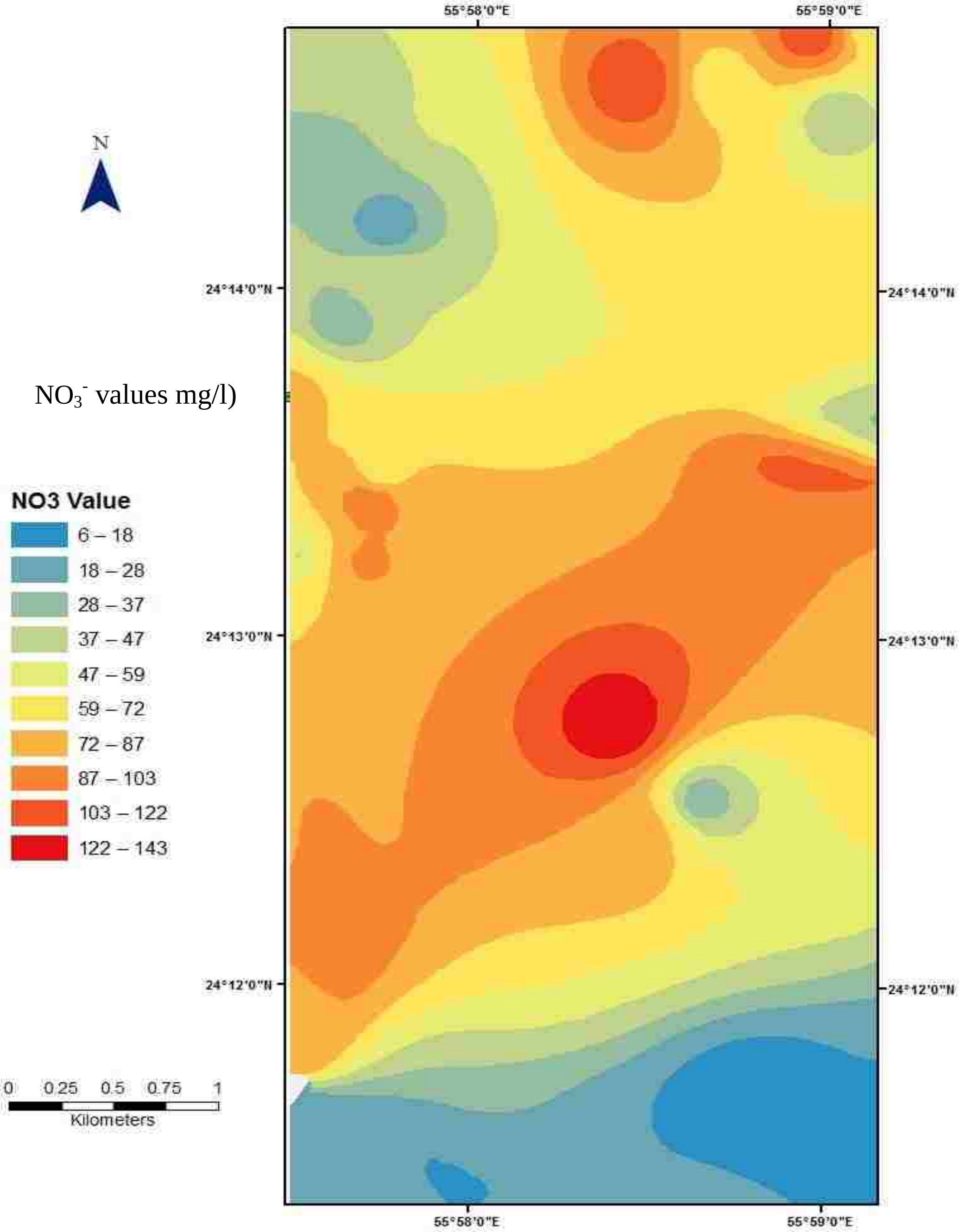
Nitrate (NO_3^-) is found naturally in the environment and is an important plant nutrient. The organic nitrogen fertilizer used in agriculture includes animal manures, human wastes, composites, sewage sludge, legume crops, and green manure crops. The most common inorganic nitrogen fertilizers contain nitrate (NO_3^-) and/or ammonium (NH_4).

The source of nitrate (NO_3^-) in groundwater, depends on the leaching, soil type and the amount of water in precipitation or irrigation (Mahler et al., 1990). Nitrate (NO_3^-) concentration in the study area varied from 6 mg/l in well No 19 to 143 mg/l in well No 16 with an average of 62 mg/l. The fluctuation in the measured values along the study area might be ascribed relatively to agricultural activities. As seen in (Figure 36), the highest values of (NO_3^-) is in the northern and central parts of the study area where high agriculture activities and fertilizer are used.

In comparison with the WHO's guideline (Mahler et al., 1990) as presented in table 7 and (Figure 37), most collected groundwater samples are safe for drinking purposes. The table shows five categories of NO_3^- concentration (mg/l) in the water. Each category referred to health effects on humans or livestock. The safe rate of NO_3^- in the drinking water starts from 0-44mg/l. It is dangerous in use if this concentration reaches up to 440 mg/l. Most infants less than six months old are in danger of nitrate poisoning and this might cause blue-baby syndrome when it reacts with haemoglobin to make a molecular called *Methaemoglobinaemia*. Death could also occur when 70% of the haemoglobin is converted to this compound. Nitrate might also interact with organic compounds to form N-nitrosamines, which could cause cancer. Additionally, livestock are affected by increasing nitrate when the bacteria in the animal rumen converts nitrate to toxic nitrate. Symptoms in animals include lack of coordination, laboured breathing, blue membranes, vomiting, and abortions (Mahler et al., 1990).

Table 7: Drinking water guidelines for water with known concentration of nitrate.

(NO_3^-) mg/l	Guideline	No of wells	%
0 to 44	Safe for humans and livestock	7	35%
45 to 88	Generally safe for human adults and livestock .Do not use for human infants.	8	40%
89 to 176	Short-term use of human adults acceptable. Short-term use for all livestock is acceptable unless feed sources are high in nitrate. Long-term use poses a risk.	5	25%
177 to 440	Moderate to high risk for human adults. Moderate to high risk for mature livestock if the feed is low in nitrate. Do not use for human infants.	0	0%
> 440	Do not use	0	0%



NO_3^- values mg/l)

Figure 36: Distribution of NO_3^- in the study area.

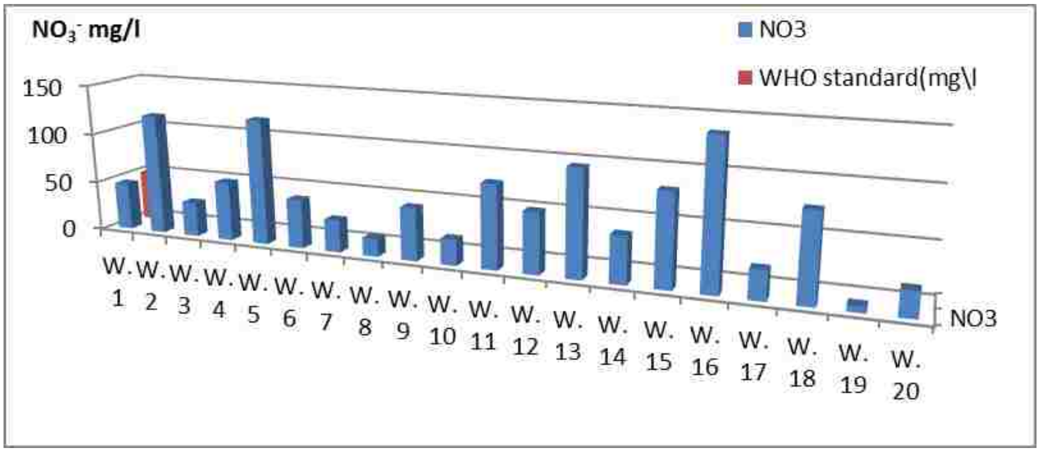


Figure 37: NO_3^- concentration of GW compared with WHO (2011) standard.

5.4. Ionic Ratio

Studying the ionic ratio is widely used to determine the effect of saline water and ion exchange upon the groundwater quality. A lower ratio of Na^+/Cl^- than seawater value (0.88) reveals seawater intrusion or mixing with deep saline water (Martens et al., 2011).

Accordingly, the values of Na^+/Cl^- versus Cl^- concentration (meq/l) of 20 groundwater samples in the study area, ranged from 0.48 to 1.23. (Figure 38) shows that two samples reflect Na^+/Cl^- molar ratio greater than 1. This result might be attributed to weathering of silicate process during the recharge events. The majority of samples (90%) present lower Na^+/Cl^- ratio, due to the dominance of Cl^- ions, which are present in the groundwater as sodium chloride.

Chloride value exceeding sodium may be due to the cation exchange occurring when the deep saline water mixes with freshwater from upper aquifer or due to pollution by anthropogenic activities including fertilizer, human or animal wastes, and agriculture applications (Jones et al., 1999). The other useful ionic ratio is Cl^-/HCO_3^- that is usesd to study salinization phenomena and its effect on groundwater quality (Todd, 1980). This ratio is classified as follows:

- < 0.5 for Unaffected of salinization
- 0.5 to 6.6 for slightly and moderately affected
- 6.6 For strongly affected

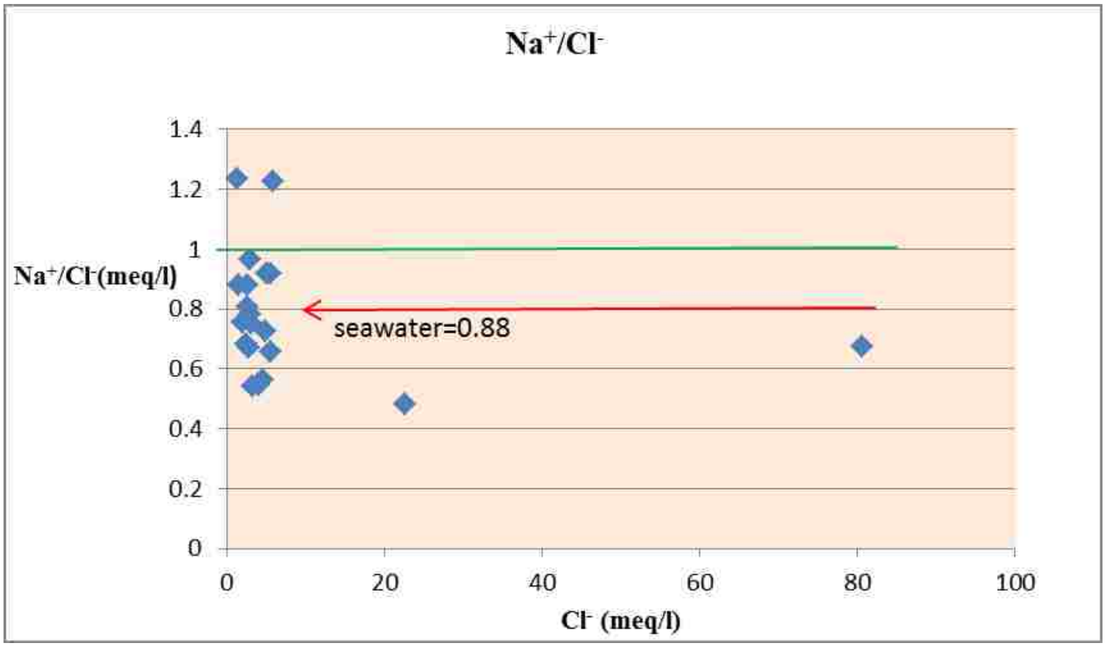


Figure 38: Molar ratio of Na^+/Cl^- vs. Cl^- (meq/l) concentration in GW samples.

Based on the values of 20 groundwater samples of Cl^-/HCO_3^- ratio, only two samples of groundwater were unaffected, 85% of samples were slightly and moderately affected by salinization process, and just one sample (well No.8) was strongly affected by the deep saline water. In addition, using ratio of (Ca^{2+}/Na^+ and Cl^-/HCO_3^-) versus TDS is useful to evaluate the effect of saline water mixing with fresh water and the effect of cation exchange processes (Martens et al., 2011).

The ratio of Ca^{2+}/Na^{+} (Figure 39A) stayed very low for most samples and it had negative correlation with the TDS value. This case might be attributed to the evaporation of rainfall, absorption of Na^{+} by clays, and by return flow of irrigation which is a common process in arid and semi-arid areas (Ghassemi et al., 1995). Moreover, the Cl^{-}/HCO$_3^{-}$ ratio versus TDS values (Figure 39B) shows negative correlation between the two variables except for well No.8, which shows a positive correlation. This result was due to high concentration of Cl^{-} ion caused by deep saline-water mixing.

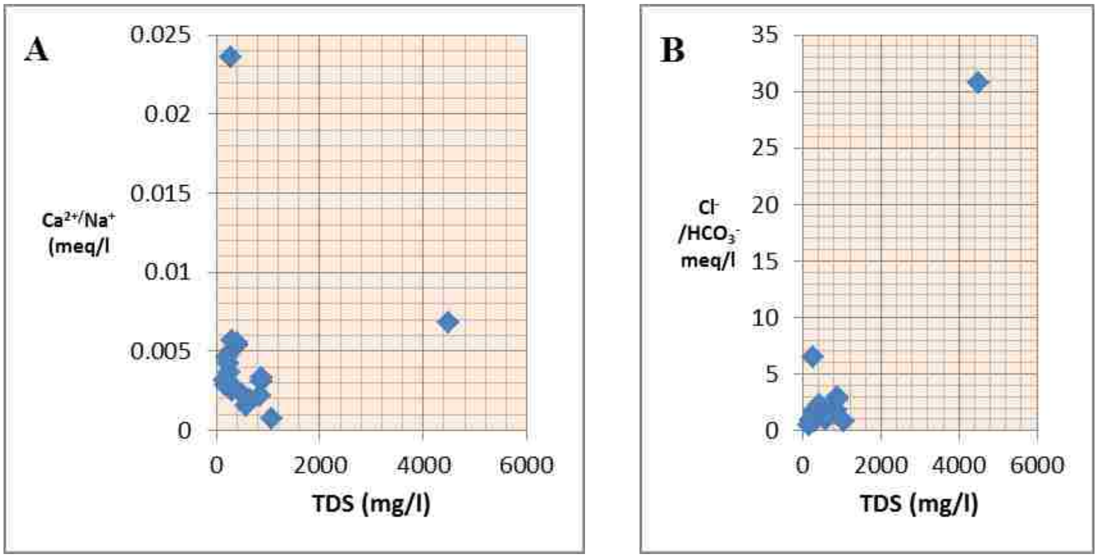


Figure 39 A&B: The ratio of Ca$^{2+/}$Na^{+} and Cl^{-}/HCO$_3$- versus TDS.

Furthermore, the ratio of Ca^{2+}+ Mg^{2+} versus HCO$_3^{-}$shows that the majority of samples (Figure 40) fall above the line (1:1) suggesting an excess of alkaline earth elements (Ca^{2+}+ Mg^{2+} over than HCO$_3^{-}$) and also may be is the fact that reflecting extra sources of Ca^{2+} and Mg^{2+} ions supplied by silicate weathering (Zhang et al., 1995). Minor representations are also observed in bicarbonate ratio which may be due to the reaction of the feldspar minerals with carbonic acid in the presence of water (rainfall), which releases HCO^-_3 (Lakshmanan et al., 2003).

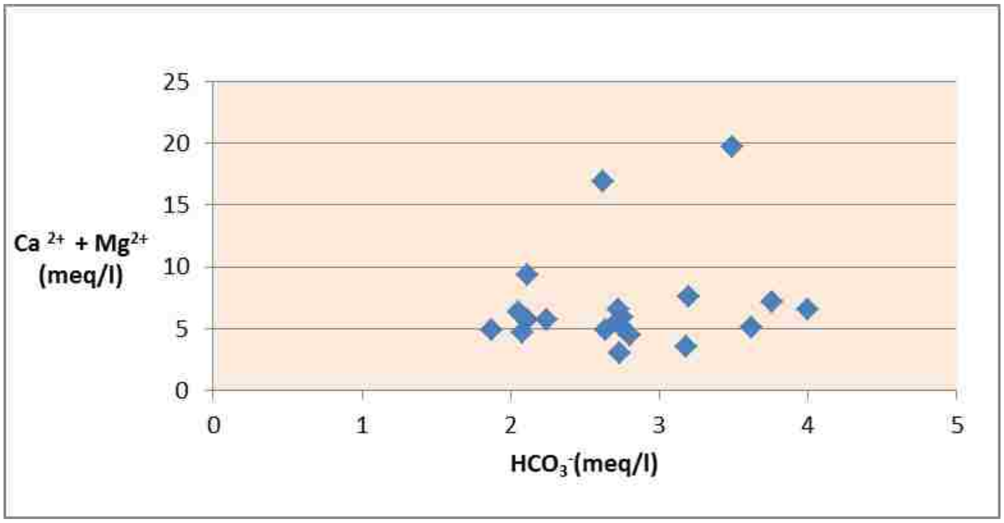


Figure 40: Ratio of Ca^{2+} Mg^{2+} versus HCO_3^- for collected GW samples.

Additionally, the plot of (Ca^{2+}+Mg^{2+}) versus (HCO_3^- + SO_4^{2-}) is used to determine the ion exchange processes (Figure 41). If ion exchange is the process, the points shift to the left side of the plot due to excess of SO_4^{2-} +HCO_3^-, while if reverse ions exchange is the process, the points shift to the right due to excess ofCa^{2+} +Mg^{2+} (Fisher & Mullican, 1997).

According to the results, a total of 20% of groundwater samples which are located in the northeast of the study area fall left of the 1:1 line indicating ion exchange process which may be due to the excess of bicarbonate SO_4^{2-}, and 60% of samples, which are located in the centre and south of the study area, fall right of the 1:1 line indicating excess of calcium and magnesium in groundwater exchanged with sodium from aquifer materials.

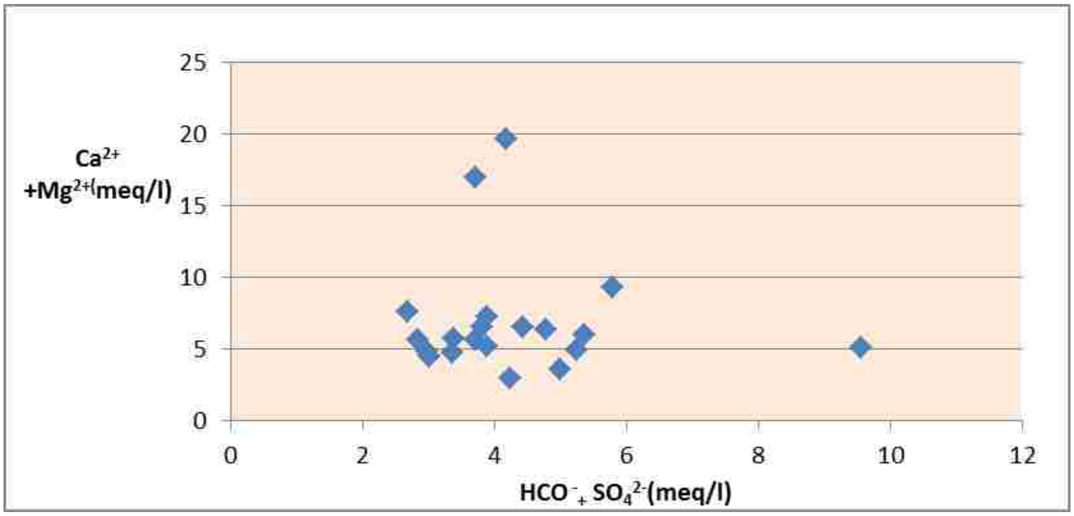


Figure 41: Ratio of ($Ca^{2+}Mg^{2+}$) versus (HCO_3^- + SO_4^{2-}) for GW samples

On the other hand, (Figure 42) shows correlation between NO_3^- versus TDS. Elevated NO_3^- concentrations among the study area from northeast toward southwest of study area is observed, while the negative correlation with TDS can be seen for all samples except well No.8, which has the highest TDS value. The highest NO_3^- concentrations were recorded due to intensive agricultural activity in this zone.

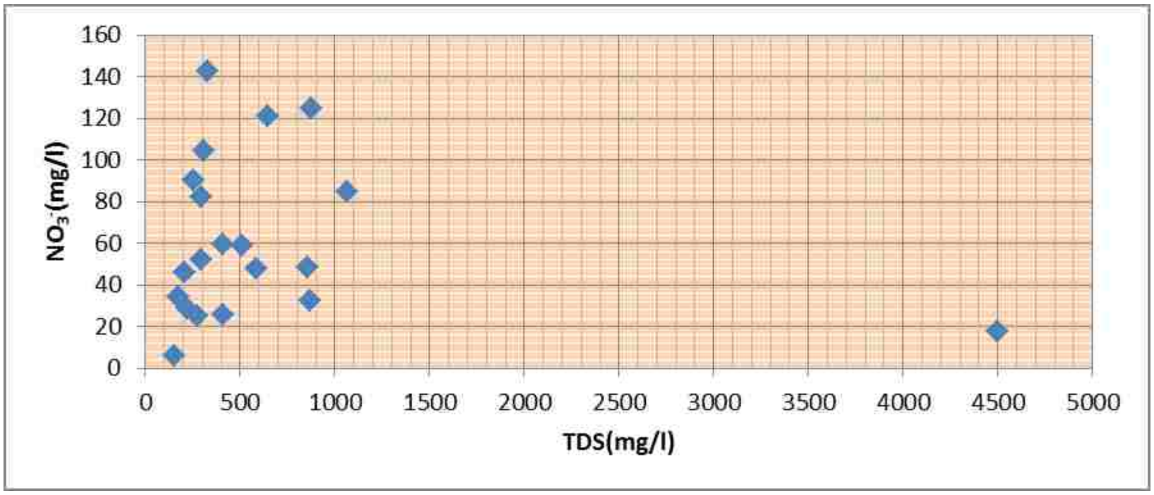


Figure 42: NO_3^- - TDS plot for groundwater samples.

According to the ratio of SO_4^{2-} versus Cl^- (Figure 43) a negative correlation between two variables of most samples can be seen. The possible extra source of SO_4^{2-} is the dissolution of gypsum from the upper aquifer during the recharge event in addition to synthetic fertilizer that is commonly used in the study area.

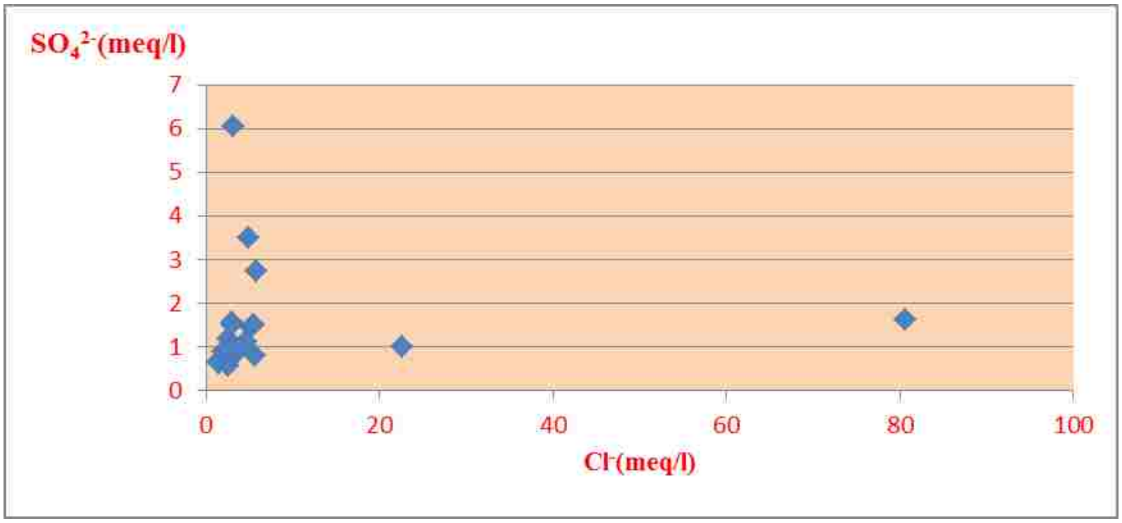


Figure 43: SO_4^{2-} ratio versus TDS relationship for groundwater samples.

5.5. Trace Metals

Heavy metals tend to form insoluble compounds in an alkaline environment, such as that found in limestone. Thus an alkaline aquifer enhances the precipitation of heavy metals (El Gawad et al., 2008). Therefore, a study area which is abundant in this feature of geology and aquifer lithology may affect the concentration of some minerals in the groundwater. Accordingly, about 16 trace elements have been analysed in this study. These were: Aluminium (Al), Arsenic (As), Barium (Ba), Cadmium (Cd), Chromium (Cr), Cobalt (Co), Copper (Cu), Iron (Fe), Lead (Pb), Manganese (Mn), Molybdenum (Mo), Nickel (Ni) Phosphorus (P), Strontium (Sr), Vanadium (V),and Zinc (Zn). (Table 8) shows the results of trace metals concentration from the study area compared with

WHO drinking water standard (2011). The data indicate that low and not significant concentrations of most trace elements are faund in the 20 samples of groundwater in the study area. Abnormal concentrations of the element Barium (Ba) appeared in all samples.

Table 8: Trace elements concentration (mg/l) in GW compared with WHO standards

Trace elements	Maximum value	Minimum value	Average mg/l	WHO Standard (2011)	% wells exceeded The limit	No of wells
Aluminium (Al)	0.025	0.005	0.015	0.1	0%	0
Arsenic (As)	0.105	0.009	0.057	0.01	0%	0
Barium (Ba)	605.588	2.034	303.811	0.7	100%	20
Cadmium (Cd)	0.0055	0.0008	0.0032	0.003	20%	4
Chromium (Cr)	0.034	0.005	0.019	0.05	0%	0
Cobalt (Co)	0.005	0.005	0.005	---	---	---
Copper (Cu)	0.004	0.0003	0.002	2	0%	0
Iron (Fe)	0.107	0.001	0.054	1	0%	0
Lead (Pb)	0.011	0.011	0.011	0.01	0%	0
Manganese (Mn)	0.013	0.001	0.007	0.4	0%	0
Molybdenum (Mo)	0.096	0.005	0.050	0.07	5%	1
Nickel (Ni)	0.022	0.003	0.0125	0.07	0%	0
Phosphorus (P)	0.066	0.012	0.039	---	---	---
Strontium (Sr)	25.284	0.726	13.005	---	---	---
Vanadium (V)	0.012	0.004	0.008	---	---	---
Zinc (Z)	0.551	0.001	0.276	3	0%	0

5.5.1. Barium (Ba)

Organic barium compounds are ionic and are hydrolysed in water. It is a common minor constituent of alkali feldspar (Evans, 1987). The concentration of barium ions in natural aquatic systems is limited by the presence of naturally occurring anions and possibly also by the adsorption of these ions onto metal oxides and hydroxides (Hem, 1985).

Naturally, barium is present as a trace element in both sedimentary and igneous rocks because it is not found free in nature. It occurs in a number of compounds, most commonly barium sulphate (barite) and barium carbonate. Barium in water comes primarily from natural sources. The acetate, nitrate and halides are soluble in water, but the carbonate, chromate, fluoride, oxalate, phosphate and sulphate are insoluble. The solubility of barium compounds increases as the pH level decreases (Organization, 2006).

The highest concentration was detected in well No.8 as 605 mg/l and the lowest one was well No.17 as 2 mg/l as plotted (Figure 44). The highest levels to be found in this well, which has a deep level of brackish water, might be associated with groundwater of low pH. The presence of high concentrations of barium may come from granite-like igneous rocks; alkaline igneous and volcanic rocks and manganese-rich sedimentary rocks (Organization, 2006). Therefore, sedimentary rocks such as limestone are the dominant geologic deposition in the study area, which may lead to a higher concentration of barium.

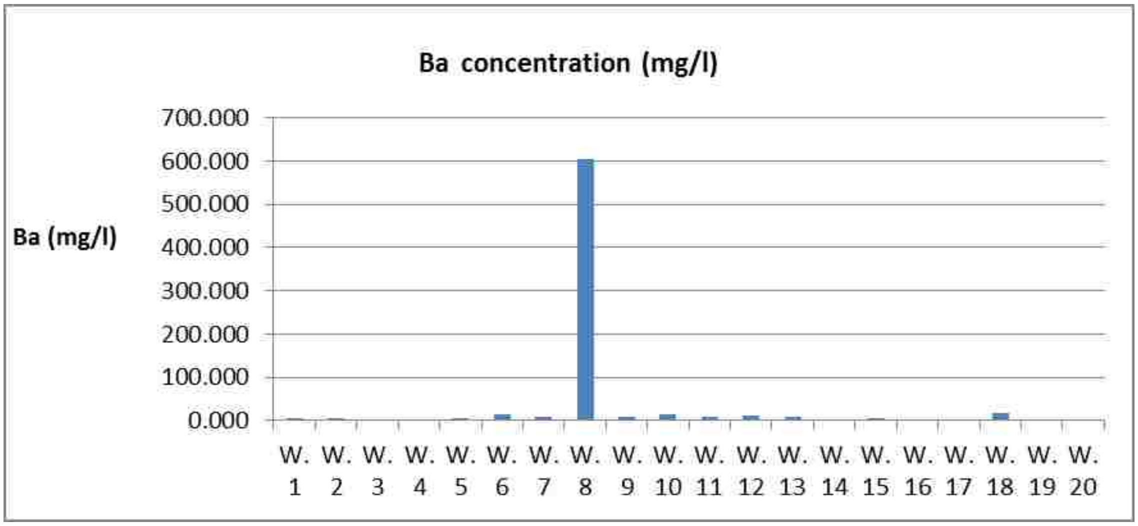


Figure 44: Ba (mg/l) concentration in collected GW samples.

5.5.2. Strontium (Sr)

Strontium (Sr), one of the dissolved polyvalent metallic ions which comes from sedimentary rocks (El Gawad et al., 2008), appeared in moderate average amount concentration of 2.661mg/l. The maximum value is in well No.8 (25mg/l), whereas the lowest concentration was in well No.3 as (0.6) mg/l. The elevated value in sample No.8, which is in the deep well (200 m), may be ascribed to dissolving of limestone rocks, which are dominant in the study area.

5.5.3. Cadmium (Cd)

Cadmium is released to the environment from wastewater. The highest portion of cadmium is caused by contamination from fertilizers. In addition, contamination of the drinking water may be due to impurities in the zinc of galvanized pipes and some metal fittings (Bouchard et al., 2011).

The highest concentration of cadmium was recorded in well No.18 (0.005mg/l) and the lowest value was in well No.19 (0.0008 mg/l) with an average of 0.003 mg/l. There were four wells that exceeded the limit of WHO guideline (0.003mg/l). The toxic concentration of cadmium depends on salinity and hardness (Fetter, 1988). No significant values were observed of the 13 other trace metals (Al, As, Co, Cr, Cu, Fe, Mn, Mo, Ni, P, Pb, V, and Zn).

5.6. Hydrochemical Water Types

Hydrochemical water types are a function of solution kinetics, rock-water interactions, geology and several contamination sources. Hydrochemical facies is used to describe the quantities of water that differ in their chemical composition. This is a convenient method to classify and compare water types that based on ionic composition and ratio of anion and cation (Srinivasamoorthy et al., 2014). The water type is represented by each anion and cation that exceeds 15 equivalent percentages, and then arranged from the lowest to the highest cationic and anionic concentrations in the hydrochemical formula (Altoviski, 1962). The cation distribution indicates that most samples are dominant in mixed cation and magnesium type. The other few samples are lying in the sodium potassium type. In the anion triangle, there is a mixed anion and other samples are lying in the chloride type water, while one sample fell in the bicarbonate field. The diamond shaped part above summarizes that most of the groundwater samples analysed are falling in the field of mixed $MgHCO_3$ and CaCl type of water indicating salinization and cation exchange reaction. Some samples also represent NaCl types, which indicate up coning of deep saltwater.

Piper, (1944) plotted a convenient method to classify and compare water types based on ionic composition. It is proposed as plotting the chemical data on a tri-linear diagram (Figure 45). The plot shows that three main types of water have been identified based on varying ionic concentration. These types of water (Appendix-Table1) are distributed in the study area as the following:

- Mg^{2+}, Bicarbonate type :(samples 1, 3, 12, 13, 14, 15, 16, 17, 19, and 20)
- Mg^{2+}, Chloride type: (samples 2, 4, 5, 9, 11, and 18)
- Na^{+}, Chloride type: (samples 6, 7, 8, and 10)

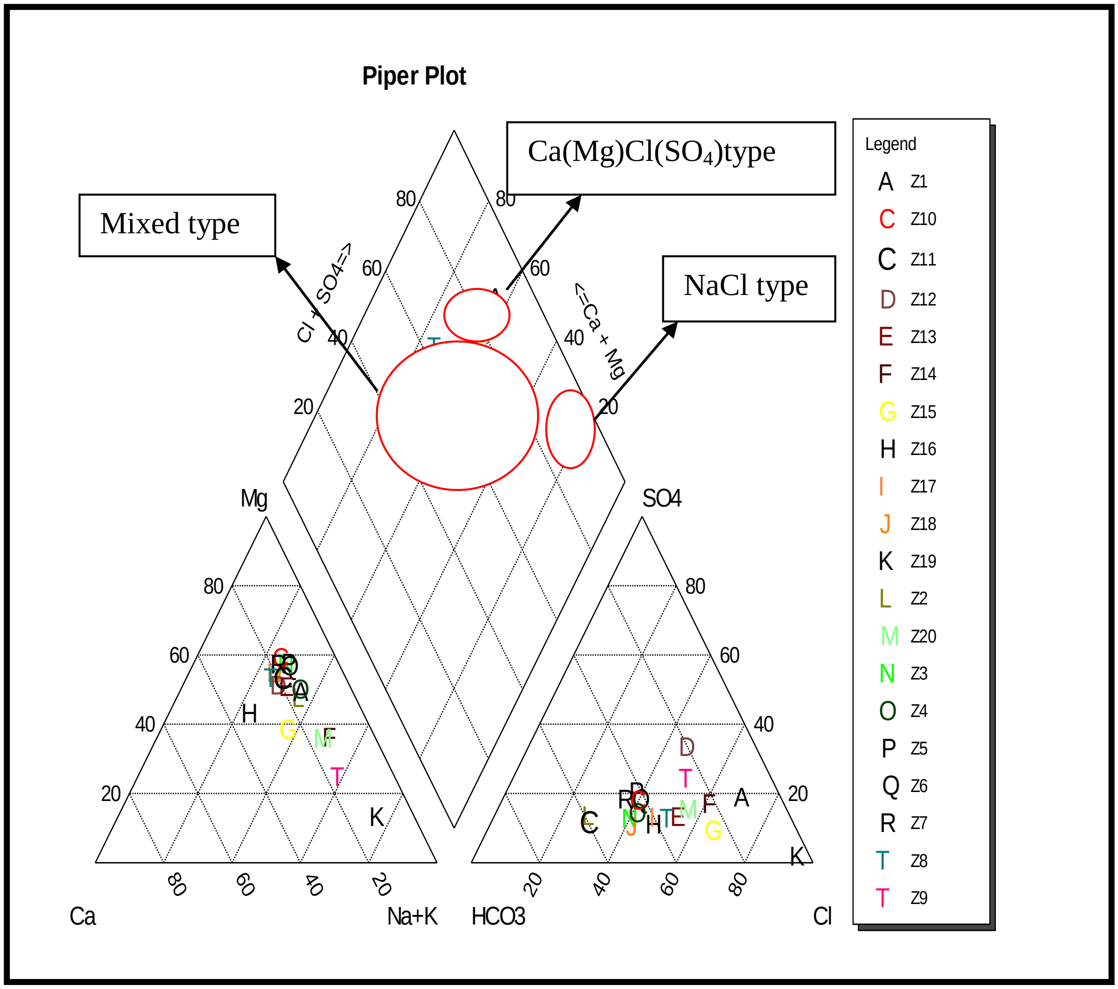


Figure 45: Piper diagram of groundwater samples of study area.

The majority of the groundwater samples (50%) are classified as Mg-HCO_3^- type. This indicates recent recharge by freshwater and water with temporary hardness (Handa, 1979). The existence of bicarbonate anion with higher concentration may indicate a higher groundwater recharge in the eastern and central part of the study area, where the depleted rainwater dissolves the ophiolite rocks that form the catchment areas of Wadi Al Jizi and Mahada mountains. In addition, dissolution of carbonate and dolomite rocks may lead to increase the concentration of magnesium in the groundwater (Murad et al., 2012).

The rest of the water types were classified as MgCl (30%) and NaCl (20%). The presence of these types may be due to up coning of deep brackish water where salinization condition and cation exchange reaction is occurring (Walraevens & Van Camp, 2005). However, there is evidence of that according to the field survey. The farmer in the area found that saline water can be produced if wells are drilled deeper than 150 feet. The three types of water identified in the study area clearly illustrate the water type zonation (Figure 46).

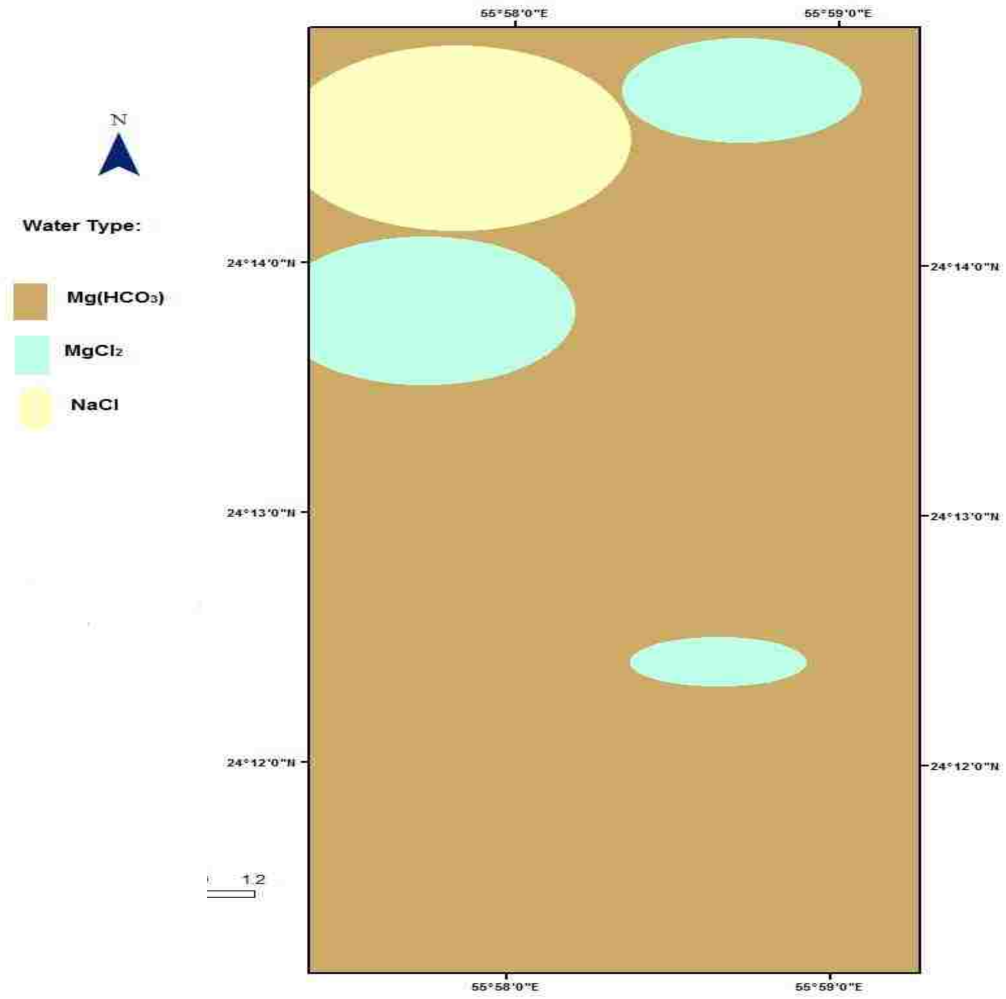


Figure 46: Water type zonation map in the study area.

5.7. Water Origin and Hypothetical Salt Combinations

The calculated results according to Sulin (1948) reveal three water genesis represented by $MgCl_2$, $CaCl_2$, and Na_2SO_4 respectively (Figure 47). As shown in the graphical representation, $MgCl_2$ is the dominant water origin occuping most of the study area while $CaCl_2$ and Na_2SO_4 come in second and third respectively.

The Na_2SO_4 zone reveals the inflowing of meteoric water recharge into the shallow aquifer. The calcium chloride ($CaCl_2$) zone reflects the impact of up-coning of old marine water origin due to severe groundwater exploitation, whereas the appearance of $MgCl_2$ genesis is not recovered and shows the recent marine origin, but it may indicate fresh water richness in magnesium which is coming from the dissolution of magnetite mineral found in the ophiolite rocks which are distributed within the area of study. Salts of permanent hardness represented by $CaSO_4$, $MgSO_4$ appear within few samples while salts of temporary hardness occur among the almost samples and are represented by Ca (HCO_3) and Mg $(HCO_3)_2$.

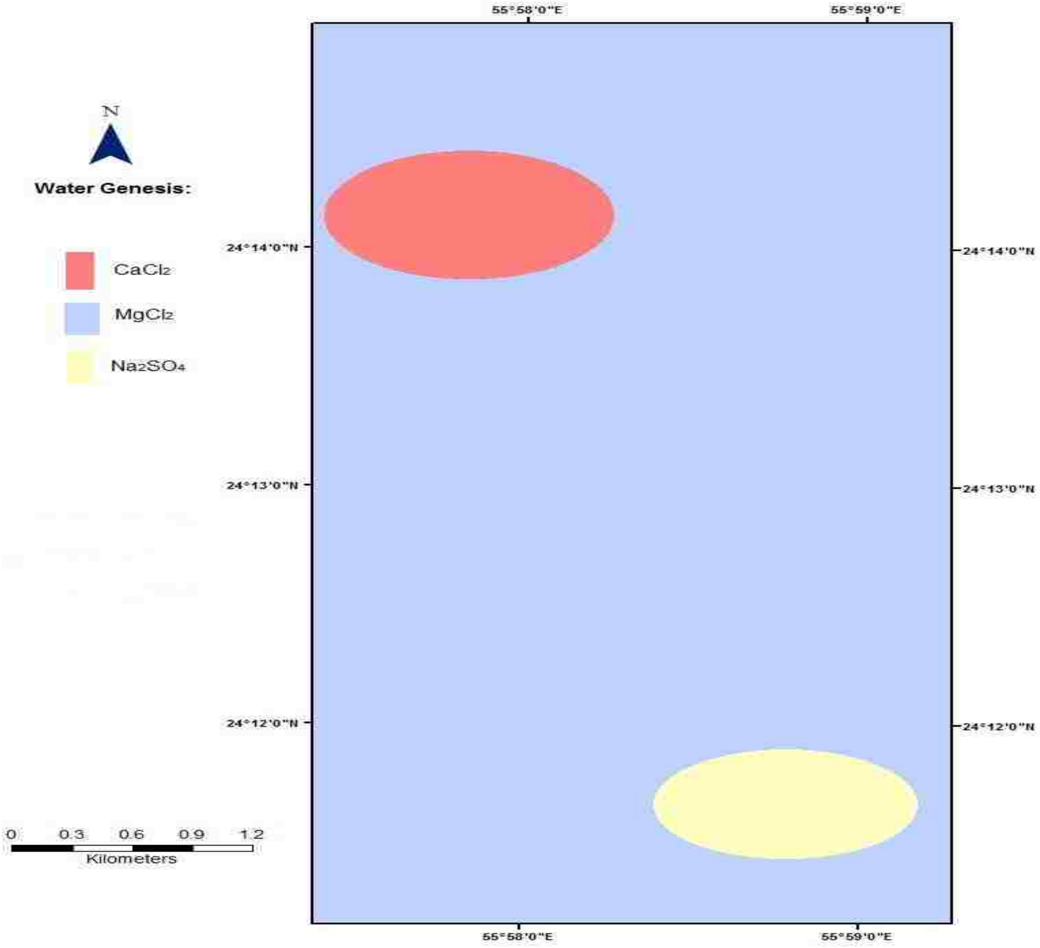


Figure 47 Water genesis zonation map of the study area.

5.8. Irrigation Water Quality

Crop yield and deteriorated soil fertility may be reduced by chemical constituents caused by human activities and natural environment, which are derived from irrigation water pumped from wells. It is also affected by nature of plants irrigated, soil type, climate, and the method of irrigation and drainage.

When the irrigation water reaches the soil, salts introduce into the root zone and plant roots take in water but absorb very little salt from the soil solution. Moreover, water evaporates from the soil surface but salts remain behind. The processes result in a gradual accumulation of salt in the root zone affecting the plants through salt hazards, water deficiency and toxicity. Knowledge of irrigation water quality is critical to understand what management changes are necessary for long-term productivity (Jalali, 2011).

Accordingly, irrigated agricultural crops need very good quality water. It is necessary to have detailed information concerning the quality of irrigation water and its effect on soils and crops. Hence, there are numerous parameters used to define irrigation water quality to assess salinity hazards and determine management strategies. Consequently, these tools will provide visual evaluation (e.g. Charts and Tables) of irrigation water quality.

5.8.1. Sodium Adsorption Ratio (SAR)

Sodium adsorption ratio is expressed as sodium content or alkali hazard. It is important for determining the quality of water used for irrigation purposes. Higher salinity reduces the osmotic activity of plants and prevents water from reaching the branches and leaves of plants resulting in inferior production (Marghade et al., 2011).

Plants are sensitive in varying degrees to soil salinity, and when these exceeding a certain limit; their growth is impaired, thus lowering their productivity. Sodium adsorption ratio (SAR), along with pH, characterizes salt-affected soils. It is an easily measured property that gives information on the comparative concentrations of Na^+, Ca^{2+}, and Mg^{2+} in soil solutions. The SAR is computed using the following formula as (Hem, 1985).

$$\text{SAR} = \frac{Na+}{\sqrt{(Ca+Mg)/2}}$$

$[Na^+]$, $[Ca^{2+}]$, and $[Mg^{2+}]$ are concentration in mill equivalent weight of sodium, calcium, and magnesium ions are in the soil solution. When the SAR exceeds 12, it means serious physical soil problems arise and plants have difficulty absorbing water (Sherif et al., 2012). Accordingly, the SAR values of 20 samples range from 1.44 to 26.4 with an average of 3.72. The analytical data is plotted on the Wilcox Diagram (Figure 48). The majority of water samples in the study area clusters in C2 and C3 S1 zones indicating medium to high salinity and low sodium water, which can be used for irrigation in almost all types of soil with little danger of exchangeable sodium (Kumar et al., 2007). The exceptional case of well No.8, which clusters in C4-S1 zone, indicat very high damaging risk if it is used for irrigation because the higher salinity of this water reduces the osmotic activity of plants and prevents water from reaching the branches and leaves of plants and then decreases productivity and growth (Marghade et al., 2011).

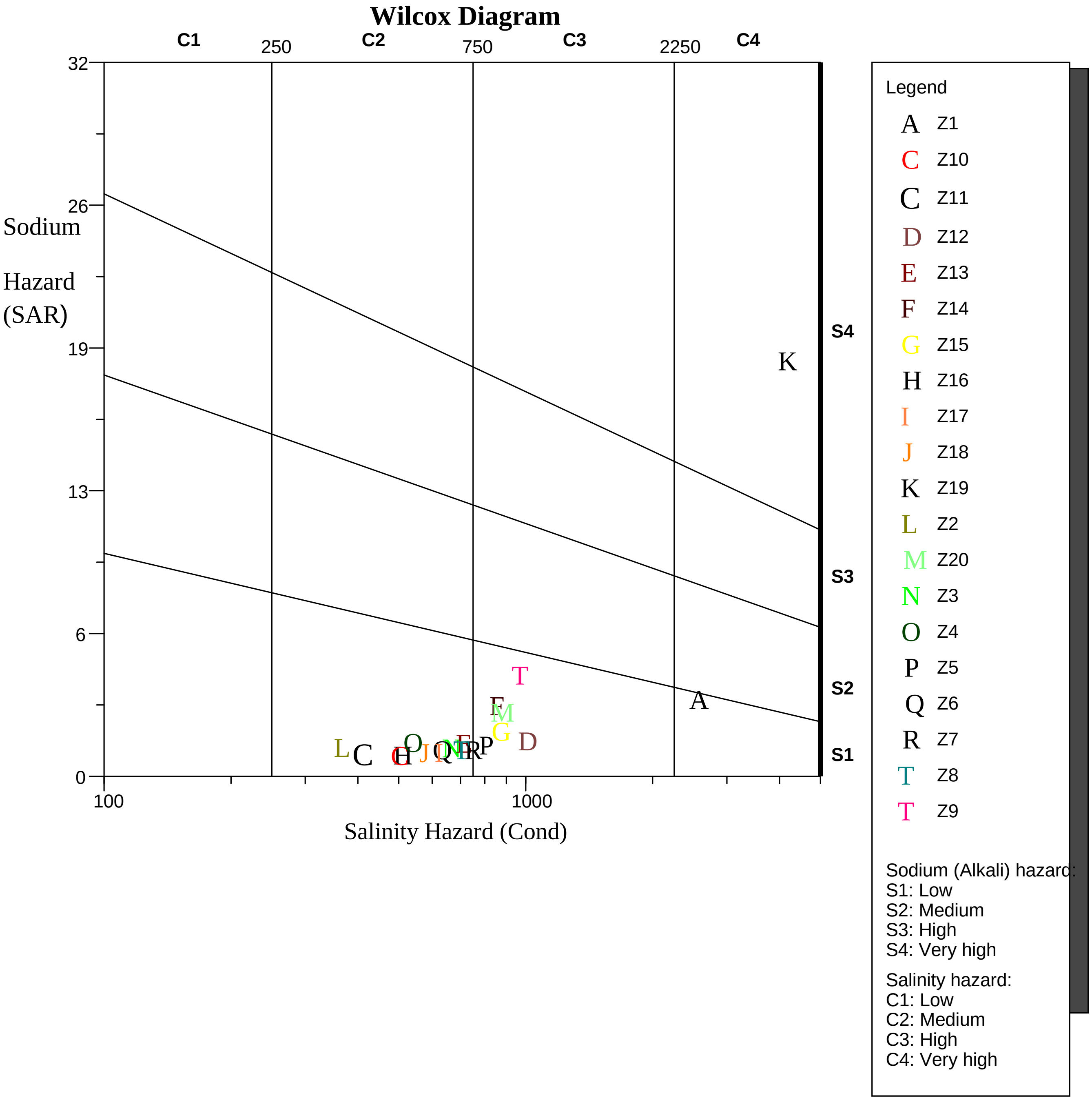


Figure 48: Water classification according to EC and SAR values.

5.8.2. Total Hardness

Srinivasamoorthy et al., (2014) indicated that the precipitation of Ca^{2+} and Mg^{2+} ions cause action of soap in water that affects the temporary and permanent hardness of water. The temporary hardness is mainly due to calcium carbonate in water that is removed during heating, while permanent hardness is due to Ca^{2+} and Mg^{2+} ions that are removed by ion-exchange processes.

Hardness of water limits the uses of water for industrial purposes; causing scaling of pots, boilers and irrigation pipes. In some studies, a significant correlation was observed between hardness and heart diseases. In contrast a number of epidemiological studies suggest that water hardness protects against diseases (WHO, 2006). The total hardness (TH) expressed in mg/l as determined by Todd (1980) is follows TH (mg/l) = 2.497 Ca^{2+}+ 4.115 Mg^{2+}. The class of water according the TH vary from soft water (< 75 mg/l) to very hard water (>300 mg/l) as shown in the (Table 9). The most desirable limit for TH is 80 –100 mg/l $CaCO_3$ (Freeze & Cherry, 1979). The TH in the study area ranges from148.6 mg/l to 984 mg/l with an average of 347 mg/l. Most wells were tending to hard and very hard category.

Table 9: Classification of GW samples according the TH in the study area.

	Rang (mg/l)	Class	No of wells	Percentage
TH	<75	Soft	0	0%
	75 – 150	Moderately hard	1	5%
	150 – 300	Hard	11	55%
	>300	Very hard	8	40%

5.8.3. Magnesium Ratio (MR)

The measure of the effect of magnesium in irrigated water is expressed as the magnesium ratio (MR). This ratio classifis irrigation water to suitable (≤ 50), and unsuitable (≥ 50) (Haritash et al., 2008).

Paliwal (1972) developed an index for calculating the magnesium hazards were the concentration is by mg/l using the following formula:

$$MR = \frac{Mg \times 100}{Ca \times Mg}$$

The MR values in the study area (Table 10) range from 0 to 6.6 indicating 100% of wells fall below the permissible limit of 50 mg/l and suggests a favorable effect on crop yield.

Table 10: Magnesium Ratio (MR) of GW samples in the study area.

MR (mg/l)	Range	Class	No of wells	Percentage
	<50	Suitable	20	100%
	>50	Unsuitable	0	0%

5.8.4. Sodium Percentage (Na^+ %)

Sodium is expressed as percent sodium or soluble-sodium percentage (Na^+ %). It is calculated using the following formula:

$$Na^+ \% = \frac{(Na+)+(K+)}{(Ca2+)+(Mg2+)+(Na+)+(K+)} \times 100$$

Where, all ionic concentrations are expressed in meq/l.

As per the Wilcox, (1955) classification, no well lay within an excellent category where as 80% of groundwater samples represent the good category, 15% represent permissible, 5% represent doubtful and no representation is given an unsuitable limit as shown in (Table 11). The effect of dilution caused recharge events may help to increase in number of samples representing good category, and decrease in doubtful category for irrigation purposes.

Table 11: Na^+ percentage of collected GW samples in the study area.

Na^+ %	Range	Classes	No of wells	%
	< 20	Excellent	0	0%
	20 – 40	Good	16	80%
	40 – 60	Permissible	3	15%
	60 – 80	Doubtful	1	5%
	80	Unsuitable	0	0%

5.8.5. Electrical Conductivity (EC)

According to the international permissible limits for classes of irrigation water which depends on the EC values. There are five categories that evaluate the rate of EC in the water. Concentration of EC for each class indicates a description of water quality for irrigation (James, Hanks, & Jurinak, 1982). EC value varys from- 250 µS/cm to more the 3000 µS/cm- as shown in the (Table 12).

The result of collected groundwater samples show 90% of the wells within the class vary from good to permissible. Well No.18 is classed as doubtful and well No.8 as unsuitable as show in (Figure 49). This may be related to the depth of well No.18

(depth= 200 m) which is characterized by the brackish deep water. In addition, high evaporation rate and nutrient enrichment may be responsible for elevating the EC.

Table 12: Classification of GW quality based on EC.

EC	Range(μs\cm)	Classes of water	No of wells	%
	< 250	Excellent	0	0%
	250 – 750	Good	12	60%
	750 – 2000	Permissible	6	30%
	2000 - 3000	Doubtful	1	5%
	>3000	Unsuitable	1	5%

Green = Good for irrigation
Blue = Permissible for irrigation
Orange= Doubtful for irrigation
Red = Unsuitable for irrigation

Figure 49: Wells classification according to EC values of groundwater samples

5.9. Microbiological Analysis

Duplicate samples from each of the 20 private wells on farms located in the study area were subjected to biological analysis. The Millipore filtration technique was used to detect the number of colonies formed by coliform bacteria and *E.coli* in 100ml of ground water. The data are expressed as cfu/100ml (Figure 50), and Appendix, Table 2.

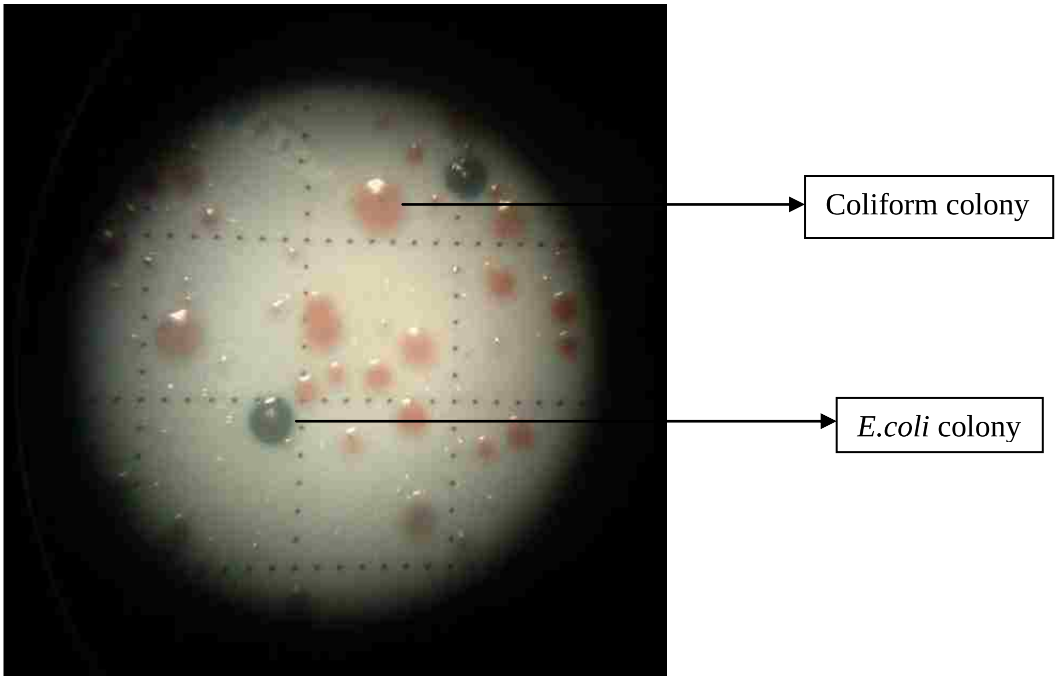


Figure 50: *E. coli* (blue) and coliforms (pink) colonies on cultured plates

5.9.1. Total Coliform Count

The membrane filtration technique was used to test water samples collected from 20 wells in the study area. The results revealed high numbers of coliform bacteria in most of the samples. Well no. 2 had the highest count (2370 cfu/100ml) while well no.16 had the lowest (6 cfu/100 ml).

The contour map (Figure 51) shows the variation in coliform count (cfu/100 ml) in the 20 samples collected from the study area. Overall, the highest counts were identified in wells located in the northeast and northwest of the study area, whereas lower counts were found in the southeast. This result might be attributed to seepage of contaminants into the aquifer due to surface runoff during or after storm events in the different catchments of the Mahdah Mountains, and other wadis that feed the study area.

Furthermore, the intensive use of manure fertilizer (Figure 52) and plant waste on farms that use flood irrigation may drive bacteria and nutrients into the groundwater. Contamination via livestock and the heavy use of manure fertilizer are major contributors to bacterial pollution in groundwater (Ayebo, Plowman, & others, 2006). According to the field survey, wells number 2, 3, 8, 9, and10, which contained high coliform counts, had low water levels. The lower number of counts in samples taken in the Southeastern part of the study area might be attributed to low levels of agricultural activity and greater distances from the runoff areas.

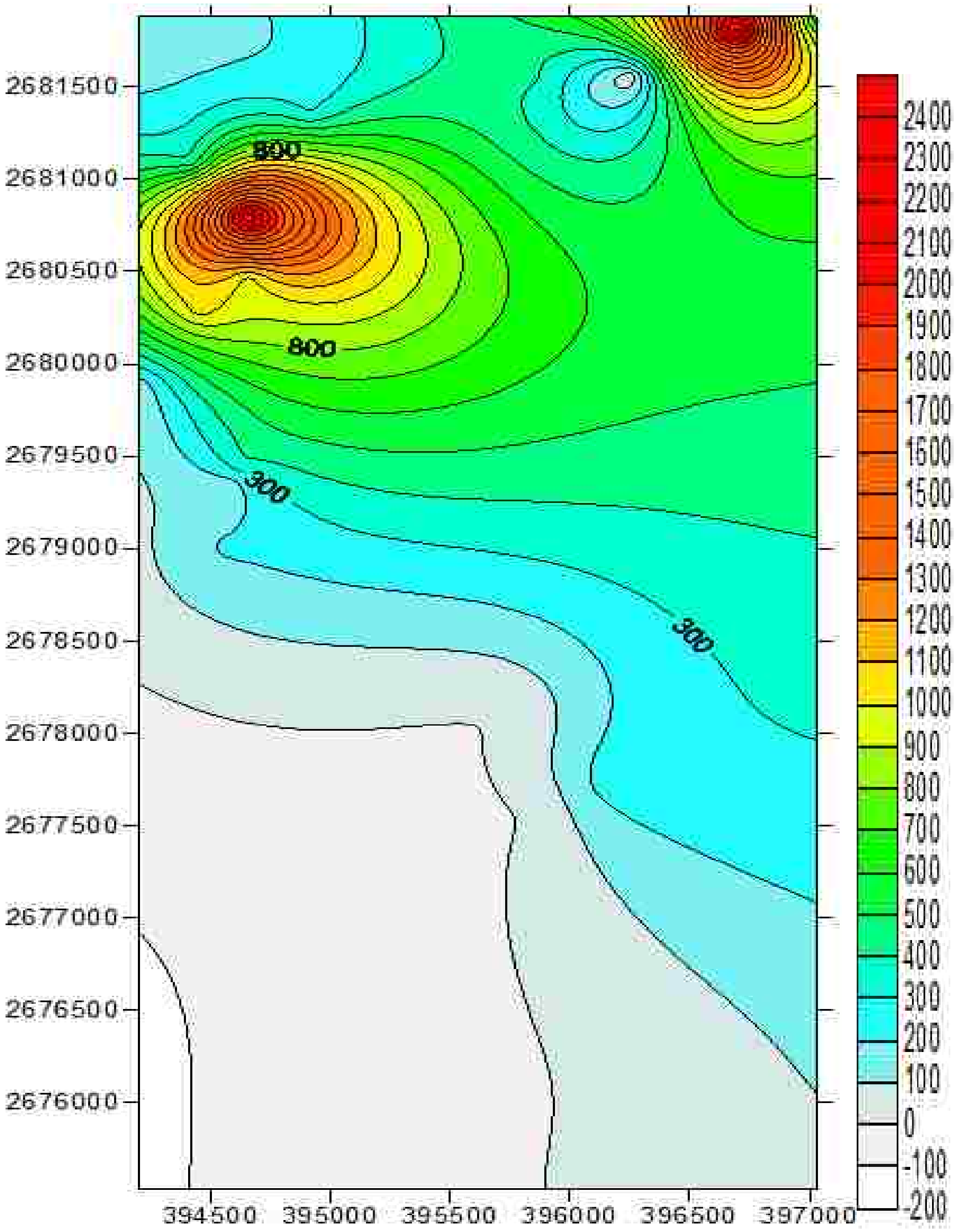


Figure 51: Contour map shows coliform bacteria counts (cfu/100 ml).

Figure 52: Manure fertilizer being used next to water wells.

According to WHO Guidelines (2011), all tested samples exceeded the acceptable level of coliform contamination of drinking water (0 cfu/100ml). However, all samples fell within the acceptable range for irrigation (<10 – 10000 cfu/100 ml). By contrast, samples from 14 wells (70%) did not meet the livestock watering standard (100cfu/100 ml), indicating that the water from these wells is not safe for this purpose (Figure 53) (Sundaram et al., 2009).

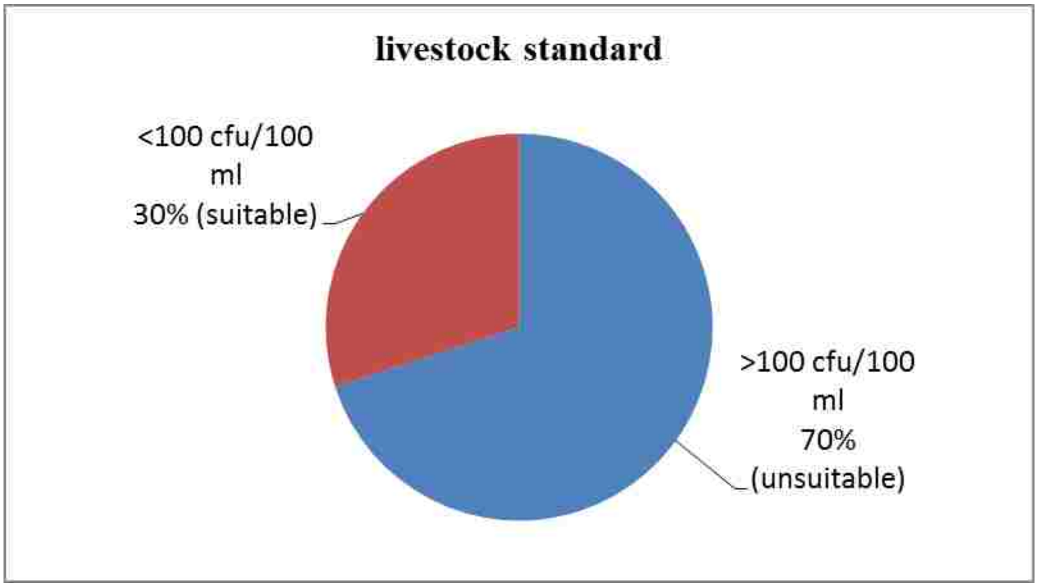


Figure 47: Percentage of GW samples exceeded or within the limits of livestock

5.9.2. Physicochemical Properties and Coliforms Presence Correlation

(Figure 54) showed that there was no statistically significant correlation between any of the physicochemical parameters measured and the number of coliform bacteria (cfu/100 ml). However, there are some positive correlations observed in EC (Regression = 0.4), Cl^- (R = 0.5), Na^+ (R = 0.5) and Ca^{2+} (R = 0.5). These constituents may be providing a helpful environment for the growth and proliferation of the coliform bacteria.

On the other hand, there is a negative correlation that appeared with pH (R = - 0.52), TH (R = -0.17), HCO_3^- (R = - 0.2) and SO_4^{2-} (R = - 0.1). From these results, it could be suggested that acidic environment is not suitable to the coliform bacteria to increas and grow. In contrast, Mg^{2+} (R = 0.05), and NO_3^- (R = - 0.06) showed approximately no correlation versus coliform existence. However, *E.coli* bacteria, which also presented in nine samples, were not significantly correlated to any of phytochemical parameters.

The result indicates that the number of coliform bacteria in the wells was independent of the concentration of anions, cations, and pH. Thus, coliform contamination in most wells may be due to the seepage of surface contaminated runoff or irrigation return flow, into the wells due to the lack of cap-seals as shown in (Figure 55).

Inspection of wells in the study area suggested that farming and agriculture activities, septic tanks, sewage infiltration systems, and sewer leakage from farmer’s houses might all contribute to microbiological contamination of well water. Moreover, in addition to surface runoff and accumulated surface water (pools/ponds) that may enter wells without adequate cap-seals, the delivery pipe from the well to the collection pool may have been a source of contamination.

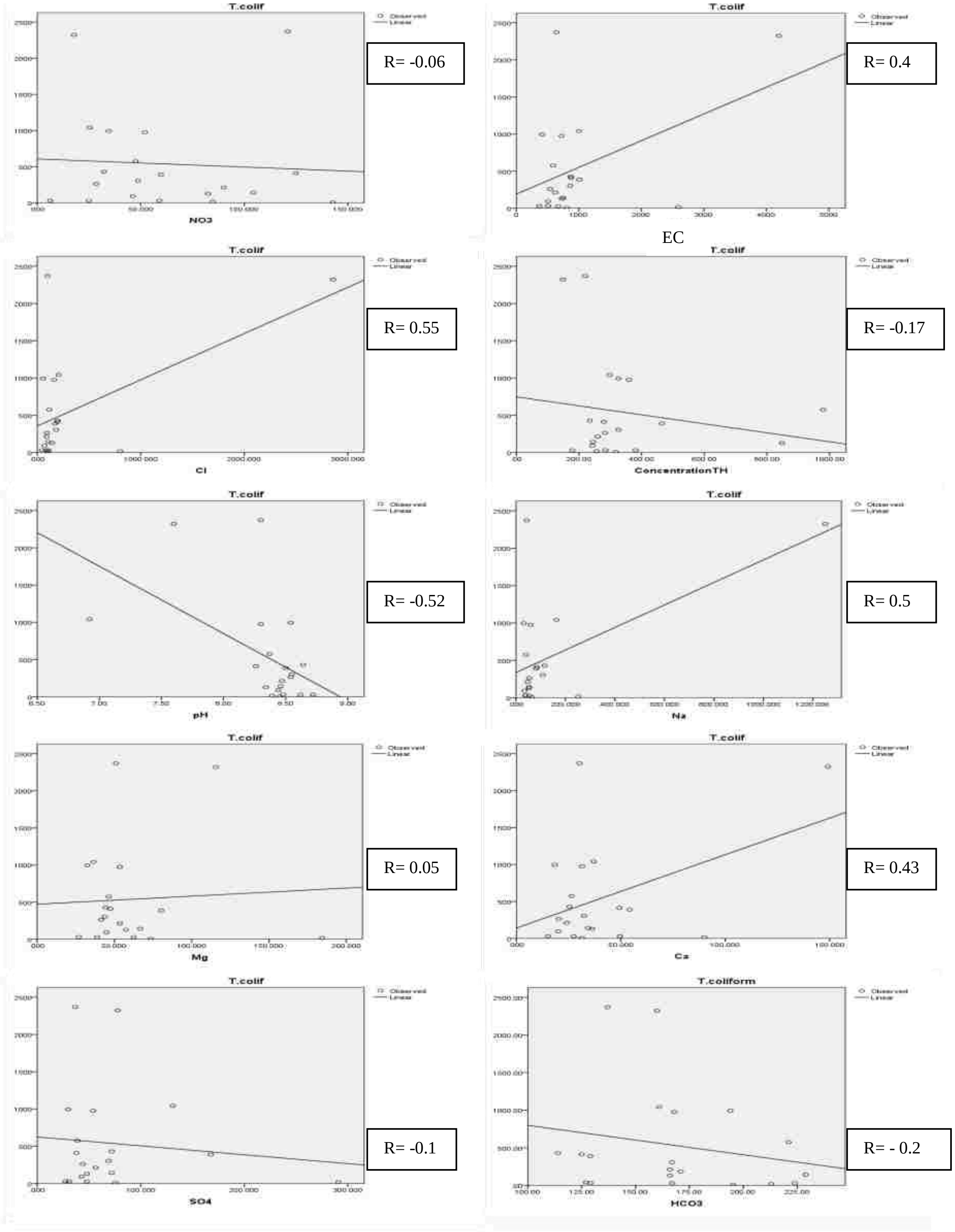


Figure 48: Concentration correlation between total coliform counts and pH/ anions/cations (mg/l).

Figure 495: Inappropriate cap-seals around wells may allow the entry of pollutants.

5.9.3. *E.coli* Bacteria

The twenty samples obtained from the study area tested for the presence of *E.coli* and nine of these twenty samples contained *E.coli.* The highest counts were detected in well No.17 (260 cfu/100 ml) and well No.3 (222 cfu/100 ml).

All other wells contained low counts (ranging from 1 to 82 cfu/100ml) as illustrated in (Figure 56). The presence of *E.coli* in the water wells might be attributed to several sources. The major source may be septic tanks, which are located very close to the wells as shown in (Figure 57). Moreover, the wells are also close to cattle stockades, meaning that manure debris may leach into the groundwater and contaminate the wells.

Figure 50: Geographical distribution of *E.coli*-contaminated wells in the study area.

Figure 51: The farmer's homes are close to the water wells.

5.9.4. *E.coli* Identification and Source Tracking

The eight *E.coli* 16s rRNA sequences were 100% homologous with *E. coli* K-12 (sub-strain MG 1655) (Table 13). This species was first identified in the faeces of healthy individuals in 1885 by *Theodor Escherich*. He called it *Bacterium coli commune* and it resides in the colon.

The K-12 strain was isolated from a stool sample from a convalescent diphtheria patient at Stanford University in 1922 (Bachmann, 1972). The results of the present study show that all of the *E.coli* in the contaminated wells derived from humans or animals sources. This finding is consistent with the local conditions observed at each of the sites. For example, the wells are located closer to accommodation with septic tank facilities and very close to the animal’s stockade. In addition, the wellheads are not completely sealed, which would allow contaminants such as manure fertilizers to enter the well.

Table 13: Closest matches for the eight *E.coli* isolates (Blast).

Isolate	Nearest relative	Accession no. for nearest relative	Percent sequence identity
All 8 isolates	Complete Escherichia coli K-12 (substr. MG 1655 strain) 16S ribosomal RNA sequence	NR 102804.1	100%

5.9.5. Sequence Alignment

The 16S rRNA sequences were aligned and analyzed using Clustal (v.2.1). All eight sequences aligned perfectly suggesting that all originated from the same species. Table.14 shows the alignment of the 16S rRNA sequences identified in this study.

Table 14: Shows the alignment of the 16S rRNA sequences identified in this study

```
1          GCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG
60
2          GCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG
60
3          GCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG
60
4          CGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG 59
5          GCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG
60
6          GCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG
60
9          GCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG
60
7          GCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTG
60
           ************************************************************

1          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
120
2          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
120
3          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
120
4          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
119
5          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
120
6          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
120
9          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
120
7          GCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTT
120
           ************************************************************

1          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
180
2          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
180
3          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
180
4          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
179
5          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
180
6          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
180
9          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
180
7          ACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGC
180
           ************************************************************

1          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
240
2          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
240
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3          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
240
4          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
239
5          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
240
6          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
240
9          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
240
7          ATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAG
240
           ************************************************************

1          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
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2          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
300
3          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
300
4          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
299
5          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
300
6          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
300
9          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
300
7          TCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGT
300
           ************************************************************

1          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
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2          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
360
3          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
360
4          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
359
5          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
360
6          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
360
9          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
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7          CGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCA
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           ************************************************************

1          AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
420
2          AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
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3          AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
420
4          AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
419
5          AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
420
```

```
6           AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
420
9           AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
420
7           AGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTC
420
            ************************************************************

1           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
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2           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
480
3           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
480
4           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
479
5           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
480
6           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
480
9           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
480
7           CAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCG
480
            ************************************************************

1           TCAGC 485
2           TCAGC 485
3           TCAGC 485
4           TCAGC 484
5           TCAGC 485
6           TCAGC 485
9           TCAGC 485
7           TCAGC 485
```

Chapter 6: Conclusion and Recommendation

6.1. Conclusion

The Sultanate of Oman comprises arid and semi-arid areas depending on the amount of rain during the year, although the Al-Buraymi Governorate in the north of Oman is one of the driest regions in the proximity of the Rub’ al Khali.

The people that live in the Al-Buraymi Governorate rely largely on groundwater as their main source of water for drinking, agriculture, and industrial use. The increase in the number of residents, businesses, industries, and agricultural activity in this city has led to the consumption of large amounts of groundwater from local wells in the region, and has had a significant impact on water quality.

The current study aimed to examine the quality of the groundwater in the wells and monitor faecal contamination to allow the implementation of improved management strategies. In all, water samples from 20 wells were analyzed both chemically and biologically.

The results showed that the TDS in the samples ranged from 50 ml/g to 4500 ml/g. The increased salinity of some of the samples (from the northeast of the region) may be the result of high evaporation, increasing agricultural activity, and the mixing of fresh water with salt water in deep aquifers. The majority of water samples in the study area clustered in the C2 and C3 S1 zone, indicating medium-to-high salinity and low sodium content. Almost all soil types with little or no danger of exchangeable sodium could use the water in the S1 zone for irrigation purposes.

Furthermore, coliform bacteria were identified in all water samples (ranging from 6 cfu/100 ml to 2370 cfu/100 ml) and *E. coli* was identified in nine samples. The presence of these bacteria means that none of the studied wells contain water that is safe to drink for human (according to WHO Guidelines 2011), whereas the water in 70% of the wells is not suitable for livestock. However, the water in all of the wells was suitable for irrigation.

This study identified *E.coli* in nine of the twenty wells. Biological analysis of eight *E.coli* isolates showed that contamination was due to a single *E.coli* species, which was first isolated from the human colon. This implies that the major source of faecal contamination comes from on-site septic facilities and/or cattle (on-site animal houses and manure fertilizers).

In conclusion, the results of the present study show that the water from private wells in the Al-Zoroup area is not safe to drink without treatment; however, it is suitable for irrigation. Therefore, the following recommendations should be implemented.

6.2. Recommendations

- The responsible government in each area should carry out periodic inspections of the water wells during the year.
- Use automatic pumps to avoid over-pumping.
- Put signs up which state that the water is unsuitable for drinking.
- The municipality or public health workers should teach farmers how to make well water safe for drinking.

- The size of agricultural areas could be reduced to suit the amount of water available in the well, and to avoid crops that require large amounts of water.
- The septic tanks could be placed away from the wells and checked for leakage into the aquifer.
- The use of fertilizers should be regulated so their use does not affect the quality of the well water.
- Avoiding flood irrigation by installing modern irrigation systems that require less amount of water.
- Recharge dams can be built and underground streams inspected to ensure that they are free from contaminants.
- No animals near wells.
- Avoid the use of landfills sited close to wells for agricultural waste.
- Avoid the use of water refrigerators placed outside the farm because the water is from the wells.
- Cap the wells tightly so that runoff or surface irrigation water cannot enter.

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Appendices

Table 1A: Result of chemical parameter analyses in mg/l of GW samples

Well No	**Sample ID**	**Location**	**X**	**Y**	**pH**	**Temp °C**	**EC. mS/cm**	**TDS**
W 1	231\839	Al-Zoroup	397032	2681882	8.37	33.5	587	585
W 2	336\881	Al-Zoroup	396695	2681806	8.3	32.3	644	643
W 3	332\132	Al-Zoroup	396818	2681377	8.54	33.1	414	170
W 4	231\721	Al-Zoroup	396256	2681552	8.48	39.8	509	509
W 5	334\513	Al-Zoroup	395854	2681545	8.26	33.3	876	875
W 6	336\862	Al-Zoroup	394919	2681355	8.55	34	862	857
W 7	231\519	Al-Zoroup	394421	2681094	8.64	31.2	870	871
W 8	232\011	Al-Zoroup	394657	2680796	7.6	29.6	4200	4200
W 9	231\990	Al-Zoroup	394659	2680469	8.3	27	724	297
W 10	336\926	Al-Zoroup	394455	2680288	6.92	34.9	998	410
W 11	337\990	Al-Zoroup	394199	2679883	8.34	31.5	725	297
W 12	231\956	Al-Zoroup	394628	2679472	8.5	31.5	1007	412
W 13	231\972	Al-Zoroup	394581	2679342	8.46	32.5	751	308
W 14	231\967	Al-Zoroup	394232	2679059	8.44	31.7	505	207
W 15	231\958	Al-Zoroup	394551	2679009	8.47	31.4	622	256
W 16	231\889	Al-Zoroup	395762	2678141	8.45	29.4	805	331
W 17	231\869	Al-Zoroup	396170	2677751	8.54	30.2	541	222
W 18	231\862	Al-Zoroup	395868	2677561	8.39	31.1	2590	1062
W 19	222\475	Al-Zoroup	396405	2676095	8.72	31.3	370	152
W 20	222\546	Al-Zoroup	396200	2675528	8.62	30.2	668	274

Table 1B: Results of major cation concentration in GW samples

Well. No	**Mn**	**Pb**	**Fe**	**Sr**	**Cu**	**Ba**	**Al**	**As**
W. 1	0.002	0.011	0.017	0.979	0.001	5.428	0.006	0.009
W. 2	0.001	0.011	0.019	1.106	0.002	5.829	0.013	0.009
W. 3	0.003	0.011	0.017	0.678	0.003	2.943	0.006	0.009
W. 4	0.008	0.011	0.017	0.946	0.002	3.139	0.012	0.009
W. 5	0.001	0.011	0.003	1.920	0.003	6.863	0.009	0.009
W. 6	0.001	0.011	0.017	1.681	0.001	13.701	0.006	0.009
W. 7	0.002	0.011	0.004	1.710	0.003	8.460	0.014	0.009
W. 8	0.013	0.011	0.107	25.284	0.003	605.588	0.025	0.009
W. 9	0.003	0.011	0.016	1.280	0.003	8.612	0.007	0.009
W.10	0.006	0.011	0.017	2.189	0.002	13.501	0.01	0.105
W.11	0.001	0.011	0.017	1.295	0.0003	8.352	0.008	0.064
W.12	0.004	0.011	0.002	1.940	0.003	12.265	0.015	0.009
W.13	0.002	0.011	0.013	1.310	0.002	9.794	0.007	0.009
W.14	0.001	0.011	0.017	0.899	0.003	3.833	0.01	0.009
W.15	0.001	0.011	0.002	1.140	0.003	5.391	0.005	0.084
W.16	0.001	0.011	0.017	1.531	0.003	3.848	0.019	0.009
W.17	0.002	0.011	0.017	0.949	0.003	2.631	0.006	0.065
W.18	0.003	0.011	0.017	4.216	0.003	17.070	0.025	0.009
W.19	0.001	0.011	0.005	0.726	0.003	2.034	0.007	0.009
W.20	0.001	0.011	0.001	1.445	0.004	3.975	0.006	0.009

Table 1C: Results of trace elements concentration in GW

W. No	Cd	Cr	CO	Ni	Mo	P	V	Zn
W. 1	0.001	0.020	0.005	0.007	0.018	0.030	0.005	0.024
W. 2	0.001	0.020	0.005	0.003	0.009	0.012	0.009	0.090
W. 3	0.0012	0.016	0.005	0.003	0.014	0.066	0.004	0.028
W. 4	0.0021	0.015	0.005	0.003	0.011	0.012	0.005	0.423
W. 5	0.001	0.012	0.005	0.008	0.009	0.012	0.009	0.034
W. 6	0.001	0.030	0.005	0.010	0.007	0.012	0.006	0.038
W. 7	0.001	0.030	0.005	0.003	0.025	0.012	0.010	0.016
W. 8	0.0038	0.007	0.005	0.003	0.018	0.012	0.006	0.551
W. 9	0.001	0.007	0.005	0.007	0.018	0.012	0.005	0.155
W. 10	0.0032	0.005	0.005	0.022	0.012	0.012	0.007	0.001
W. 11	0.0028	0.026	0.005	0.003	0.018	0.012	0.004	0.031
W. 12	0.0051	0.032	0.005	0.003	0.005	0.012	0.005	0.133
W. 13	0.0022	0.014	0.005	0.005	0.018	0.012	0.007	0.069
W. 14	0.001	0.013	0.005	0.016	0.009	0.012	0.005	0.037
W. 15	0.001	0.015	0.005	0.003	0.026	0.012	0.006	0.001
W. 16	0.001	0.019	0.005	0.003	0.015	0.032	0.007	0.066
W. 17	0.0028	0.011	0.005	0.003	0.028	0.012	0.004	0.063
W. 18	0.0055	0.016	0.005	0.005	0.033	0.012	0.005	0.025
W. 19	0.0008	0.009	0.005	0.006	0.040	0.012	0.009	0.035
W. 20	0.001	0.034	0.005	0.003	0.096	0.012	0.012	0.015

Table 1D: Results of major anion concentration in GW samples

Well No.	**Cl**	**SO_4**	**HCO_3**	**CO_3**	**NO_3**
W. 1	115.448	38.736	221	<1	47.573
W. 2	99.29	36.443	137	<1	120.982
W. 3	55.027	29.936	194	30	34.503
W. 4	87.077	27.321	129	7.5	58.97
W. 5	197.01	37.83	125	<1	124.845
W. 6	181.957	68.922	167	15	48.696
W. 7	194.264	72.014	114	7.5	32.181
W. 8	2858.46	77.794	160	<1	17.724
W. 9	164.147	54.084	168	4.5	52.015
W. 10	206.373	130.93	161	<1	25.4
W. 11	144.345	48.016	166	7.7	82.506
W. 12	171.708	167.682	129	10.5	59.773
W. 13	104.912	71.933	229	22.5	104.3
W. 14	70.339	42.734	127	16.5	46.169
W. 15	90.665	56.427	166	15	90.0
W. 16	102.992	75.42	195.2	30	142.8
W. 17	93.454	44.015	171	11.2	28.5
W. 18	801.354	291.056	213	21	84.804
W. 19	46.9	30.626	167	11	6.204
W. 20	110.958	48.008	224	12	25.0

Table 1E: Results of ionic rations in GW samples

W. No.	SAR	TH	Na^+	K^+	Mg^{2+}	Ca^{2+}
W.1	1.55	255.83	40.56	2.813	46.29	26.406
W.2	1.569	284.60	43.08	3.033	51.02	30.177
W.3	1.44	179.00	31.42	2.437	32.48	18.290
W.4	1.44	283.24	38.52	2.674	38.86	49.536
W.5	2.9	317.59	84.24	3.652	47.40	49.306
W.6	4.13	260.17	108.48	4.049	43.73	32.427
W.7	4.53	244.72	115.53	4.413	44.22	25.424
W.8	26.4	848.1	1249.91	13.186	115.64	149.206
W.9	2.14	297.69	60.17	3.355	53.45	31.463
W.10	6.47	242.50	163.89	4.513	36.49	37.000
W.11	1.74	325.68	51.13	3.245	57.34	36.327
W.12	2.299	464.73	80.57	3.707	80.28	54.178
W.13	1.72	359.63	53.17	3.082	66.77	34.312
W.14	1.38	234.09	34.43	2.438	44.88	19.991
W.15	1.74	278.94	47.36	2.822	53.44	24.008
W.16	2.034	380.10	64.54	3.307	73.53	31.468
W.17	2.199	220.49	53.22	2.760	41.43	20.346
W.18	4.92	979.65	251.18	5.874	184.44	90.118
W.19	1.89	148.20	37.56	2.386	26.96	15.091
W.20	1.949	325.84	54.12	3.210	62.39	27.512

Table 2: Coliform bacteria and *E.coli* colonies (cfu/100ml) in 20 samples.

Well No.	**Date of sampling**	**Total coliform**	***E.coli***
W.1	08\04\2013	575	2
W.2	08\04\2013	2370	0
W.3	01\04\2013	995	222
W.4	08\04\2013	31	0
W.5	08\04\2013	412	0
W.6	08\04\2013	305	0
W.7	08\04\2013	430	82
W.8	08\04\2013	2323	23
W.9	08\04\2013	976	0
W.10	01/04/2013	1042	2
W.11	01/04/2013	129	0
W.12	01/04/2013	389	41
W.13	01/04/2013	143	0
W.14	01/04/2013	93	0
W.15	01/04/2013	213	0
W.16	26/03/2013	6	0
W.17	26/03/2013	264	260
W.18	26/03/2013	17	0
W.19	26/03/2013	31	65
W.20	26/03/2013	29	1

Table 3: 16SrRNA sequences of nine samples of *E.coli* bacteria in GW samples.

Sample No	16SrRNA sequences
1	AGGCGGGCGGTGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAAGAGGGCTTTTCTTTCCACAGC
2	GGGCGGGGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAAAACTTTTTTCTTTTCCAAG

3	CCGGCGGGTGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAACGGGCTTTTCTCTTCCACAGGA
4	GGGCGTGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAAGAGGGCTTTTCCTTCCAAA
5	CGGCGTGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCT

	GGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAACGGCTTTTTCTTCCACAG
6	GGGCGGTGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAAGAGGCTTTTCTTTCCACA
7	CCGGCGGGGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCTCTTTGGTCTTGCGACGTTATGCGGT

	ATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAAGAGGCTTTTCTTTCCACA
8	AAACGTTGTCGACGGCAGGGCTGGGTAGCGATCGCTTTCGCCCAGTATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGGTGTGGCTGGTCATCCCTCTTCAGACCAGGCTAGGGATCGTTCGCCCCTTATGGTGGAGGCCCGGTTTACCCCCCACCCTAACTAGGCTAAATTCCCCATTCTTGGGGGCCAACAATTCCGGAATGGCCAAGAAGGCCTCGGGAAGAGGTCCCCCGCTCTTTGGTACTTCGCGCGGACGGGTTATTGCCGGAATATAAGGCCTACCCGG
9	CGGGCGGTGCGCTTTACGCCCAGTAATTCCGATTAACGCTTGCACCCTCCGTATTACCGCGGCTGCTGGCACGGAGTTAGCCGGTGCTTCTTCTGCGGGTAACGTCAATGAGCAAAGGTATTAACTTTACTCCCTTCCTCCCCGCTGAAAGTACTTTACAACCCGAAGGCCTTCTTCATACACGCGGCATGGCTGCATCAGGCTTGCGCCCATTGTGCAATATTCCCCACTGCTGCCTCCCGTAGGAGTCTGGACCGTGTCTCAGTTCCAGTGTGGCTGGTCATCCTCTCAGACCAGCTAGGGATCGTCGCCTAGGTGAGCCGTTACCCCACCTACTAGCTAATCCCATCTGGGCACATCCGATGGCAAGAGGCCCGAAGGTCCCCCTCTTTGGTCTTGCGACGTTATGCGGTATTAGCTACCGTTTCCAGTAGTTATCCCCCTCCATCAGGCAGTTTCCCAGACATTACTCACCCGTCCGCCACTCGTCAGCAAAGAACGGCTTTTCTTTCCAAA