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Microbiology of Water in Wadi Al-Arrroub Drainage Basin

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Dedication

To

My parents who light up my life

My brothers and sisters for their support and help

My in laws for their encouragement

My beloved wife for her patience and endless encouragement
And my lovely daughter “Jana”

All my teachers and friends with respect

Declaration

I certify that this thesis submitted for the degree of Master is the result of my own research, except where otherwise acknowledged, and that this thesis (or any part of the same) has not been submitted for a higher degree to any other university or institution.

Signed: ……

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Abstract

Water resources in Palestine are threatened by many dangers; overexploitation of the Palestinian groundwater by the Israelis and pollution of groundwater by wide spread Israeli settlements on Palestinian land. These factors, along with poor awareness of protection of the environment by the Palestinians, seriously threaten the Palestinian water resources. From the public health perspective, this increases the level of water pollution which results in many health risks.

The study of the microbiology of a water source is an important aspect in evaluating the quality of water. The global burden of infectious waterborne disease is considerable. Reported cases highly underestimate the real incidence of worldwide waterborne diseases, in this regard Palestine is no exception.

Several studies in the West Bank dealt with the microbiology of water and pointed to the poor quality of water used for human consumption. These studies examined few microbiological indicator bacteria in cisterns, dug wells and swimming pools. One recent study (Atteyeh, 2007) examined the presence of DNA of *Helicobacter pylori* and other pathogens (presence of DNA rather than viable bacteria) in three Palestinian water resources, one of which is Wadi Al-Arroub.

In this study the microbiology of a water basin in a heavily populated area of the West Bank, namely Wadi Al-Arroub was examined. The study focused on groundwater samples from 9 deep wells and 11 springs and identified several indicator bacteria, several bacterial pathogens, two protozoan pathogens, and nitrate level.

The study was conducted in the period between May 2005 and January 2006 on water samples from Wadi Al-Arroub drainage basin in Palestine. The study catchment with an area of 61 km^2 is a sub-basin of the Dead Sea-Jordan River Basin and part of the Eastern Basin of the Mountain Aquifer.

The objective of this study is to provide information about Wadi Al-Arroub drainage catchment by identifying the different microbiological pollutants, their possible sources and the impact on water resources, and to highlight possible measures to improve the situation.

Total coliform (TC), Faecal coliform (FC), Faecal streptococci, Total Viable Count, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Entameoba histolytica*, *Giardia intestinalis*, and nitrate (NO_3) level were analyzed for ground water resources.

Eighty five samples were collected; the TC bacteria isolated were identified in 17 (52%) out of 33 well water samples and 33 (94%) out of 35 spring water samples. Samples analyzed for FC were not detected in well water samples, but 35 (80%) out of 44 spring water samples had FC. This indicates that spring water is highly contaminated with wastewater infiltration from cesspits and stream wastewater. Faecal streptococci were detected in 5 (12%) out of 41 well water samples and 38 (86%) out of 44 spring water samples. *Staphylococcus aureus* was identified in 9 (22%) out of 41 well water samples and 36 (82%) out of 44 spring water samples. *Pseudomonas aeruginosa* was detected in 3 (7%) out of 41 well water samples and 8 (18%) out of 44 spring water samples.

The Total coliform bacterial isolates were differentiated into *Enterobacter, Klebsiella, Citrobacter, Escherichia coli* and Faecal coliform and were identified at 48 %, 9%, 12%, 15% and 0% respectively in groundwater well samples. And were identified at 46%, 54%, 46%, 71%, and 80% respectively in spring water samples. This indicates more serious spring water contamination. The identification of *E. coli* in 5/33 (15%) of well water samples and in 25/35 (71%) of spring water samples is an indication of faecal contamination of these water sources.

All well water samples had a Total Viable Count (TVC) less than 500CFU/ ml after chlorination which agrees with the WHO guidelines for drinking water ≤ 500CFU/ ml. Protozoal pathogens such as *E. histolytica* and *G. intestinalis* were not detected in any spring water samples in spite of the presence of bacterial indicators and pathogens.

The nitrate (NO_3) concentration (< WHO guideline 45 mg/l) is higher in spring water samples (75%) than in well water samples (20%). Nitrate is a major component of fertilizers which are overused in agriculture. Another source of nitrate may be wastewater from different sources especially poorly designed cesspits and stream wastewater.

These results show that tested springs are highly polluted and to a greater extent than the deep wells. This is directly related to the rocks characteristics and depth of ground water. Specific measures have to be taken to improve the situation of spring water, aimed at reducing the risk of waterborne infectious diseases.

ميكروبيولوجيا المياه في حوض وادي العروب

مصادر المياه الفلسطينية مهددة بكثير من الأخطار أهمها الإستغلال الكبير للمياه الجوفية الفلسطينية من قبل الإسرائيليين، و كذلك التلوث لهذه المياه من قبل المستوطنات الإسرائيلية المنتشرة بشكل واسع على الأراضي الفلسطينية. هذه العوامل مع عدم الإهتمام الكافي بحماية البيئة من قبل الفلسطينيين،يهدد مباشرةً مصادر المياه الفلسطينية، و يزيد من مستوى تلوث المياه و الأخطار الصحية الناتجة عنه.

فالدراسة الميكروبيولوجية لمصادر المياه احدى الطرق عالية الأهمية في تقييم جودة المياه، فالعبئ العالمي للامراض المنتقلة عن طريق المياه جديرة بالإهتمام، فالحالات المسجلة أقل تقديراً للأمراض المنتقلة عبر المياه، وفلسطين لا تستثنى من ذلك.

بعض الدراسات في الضفة الغربية اعتنت بالدراسة الميكروبيولوجية للمياه و اشارت الى المياه سيئة الجودة المستخدمة من قبل الإنسان. هذه الدراسات استخدمت عدداً قليلاً من المؤشرات البكتيرية لفحص جودة المياه في الخزانات المنزلية و آبار الجمع و برك السباحة. منها الدراسه الحديثة التي بحثت عن وجود المادة الوراثية (DNA) للممرض *H. pylori* و ممرضات أخرى في ثلاث مناطق فلسطنية منها منطقة وادي العروب.

في هذه الدراسة الميكروبيولوجية لمياه حوض مكتظ بالسكان من الضفة الغربية ألا وهو حوض وادي العروب، تم التركيز على فحص عينات من المياه الجوفية الصادرة من تسعة آبار جوفية و احدى عشرة نبعة، حيث تم تحديد العديد من المؤشرات البكتيرية و العديد من مسببات الأمراض البكتيرية و اثنان من مسببات الأمراض الطفيلية وكذلك مستوى النيتريت في الماء.

في هذه الدراسة و التي تمت في الفترة ما بين أيار 2005- كانون ثاني 2006م على عينات المياه الجوفية في حوض وادي العروب في فلسطين، و البالغة مساحته قرابة 61 كم^2 ، و هو جزء من حوض وادي الأردن – البحر الميت و الحوض الشرقي للخزان الجبلي.

تهدف هذه الدراسة الى تقديم معلومات حول منطقة وادي العروب و ذلك بتحديد العديد من الملوثات الميكروبيولوجية و ربما مصادرها و كذلك تأثيرها على مصادر المياه في المنطقة ثم تحديد الإجراءات اللازمة لمكافحة التلوث.

تم قياس كل من , *P. aeruginosa* , *S. aureus* , TVC , Faecal streptococci , FC , TC, *Giardia intestinalis* , *Entamoeba histolytica* و النيترات في مصادر المياه الجوفية في المنطقة.

خمس و ثمانون عينة تم جمعها، حيث تم تحديد بكتيريا Total coliform في 17 عينة (52%) من أصل 33 عينة من مياه الآبار الجوفية و في 33 عينة (94%) من أصل 35 عينة من مياه الينابيع. العينات التي تم تحليلها لبكتيريا Faecal coliform لم توجد في جميع عينات مياه الآبار الجوفية أما مياه الينابيع فتم ايجادها في 35 عينة (80%) من مجموع 44 عينة. بكتيريا Faecal streptococci وجدت في 5 عينات (12%) من أصل 41 عينة من مياه الآبار الجوفية و في 38 عينة (86%) من أصل 44 عينة من مياه الينابيع. بكتيريا *S. aureus* تم تحديدها في 9 عينات (22%) من أصل 41 عينة من مياه الآبار الجوفية و 36 عينة (82%) من أصل 44

عينة من مياه الينابيع. بكتيريا *P. aeruginosa* وجدت في 3 عينات (7%) من 41 عينة من مياه اللآبار الجوفية و 8 عينات (18%) من 44 عينة من مياه الينابيع.

المعزولات البكتيرية لـِ Total coliform تم تصنيفها الى *Enterobacter , Klebsiella, Citrobacter ,* و *Escherichia coli* و Faecal coliform حيث تم تحديدها بالنسب 48%، 9%، 12%، 15%، و 0 % على التوالي في عينات مياه الآبار الجوفية. أما المعزولات البكتيرية لـِ Total coliform في عينات مياه الينابيع تم تصنيفها الى *Enterobacter Klebsiella, Citrobacter, Escherichia coli* , و Faecal coliform و وجدت بالنسب 46%، 54%، 46%، 71%، و 80%، على التوالي؛ و هذا يشير الى تلوث حقيقي لمياه الينابيع. تحديد *E. coli* في 5 عينات (15%) من مياه الآبار الجوفية و 25 عينة (71%) من مياه الينابيع؛ دليل على وجود التلوث البرازي في مصادر المياه.

جميع العينات من الآبار الجوفية التي أجري لها فحص TVC بعد نظام الكلورة كانت أقل من 500CFU/ ml و هذا يتفق مع المعايير التي وضعتها منظمة الصحة العالمية و كذلك المعايير الفلسطينية لمياه الشرب وهي أقل من 500CFU/ ml ، مما يدل على أن نظام الكلورة المقام على الآبار الجوفية ذو كفاءة عالية.
الممرضات الطفيلية مثل *Giardia intestinalis and Entamoeba histolytica* لم يتم ايجادها في أي من عينات الآبار الجوفية و كذلك عينات الينابيع، بالرغم من وجود مؤشرات و ممرضات بكتيرية.

تركيز النيترات في مياه الينابيع كان أعلى منه في مياه الآبار الجوفية حسب معايير منظمة الصحة العالمية (< 45 ملغم / لتر) . النيترات من المكونات الرئيسية للسماد الذي يستعمل كثيراً في الزراعة. و لوجود تلوث عالي في مياه الينابيع بالميكروبات القادمة من مياه المجاري.
هذه النتائج تبين أن الينابيع التي تم فحصها عالية التلوث بالموشرات و الممرضات البكتيرية و كذلك النيترات بالمقارنة مع الآبار الجوفية، و هذا يعود بشكل مباشر الى خصائص الصخور و عمق المياه الجوفية. لذا يجب أن تكون هناك إجراءات محددة لتحسين نوعية مياه الينابيع لتقليل أخطار الأمراض المنتقلة عن طريق المياه.

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Abbreviations

%:	percent
°:	degree
°C:	degree centigrade (Celsius scale)
AET:	Actual Evapotranspiration
AD:	after the birth of Christ (anno domini in Latin)
APHA:	American Public Health Agency
ARIJ:	Applied Research Institute Jerusalem
ArrLPA:	Arroub lower perched aquifer
ArrUPA:	Arroub upper perched aquifer
BC:	Before Christ
CFU:	Colony Forming Unit
DNA:	Deoxyribonucleic Acid
E:	East
E. coli:	*Escherichia coli*
e.g.:	for example
ET:	Evapotranspiration
et al.	And others
E. histolytica:	*Entameoba histolytica*
FC:	Faecal coliform
F. streptococci:	Faecal streptococci
Fig:	Figure
G. intestinalis:	*Giardia intestinalis*
h:	hour
HPC:	Heterotrophic plate counts
$Km^{2:}$	Kilometer square
Lab:	Laboratory
m^2:	Meter square
m^3:	Cubic meter
MCM:	Million Cubic Meters

masl:	meters above sea level
mbsl:	meters below sea level
mbgl:	meter below ground level
mg/l:	milligram per liter
min:	minutes
ml:	milliliter
µm:	micrometer
mm:	millimeter
MOPIC:	Ministry of Planning and International Cooperation
N:	nitrogen
N:	North
NAS:	National Academy of Sciences
ND:	Not Done
NF:	Not Found
nm:	nanometer
$NO_{3:}$	Nitrate
O:	Oxygen
PCBS:	Palestinian Central Bureau of Statistics
PCR:	Polymerase chain reaction.
PG:	Palestinian Grid
PNAMO:	Palestinian National Authority – Meteorological Office
P. aeruginosa:	*Pseudomonas aeruginosa*
PET:	Potential evapotranspiration
PWA:	Palestinian Water Authority
R:	Round
Spp:	species
S. aureus:	*Staphylococcus aureus*
STEC:	Shiga toxigenic *Escherichia coli*
T:	Temperature
TC:	Total coliform
TMTC:	Too Many To Count

TVC:	Total Viable Count
UK:	United Kingdome
UNCED:	United Nations Conference on Environment and Development
UNRWA:	United Nations Relief and Works Agency
UV:	Ultra violet
USA:	United States of America
WHO:	World Health Organization
WSP:	Water Safety Plans

Chapter 1

Introduction

All through historical times to the present water has been a scarce commodity in the region between the Jordan River and the Mediterranean Sea i.e. Palestine. The scarcity of water resources in the West Bank is due to arid to semi-arid climate, over exploitation, mismanagement as well as the fact that these resources are shared with Israel gives it great importance. Water is the most valuable and precious resource of the region. From the onset of the Madrid peace process in 1991, discussions on water were included in both the bilateral and multilateral peace negotiations. The area of this study, Wadi Al-Arroub drainage basin, suffers from water scarcity as well as the whole West Bank. Wadi Al-Arroub drainage basin enjoys a Mediterranean climate with hot and dry summers and mild and wet winters. The average annual rainfall from 1953 to 2001 was about 607 mm (Qannam, 2003).

The inhabitants of Wadi Al-Arroub experience frequent interruptions or even absence of tap water supply for long periods. This forces them to utilize water from unprotected springs and open pits, or to pay 4-5 folds the price for obtaining water to fulfill their basic domestic needs. The scarcity of water is also a main reason, which causes farmers not to cultivate their land and thus allowed urban expansion on agricultural land.

Wadi Al-Arroub drainage basin with an area of 61 Km^2 is situated midway between Hebron and Bethlehem. Wadi Al-Arroub drainage basin was chosen for the present study, because of lack of data concerning the microbiology of this drainage basin. Further more, evidence of serious pollution from the many springs in this basin as well as the sewage flow along the talweg is a significant health hazard for the local inhabitants. (Talweg is a geomorphological term meaning the line of lowest points along a valley floor).

Qannam, 2003 conducted a hydrogeological, hydrochemical and environmental study in Wadi AL-Arroub drainage basin, south of the West Bank. The hydrochemical study which involved sampling and analysis of water samples from the deep wells and springs, showed that rainwater is the only source of ground water recharge. Mixing with wastewater leaking from the poorly designed cesspits

and the wastewater conduit and/or the infiltration of the leachates from washing piles of animal dung by the rainfall in winter are the main factors responsible for the modifications of water types and quality recorded in the area. Qannam (2003) analyzed water samples classified into two main groups: Group A consisted of three subgroups. The first subgroup included the springs and dug wells located between houses showing slight contamination; the second subgroup included the deep wells and the third subgroup included the dug wells and springs away from the conduit and housing showing good water quality. Group B included the dug wells close to the conduit, which showed the highest contamination (Qannam, 2003).

According to Qannam's results, study of the microbiology of groundwater wells and springs in Wadi Al-Arroub is of utmost importance due to its utilization for domestic and agricultural uses, which directly affect human health.

The safe quality of water supplied to communities is an important consideration in the protection of human health and well-being. Drinking-water should be suitable for human consumption and for all usual domestic purposes. When a guideline value is exceeded, the cause should be investigated and corrective action taken.

In most countries, the principal risks to human health associated with the consumption of polluted water are microbiological in nature (although the importance of chemical contamination should not be underestimated). As indicated in “Agenda 21” of UNCED, “An estimated 80% of all diseases and over one-third of deaths in developing countries are caused by the consumption of contaminated water and on average as much as one-tenth of each person’s productive time is sacrificed to water-related diseases” (WHO, 2004)

The risk of acquiring a waterborne infection increases with the level of contamination by pathogenic microorganisms. However, the relationship is not necessarily a simple one and depends very much on factors such as infectious dose and host susceptibility. Drinking water is only one vehicle for disease transmission.

The basic and essential requirements to ensure the safety of drinking-water are a “framework” for safe drinking-water, comprising health-based targets established by a competent health authority; adequate and properly managed systems (adequate infrastructure, proper monitoring and effective planning and management); and a system of independent surveillance (WHO, 2004).

Several studies in the West Bank dealt with the microbiology of water and pointed to the poor quality of water used for human consumption. These studies examined few microbiological indicator bacteria in cisterns, springs, dug wells and swimming pools.

Al-Khatib and Orabi, 2004 showed that 87% of tested drinking-water of rain-fed cisterns in three villages in Ramallah and Al-Bireh (northern Jerusalem) were highly contaminated, 10.5% of tested samples had low contamination, and unfortunately, only 5% of tested samples were not contaminated and were suitable for drinking without treatment.

Al-Khatib and Salah, 2003 studied Fifty-eight water samples, collected from 46 swimming pools, and examined them for Coliforms and bacterial species including Streptococci, Salmonellae, and Staphylococcus. Salmonellae were isolated in 21 out of 23 samples. All of the examined samples from the swimming pools water were unacceptable according to the Palestinian and WHO standards.

Scarpa, 1999 was mainly concerned with the quality of domestic and irrigation spring water for a group of Palestinian villages in the southern West Bank. Analysis of the water sampled from 75 of these springs showed that all the springs were contaminated with bacteria (Coliforms). Chemical pollutants were also observed in many of the springs.

Bdair, 2005 studied the prevalence and characterization of Shiga toxigenic *Escherichia coli* (STEC) in Tulkarm and Jenin domestic wells in the West Bank.

Bdair's study involved 94 domestic water samples collected from 26 domestic wells between March 2004 and March 2006 from two cities (Tulkarm [58 samples] and Jenin [36 samples]) in the northern part of the West Bank.

The Total coliform bacterial isolates were identified in (67%) (63/94) of the water samples analyzed in this study. The differentiation into Faecal coliform, *Enterobacter spp, Klebsiella spp, Citrobacter spp and Escherichia coli* in Tulkarm water samples was 7%, 29%, 12%, 12%, and 24% respectively whereas the differentiation of Total coliform bacterial isolates in Jenin into Faecal coliform, *Enterobacter spp, Klebsiella spp, Citrobacter spp and Escherichia coli* was 33%, 53%, 42%, 39%, and 83% respectively. These results showed that there is more serious domestic water contamination in Jenin at the time of the study.

Bdair's results pointed to the extent of contamination of domestic wells in Tulkarm and Jenin in the northern part of the West Bank.

In a recent study (Atteyeh, 2007) *Helicobacter pylori* DNA in Palestinian water samples from different districts (Wadi Al-Arroub, Jericho and Tulkarem) was studied using polymerase chain reaction (PCR). The study showed that Wadi Al-Arroub water samples had the highest prevalence for *H. pylori* (3/13), with additional contamination with either enteric bacteria or *Legionella*. The study recommended investigation of Palestinian water quality especially biological.
Atteyeh's study using PCR technique detected DNA that is the presence or absence of *H. pylori*, enteric bacteria or *Legionella*.

The microbiological contamination of drinking-water supplies can have serious health consequences for consumers. This reasoning led us to conduct this study, the first of its kind in the West Bank, which detect the presence of viable indicator bacteria, two bacterial pathogens two protozoan pathogens and nitrate level in nine deep wells and eleven springs in Wadi Al-Arroub drainage basin (Fig 1.1).

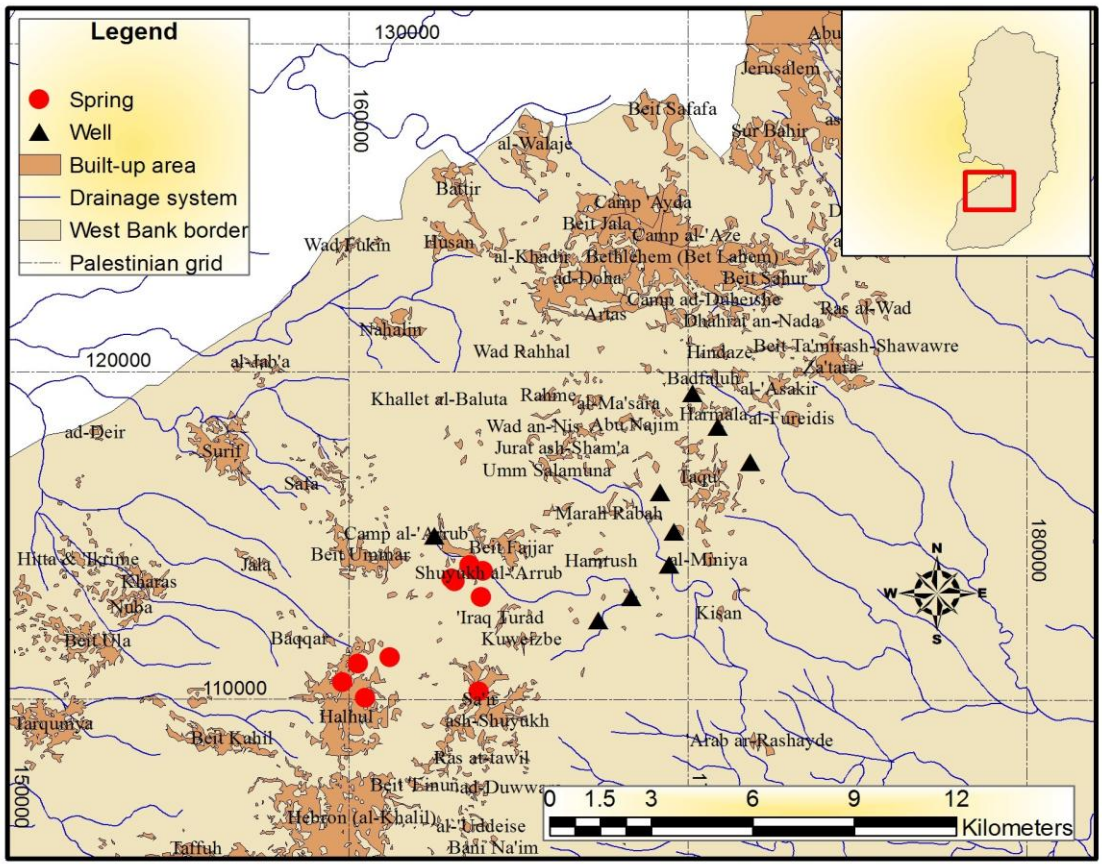

Fig 1.1: Location map of the deep wells and springs of Wadi Al-Arroub drainage basin.

1.1 Location

Wadi Al-Arroub drainage basin drains part of the Hebron Mountains it is a sub-basin of the Jordan River-Dead Sea basin. Wadi Al-Arroub is a tributary to Wadi Ghar joining it at the Western edge of the Jerusalem Desert. Wadi Al-Arroub drainage basin is part of the Hebron district which represents the Southern part of the West Bank / Palestine, that is located midway between Hebron and Bethlehem. The study area lies within 35°05`00" and 35°13`51" longitude 31°32`50" and 31°40`20" latitude respectively between 108 -120 N and 158-170 E referenced on the Palestinian Grid (Fig 1.2) (Qannam, 2003).

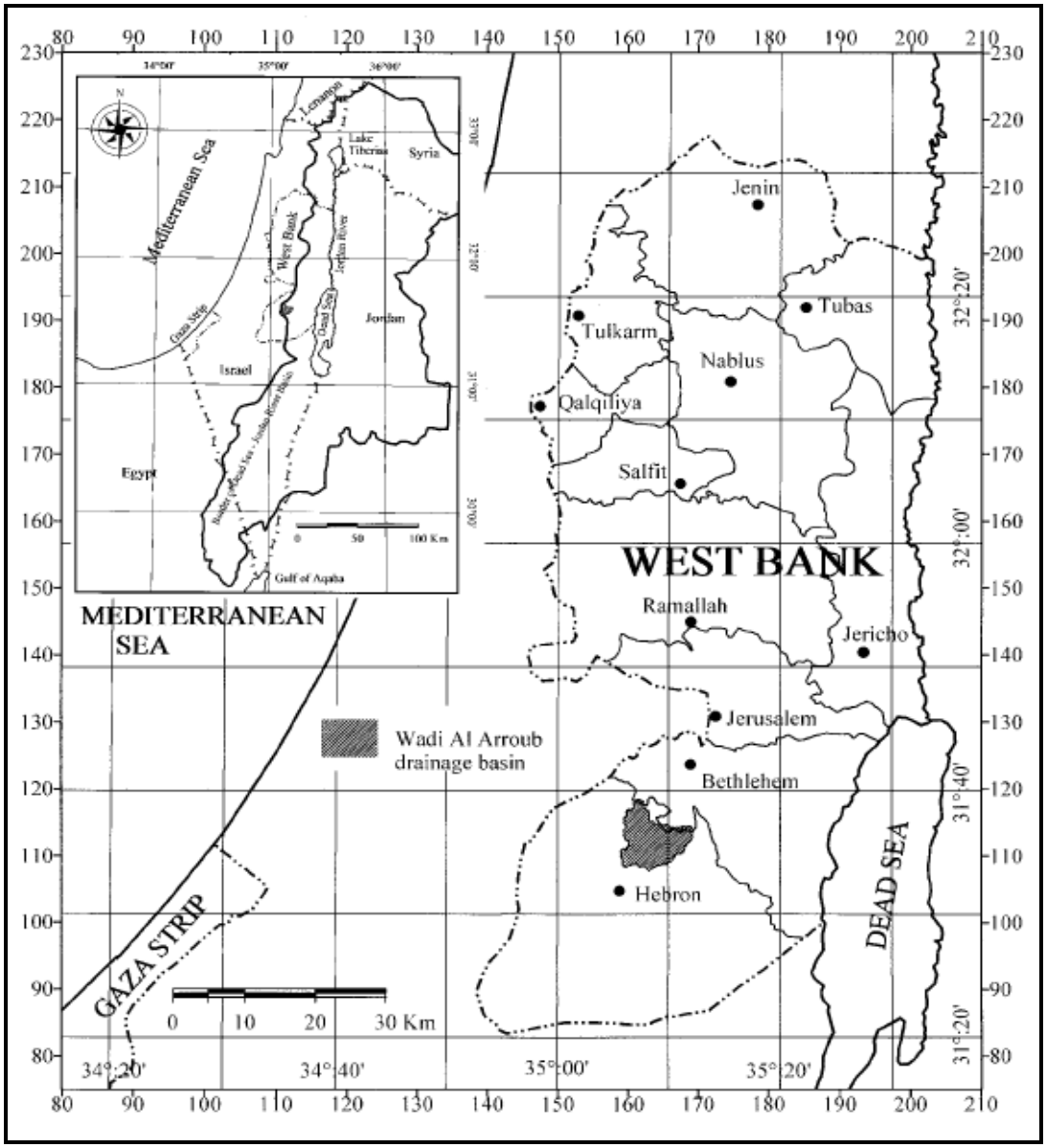

Fig. 1.2: Map of Wadi Al-Arroub drainage basin (Qannam, 2003).

1.2 Land use

Wadi Al-Arroub drainage basin includes within its boundaries all or part of the nine Palestinian communities, Arroub Camp, Shuyukh Al-Arroub, Kuweisiba, Urqan Tarrad, Si'ir, Esh-Shuyukh, Beit Ummar, Halhul and Beit Fajjar and parts of three Israeli settlements: Kefar Ezyon, Karme Zur and Asfar. Various land use activities could be identified in this area (Fig 1.3 and Table 1.1).

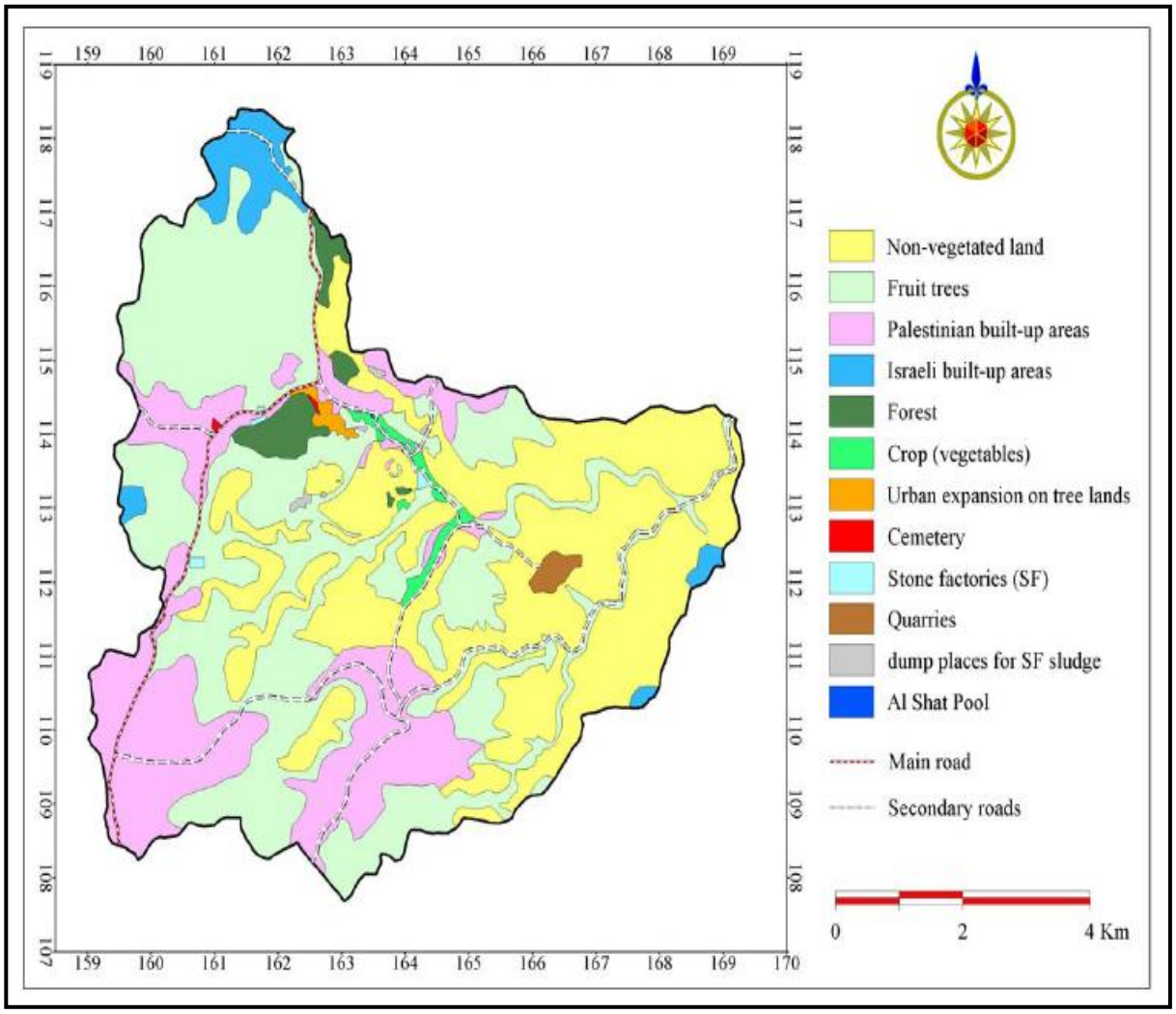

Fig 1.3: Land use map of Wadi Al-Arroub drainage basin (1999) (Qannam, 2003).

Table 1.1: The area and the distribution of the main land use activities in Wadi Al-Arroub drainage Basin (Qannam, 2003).

Land use	Area (m^2)	Percentage
Congested Palestinian areas	1.14×10^7	19.34
Israeli settlements	1.96×10^6	3.23
Forest	1.44×10^6	2.37
Fruit trees (Almonds, peaches, plums, grapes, etc.)	2.12×10^7	34.81
Unexploited land	2.33×10^7	38.37
Crops (mainly vegetables)	7.00×10^5	1.15
Stone factories	7.91×10^4	0.13
Cemeteries	3.17×10^4	0.05
Quarries	5.79×10^4	0.10
Dumps (Solid waste and sludge of stone factories)	5.99×10^4	0.10
Tree-land endangered by urban expansion	2.06×10^5	0.34

1.3 Population

Based on the study of the Palestinian Central Bureau of Statistics (PCBS) (2006), the Palestinian population of Wadi Al-Arroub drainage basin was estimated to be about 69,000 inhabitants; 8360 in Arroub Camp, 1380 in Shuyukh Al-Arroub, 985 in Kuweisiba and Urqan Tarrad, 13450 in Si'ir, 8430 in Esh-Shuyuk and approximately 1935 in Beit Fajjar, 12660 in Beit Ummar and 21800 in Halhul.

1.4 Soils

Despite the small size of West Bank, a variety of soils can be found. The major causes of this variety are the extreme conditions which form these soils: climate, arid in the East and wet in the mountainous ridge; variable geology: sedimentary rocks, sand dunes, alluvium, etc., and different topographic circumstances: topography varying from 400 meters above sea level (masl) at the Western edge to 1000 masl at the mountain ridge to 410 meters below sea level (mbsl) at the Dead Sea area. In addition, physical weathering from both water and wind modifies the soils (Qannam, 2003).

According to Orni and Efrat (1980), Wadi Al-Arroub drainage basin includes within its boundary two main soil associations: Terra Rossa stemming from dolomite and hard limestone. Terra Rossa has a reddish brown color and its depth varies between 0.5 and 2 meters. The second soil dominating are the brown and pales rendzinas with reddish and brown color, loamy with 30 % stones. These soils develop from marl and chalk. According to Dan et al. (1975), the brown rendzina has a clayey soil texture with 59.5 %, 16.6 %, and 23.9 % of clay, silt and sand respectively, while the pale rendzina has sandy clay soil texture with 45.3 %, 0 %, and 54.8 % of clay, silt, and sand respectively.

1.5 The importance of Wadi Al-Arroub drainage basin

According to the Ministry of Planning and International Cooperation (MOPIC) (1998), Palestinian Authority, Wadi Al-Arroub drainage basin is a highly sensitive recharge area, a highly valuable agricultural land, of high ecological significance, and of a second-degree landscape. This was based on the relatively high rainfall averages in this area, being part of the recharge area for the Upper and Lower regional aquifers of the West Bank, karstic features, fertile soil, about 55 springs and dug wells, and about 40 % of the area is being used for cropping and forestry. During the time of the Romans (63 BC – 325 AD) the water of the springs namely that from Ein El-Fawwar, Kuweisiba, El-Bas, Dilbi, and Al-Baradah was collected in a pool (Al-Shat, storage capacity of 20,000 m^3) and conveyed through a 40 km stone-carved rock channel (Al-Sabeel) to the city of Jerusalem. The pool and the channel were rehabilitated three times during the Islamic period (1483, 1505, and 1520) (Taha, 1999). This gives the area a historical as well as touristic importance (Qannam, 2003).

1.6 Aims of the Study:

1. The main aim of this study is to carry out specific bacteriological (indicators and pathogens), protozoal, and nitrate tests on groundwater wells and springs in order to determine the biological water quality in Wadi Al-Arroub Drainage basin.

2. Detection of indicator pollutants Total Viable Count (TVC), Total coliform (TC), Faecal coliform (FC), Faecal streptococci, and nitrate that would instigate research for possible sources of contamination which affect the water resources.

3. Determination of the prevalence of Total coliform categories including *Enterobacter, Citrobacter, Klebsiella and E. coli* in groundwater wells and springs in Wadi Al-Arroub drainage basin.

4. Detection of certain human bacterial and protozoan pathogens namely; *Staphylococcus aureus, Pseudomonas aeruginosa*, *Entameoba histolytica* and *Giardia intestinalis* in tested groundwater.

5. Assessment of chlorination treatment efficiency by doing Total Viable Count and coliform tests on the samples before and after chlorination.

1.7 Microbes of Concern

1.7.1 Indicator Organisms (Bacteria):

1.7.1.1 Total Viable Count (TVC):

1.7.1.1.1 General description:

Heterotrophic plate count (HPC) measurement detects a wide spectrum of heterotrophic microorganisms, including bacteria and fungi, based on the ability of the organisms to grow on rich growth media, without inhibitory or selective agents, over a specified incubation period and at a defined temperature. The spectrum of organisms detected by HPC testing includes organisms sensitive to disinfection processes, such as coliform bacteria; organisms resistant to disinfection, such as spore formers; and organisms that rapidly proliferate in treated water in the absence of residual disinfectants. The tests detect only a small proportion of the microorganisms that are present in water. The population recovered will differ according to the method and conditions applied. Although standard methods have been developed, there is no single universal HPC measurement (Ashbolt, et al., 2001).

1.7.1.1.2 Indicator value:

The test has little value as an index of pathogen presence but can be useful in operational monitoring as a treatment and disinfectant indicator, where the objective is to keep numbers as low as possible. In addition, HPC measurement can be used in assessing the cleanliness and integrity of distribution systems and the presence of biofilms (Bartram, 2003).

1.7.1.1.3 Source and occurrence:

Heterotrophic microorganisms include both members of the natural (typically nonhazardous) microbial flora of water environments and organisms present in a

range of pollution sources. They occur in large numbers in raw water sources. The actual organisms detected by HPC tests vary widely between locations and between consecutive samples. Numbers of HPC organisms are reduced significantly by disinfection practices, such as chlorination, ozonation and UV light irradiation. However, in practice, none of the disinfection processes sterilizes water; under suitable conditions, such as the absence of disinfectant residuals, HPC organisms can grow rapidly (Ashbolt, et al., 2001).

1.7.1.1.4 Significance in drinking water

After disinfection, numbers would be expected to be low; for most uses of HPC test results, however, actual numbers are of less value than changes in numbers at particular locations. In distribution systems, increasing numbers can indicate deterioration in cleanliness, possibly stagnation and the potential development of biofilms (Bartram, 2003).

1.7.1.2 Total coliform bacteria*:*

1.7.1.2.1 General description:

Total coliform bacteria include a wide range of aerobic and facultative anaerobic, Gram-negative, non-spore-forming bacilli. *Escherichia coli* and thermotolerant coliforms are a subset of the Total coliform group that can ferment lactose at higher temperatures. Traditionally, coliform bacteria were regarded as belonging to the genera Escherichia, Citrobacter, Klebsiella and Enterobacter, the Total coliform group includes both faecal and environmental species (Ashbolt, et al., 2001).

1.7.1.2.2 Indicator value:

Total coliform*s* include organisms that can survive and grow in water. Hence, they are not useful as an index of faecal pathogens, but they can be used as an indicator of treatment effectiveness and to assess the cleanliness and integrity of distribution systems and the potential presence of biofilms. In addition, Total coliform*s* are far more sensitive to disinfection than are enteric viruses and protozoa. Heterotrophic

plate counts (HPC) measurements detect a wider range of microorganisms and are generally considered a better indicator of distribution system integrity and cleanliness (Ashbolt, et al., 2001).

1.7.1.2.3 Source and occurrence:

Total coliform bacteria (excluding *E. coli*) occur in both sewage and natural waters. Some of these bacteria are excreted in the faeces of humans and animals, but many coliforms are heterotrophic and able to multiply in water and soil environments. Total coliforms can also survive and grow in water distribution systems, particularly in the presence of biofilms (Sueiro, 2001)

1.7.1.2.4 Significance in drinking water:

Total coliforms should be absent immediately after disinfection, and the presence of these organisms indicates inadequate treatment. The presence of Total coliforms in distribution systems and stored water supplies can reveal regrowth and possible biofilm formation or contamination through ingress of foreign material, including soil or plants (Grabow, et al., 1996).

1.7.1.2.5 Differentiation of Total coliform:

The coliform group has been defined as, all organisms that produce a colony with a golden-green metallic sheen within 24 hours on an Endo-type medium containing lactose. Differentiation of coliforms is necessary to determine the nature of pollution. It is particularly important to distinguish the presence of *E. coli* (APHA, 1995).

It was recognized at an early date that some strains included in the Total coliform group were not common in faecal material, organisms of *the Klebsiella, Enterobacter* and *Citrobacter* genera have been found in soils and on vegetation, in faeces however, they are present in much smaller numbers than *E. coli*. This is characteristically the predominant coliform in warm-blooded animal intestines (Frank and Skinner, 1941).

Although all the coliform genera *E.coli*, *Klebsiella, Enterobacter* and *Citrobacter* are present in fresh faeces and fresh pollution from feacal sources, they may not all persist in water for the same length of time. *E. coli* for example, is generally most sensitive to environmental stresses and least likely to grow in the environment. *Klebsiella*, *Enterobacter* and *Citrobacter* on the other hand are more likely to persist and to grow on organic-rich materials or in organic-rich water (APHA, 1995).

1.7.1.3 Faecal coliform:

1.7.1.3.1 General description:

Total coliform bacteria that are able to ferment lactose at 44–45 °C are known as thermotolerant coliforms. *Escherichia coli*, the predominant genus in most waters, is present in very high numbers in human and animal faeces and is rarely found in the absence of faecal contamination, although there is some evidence for growth in tropical soils (Ashbolt, *et a*l., 2001).

1.7.1.3.2 Indicator value:

Escherichia coli is considered the most suitable index of faecal contamination. In most circumstances, populations of thermotolerant coliforms are composed predominantly of *E. coli*; as a result, this group is regarded as a less reliable but acceptable index of faecal pollution. *Escherichia coli* (or, alternatively, thermotolerant coliforms) is the first organism of choice in monitoring programmes for verification, including surveillance of drinking-water quality. These organisms are also used as disinfection indicators, but testing is far slower and less reliable than direct measurement of disinfectant residual. In addition, *E. coli* is far more sensitive to disinfection than are enteric viruses and protozoa (Ashbolt, et al. 2001).

1.7.1.3.3 Source and occurrence:

Escherichia coli occurs in high numbers in human and animal faeces, sewage and water subject to recent faecal pollution. Water temperatures and nutrient conditions

present in drinking-water distribution systems are highly unlikely to support the growth of these organisms (Sueiro, 2001).

1.7.1.3.4 Significance in drinking water:

The presence of *E. coli* (or, alternatively, thermotolerant coliforms) provides evidence of recent faecal contamination, and detection should lead to consideration of further action, which could include further sampling and investigation of potential sources such as inadequate treatment or breaches in distribution system integrity (George, 2001).

1.7.1.4 Faecal streptococci:

1.7.1.4.1 General description:

Intestinal enterococci are a subgroup of the larger group of organisms defined as Faecal streptococci, comprising species of the genus *Streptococcus*. These bacteria are Gram-positive and relatively tolerant to sodium chloride and alkaline pH environments. They are facultative anaerobic and occur singly, in pairs or as short chains. Intestinal enterococci group was separated from the rest of the Faecal streptococci because they are relatively specific for faecal pollution. However, some intestinal enterococci isolated from water may occasionally also originate from other habitats, including soil, in the absence of faecal pollution (Pinto, 1999).

1.7.1.4.2 Indicator value:

The intestinal enterococci group can be used as an index of faecal pollution. Most species do not multiply in water environments. The numbers of intestinal enterococci in human faeces are generally about an order of magnitude lower than those of *E. coli.* Important advantages of this group are that they tend to survive longer in water environments than *E. coli* (or thermotolerant coliforms), are more resistant to drying and are more resistant to chlorination. Intestinal enterococci have been used in testing of raw water as an index of faecal pathogens that survive longer than *E. coli* and in drinking water to augment testing for *E. coli*. In addition, they

have been used to test water quality after repairs to distribution systems or after new mains have been set up (Ashbolt, *et al*., 2001).

1.7.1.4.3 Source and occurrence:

Intestinal enterococci are typically excreted in the faeces of humans and other warm-blooded animals. Some members of the group have also been detected in soil in the absence of faecal contamination. Intestinal enterococci are present in large numbers in sewage and water environments polluted by sewage or wastes from humans and animals (Ashbolt, *et al.*, 2001).

1.7.1.4.4 Significance in drinking water:

The presence of intestinal enterococci provides evidence of faecal contamination, and detection should lead to consideration of further action, which could include further sampling and investigation of potential sources (Junco, 2001).

1.7.2 Bacterial Pathogens:

1.7.2.1 *Staphylococcus aureus*:

1.7.2.1.1 General description:

Staphylococcus aureus is an aerobic or facultative anaerobic, non-motile, non-spore-forming, catalase and coagulase-positive, Gram-positive coccus, usually arranged in grapelike irregular clusters (LeChevallier and Seidler, 1980).

1.7.2.1.2 Human health effects:

Although *Staphylococcus aureus* is a common member of the human microflora, it can produce disease through two different mechanisms. One is based on the ability of the organisms to multiply and spread widely in tissues, and the other is based on the ability of the organisms to produce extra cellular enzymes and toxins. Infections based on the multiplication of the organisms are a significant problem in hospitals

and other health care facilities. Multiplication in tissues can result in manifestations such as boils, skin sepsis, post-operative wound infections, enteric infections, septicaemia, endocarditis, osteomyelitis and pneumonia. Gastrointestinal disease (enterocolitis or food poisoning) is caused by a heat-stable staphylococcal enterotoxin and characterized by projectile vomiting, diarrhoea, fever, abdominal cramps, electrolyte imbalance and loss of fluids (Antai, 1987)

1.7.2.1.3 Source and occurrence:

Staphylococcus aureus is ubiquitous in the environment but is found mainly on the skin and mucous membranes of animals. The organism is a member of the normal microbial flora of the human skin and is found in the nasopharynx of 20–30% of adults at any one time. Staphylococci are occasionally detected in the gastrointestinal tract and can be detected in sewage. *Staphylococcus aureus* can be released by human contact into water environments such as swimming pools, spa pools and other recreational waters. It has also been detected in drinking-water supplies (Antai, 1987).

1.7.2.1.4 Routes of exposure:

Hand contact is by far the most common route of transmission. Inadequate hygiene can lead to contamination of food. Foods such as ham, poultry and potato and egg salads kept at room or higher temperature offer an ideal environment for the multiplication of *S. aureus* and the release of toxins. The consumption of foods containing *S. aureus* toxins can lead to enterotoxin food poisoning within few hours (Antai, 1987).

1.7.2.1.5 Significance in drinking water:

S.aureus can occur in drinking-water supplies. Although staphylococci are slightly more resistant to chlorine residuals than *E. coli*, their presence in water is readily controlled by conventional treatment and disinfection processes. Since faecal material is not their usual source, *E. coli* (or, alternatively, thermotolerant coliforms) is not a suitable index for *S. aureus* in drinking-water supplies (LeChevallier and Seidler, 1980).

1.7.2.2 *Pseudomonas aeruginosa:*

1.7.2.2.1 General description:

Pseudomonas aeruginosa is a member of the family Pseudomonadaceae and is a polarly flagellated, aerobic, Gram-negative rod. When grown in suitable media, it produces the non-fluorescent bluish pigment pyocyanin. Many strains also produce the fluorescent green pigment pyoverdin (Hardalo and Edberg, 1997).

1.7.2.2.2 Human health effects:

Pseudomonas aeruginosa can cause a range of infections but rarely causes serious illness in healthy individuals without some predisposing factor. It predominantly colonizes damaged sites such as burn and surgical wounds, the respiratory tract of people with underlying disease and physically damaged eyes. From these sites, it may invade the body, causing destructive lesions or septicaemia and meningitis. Cystic fibrosis and immunocompromised patients are at a risk of colonization with *P. aeruginosa*, which can cause serious pulmonary infections in these patients. Water-related folliculitis and ear infections are associated with moist environments. *P. aeruginosa* is known for carrying resistance to a range of antibiotics (Hardalo and Edberg, 1997).

1.7.2.2.3 Source and occurrence:

Pseudomonas aeruginosa is a common environmental organism and can be found in faeces, soil, water and sewage. It can multiply in water environments and also on the surface of suitable organic materials in contact with water. *Pseudomonas aeruginosa* is a recognized cause of hospital-acquired infections with potentially serious complications. It has been isolated from a range of moist environments such as sinks, water baths, hot water systems, showers and spa pools (De Victorica and Galvan, 2001).

1.7.2.2.4 Routes of exposure:

The main route of infection is by exposure of susceptible tissue, notably wounds and mucous membranes, to contaminated water or contamination of surgical instruments. Cleaning of contact lenses with contaminated water can cause a form of keratitis (De Victorica and Galvan, 2001).

1.7.2.2.5 Significance in drinking water:

P. aeruginosa can be significant in settings such as health care facilities; however, there is no evidence that normal uses of drinking-water supplies are a source of infection in the general population. The presence of high numbers of *P. aeruginosa* in potable water can be associated with complaints about taste, odour and turbidity. *Pseudomonas aeruginosa* is sensitive to disinfection, and entry into distribution systems can be minimized by adequate disinfection. *Pseudomonas aeruginosa* is detected by HPC, which can be used together with parameters such as disinfectant residuals to indicate conditions that could support growth of these organisms (Bartram, 2003).

1.7.3 Protozoa Pathogens:

1.7.3.1 *Entamoeba histolytica:*

1.7.3.1.1 General description:

Entamoeba histolytica is the most prevalent intestinal protozoan pathogen worldwide and belongs to the superclass Rhizopoda in the subphylum Sarcodina. *Entamoeba* has a feeding, replicative trophozoite (diameter 10-60 µm), which, under unfavorable conditions, will develop into a dormant cyst (diameter 10-20 µm). Infection is contracted by the ingestion of cysts (Marshall, 1997).

1.7.3.1 .2 Human health effects:

About 85-95% of human infections with *Entamoeba histolytica* are asymptomatic. Acute intestinal amoebiasis has an incubation period of 1–14 weeks. Clinical disease results from the penetration of the epithelial cells in the gastrointestinal tract by the amoebic trophozoites. Approximately 10% of infected individuals present with dysentery or colitis. Symptoms of amoebic dysentery include diarrhea with cramping, lower abdominal pain, low-grade fever and the presence of blood and mucus in the stool. The ulcers produced by the invasion of the trophozoites may deepen into the classic flask-shaped ulcers of amoebic colitis. *Entamoeba histolytica* may invade other parts of the body, such as the liver, lungs and brain, sometimes with fatal outcome (Marshall, 1997).

1.7.3.1.3 Source and occurrence:

Humans are the reservoir of infection of *E. histolytica*. In the acute phase of infection, patients excrete only trophozoites that are not infectious. Chronic cases and asymptomatic carriers who excrete cysts are more important sources of infection and can discharge up to 1.5×10^7 cysts daily. *Entamoeba histolytica* can be present in sewage and contaminated water. Cysts may remain viable in suitable aquatic environments for several months at low temperature. The potential for waterborne transmission is greater in the tropics, where the carrier rate sometimes exceeds 50%, compared with more temperate regions, where the prevalence in the general population may be less than 10% (Marshall, 1997).

1.7.3.1.4 Routes of exposure:

Person-to-person contact and contamination of food by infected food handlers appear to be the most significant means of transmission, although contaminated water also plays a substantial role. Ingestion of faecally contaminated water and consumption of food crops irrigated with contaminated water can both lead to transmission of amoebiasis. Sexual transmission, particularly among male homosexuals, has also been documented (Marshall, 1997).

1.7.3.1.5 Significance in drinking water:

The transmission of *E. histolytica* by contaminated drinking water has been confirmed. The cysts are relatively resistant to disinfection and may not be inactivated by chlorination practices generally applied in the production of drinking water. Within Water Safety Plans (WSP), control measures that can be applied to manage potential risk from *E. histolytica* include prevention of source water contamination by human waste, followed by adequate treatment and protection of water during distribution. Owing to the resistance of the oocysts to disinfectants, *E. coli* (or, alternatively, thermotolerant coliforms) cannot be relied upon as an index of the presence/absence of *E. histolytica* in drinking-water supplies (Marshall, 1997).

1.7.3.2 *Giardia intestinalis:*

1.7.3.2.1 General description:

Giardia spp. is flagellated protozoa that parasitize the gastrointestinal tract of humans and certain animals. The genus *Giardia* consists of a number of species, but human infection (giardiasis) is usually assigned to *G. intestinalis*, also known as *G. lamblia* or *G. duodenalis*. *Giardia* has a relatively simple life cycle consisting of a flagellate trophozoite that multiplies in the gastrointestinal tract and an infective thickwalled cyst that is shed intermittently but in large numbers in faeces. The trophozoites are bilaterally symmetrical and ellipsoidal in shape. The cysts are ovoid in shape and 8-12µm in diameter (WHO, 2002)

1.7.3.2.2 Human health effects:

Giardia has been known as a human parasite for 200 years. After ingestion and excystation of cysts, the trophozoites attach to surfaces of the gastrointestinal tract. Infections in both children and adults may be asymptomatic. In day care centers, as many as 20% of children may carry *Giardia* and excrete cysts without clinical symptoms. The symptoms of giardiasis may result from damage caused by the trophozoites, although the mechanisms by which *Giardia* causes diarrhoea and intestinal malabsorption remain controversial. Symptoms generally include

diarrhoea and abdominal cramps; in severe cases, however, malabsorption deficiencies in the small intestine may be present, mostly among young children. Giardiasis is self-limiting in most cases, but it may be chronic in some patients, lasting more than 1 year, even in otherwise healthy people. Studies on human volunteers revealed that fewer than 10 cysts constitute a meaningful risk of infection (Stuart, 2003).

1.7.3.2.3 Source and occurrence:

Giardia can multiply in a wide range of animal species, including humans, which excrete cysts into the environment. Numbers of cysts as high as 88 000 per liter in raw sewage and 240 per liter in surface water resources have been reported. These cysts can survive for weeks to months in fresh water. Cysts also occur in recreational waters and contaminated food (Slifko, *et al*., 2000).

1.7.3.2.4 Routes of exposure:

By far the most common route of transmission of *Giardia* is person-to-person contact, particularly between children. Contaminated drinking water, recreational water and, to a lesser extent, food have been associated with outbreaks. Animals have been implicated as a source of human infectious *G. intestinalis*, but further investigations are required to determine their role (Rimhanen-Finne, 2002).

1.7.3.2.5 Significance in drinking water:

Waterborne outbreaks of giardiasis have been associated with drinking-water supplies for over 30 years; at one stage, *Giardia* was the most commonly identified cause of waterborne outbreaks in the USA. *Giardia* cysts are more resistant than enteric bacteria to oxidative disinfectants such as chlorine, but they are not as resistant as *Cryptosporidium* oocysts. The time required for 90% inactivation at a free chlorine residual of 1mg/litre is about 25–30 min. Owing to the resistance of the cysts to disinfectants, *E. coli* (or, alternatively, thermotolerant coliforms) cannot be relied upon as an index of the presence/absence of *Giardia* in drinking-water supplies (WHO, 2002).

1.8 Nitrate

Nitrate is an inorganic compound that occurs under a variety of conditions in the environment, both naturally and synthetically. Nitrate is composed of one atom of nitrogen (N) and three atoms of oxygen (O); the chemical symbol for nitrate is NO_3. Nitrate in drinking water is measured either in terms of the amount of nitrogen present or in terms of both nitrogen and oxygen. The federal standard for nitrate in drinking water is 45 mg/l. Nitrate in groundwater originates primarily from fertilizers, septic systems, and manure storage or spreading operations that elevate its concentration in groundwater above the levels acceptable for drinking water quality (NAS, 1978).

1.9 Microbiological Water Quality Guidelines

Water quality guidelines and standards recommended by various authorities are similar in that they intend to ensure the minimum risk of infection. The Guidelines state that drinking water must not contain waterborne pathogens. If guideline values are exceeded, immediate investigative action must be taken, including repeat testing, and thorough inspection of the treatment plant and its operation, the raw water source, and general hygiene of the water distribution system.

Table 1.2: Microbiological water quality guidelines (WHO, 2004 and PWA, 2006).

Organisms, others	**Guideline value (WHO and Palestinian)**
Total Viable Count	≤ 500 CFU/ ml
Total coliform	Must not be detected in any 100 ml sample
Faecal coliform	Must not be detected in any 100 ml sample
Faecal streptococci	Must not be detected in any 100 ml sample
Staphylococcus aureus	Must not be detected in any 100 ml sample
Pseudomonas aeruginosa	Must not be detected in any 100 ml sample
Entameoba histolytica	Must not be detected in any 4-5 Liters sample
Giardia intestinalis	Must not be detected in any 4-5 liters sample
Nitrate	NO_3 < 45 mg/l (WHO), NO_3 < 50 mg/l (Palestinian)

Chapter 2

Materials and Methods

2.1 Materials

2.1.1 Laboratory Work:

Water samples were directly transported to the Palestinian Water Authority (PWA) Central Laboratory in Ramallah, West-Bank and processed for Bacterial, protozoal and nitrate tests.

2.1.2 Equipments:

Water samples were collected and processed using the following equipments and tools:

1. Incubator (Millipore Corporation, Bedford, MA 01730, U.S.A)
2. Membrane filtration apparatus complete with vacuum source (electrically operated pump) and suction flask
3. Autoclave (Hiryama Hiclave HV-110, Japan)
4. Fume Hood (Biohazard Vertical Laminar Airflow Cabinet, Faster Via A.Vespaci, 46 44100 Ferrara, Italy)
5. Refrigerator (Camlab Limited, Cambridge CB4 1TH)
6. Laboratory balance (Precisa 2200 C SCS, Switzerland)
7. Bunsen burner
8. Thermometers
9. Light microscope (Euromex Microscope, NL 6803 ED Arnhem-Holland)
10. Petri dishes, sterile, plastic, 9 x 50 mm, with tight-fitting lids (Millipore Corporation, Bedford, MA 01730, U.S.A), glass ware and other small laboratory instruments.
11. Membrane filters, sterile, white, grid marked, 47 mm diameter, with 0.45 ± 0.02 μm pore size (Millipore Corporation, Bedford, MA 01730, U.S.A)
12. Colony counter (Model 560, Suntex, Taipei, Taiwan)

2.1.3 Culture Media:

Water samples were processed using the following culture media:

1. M-Endo agarless (HiMedia Laboratories Limited, Mumbai-400086, India)
2. M-FC agar base (HiMedia Laboratories Limited, Mumbai-400086, India)
3. KF streptococcus agar (Oxoid; Oxoid Ltd., Basingtoke, Hampshire, UK)
4. Pseudomonas isolation agar (Difco, Franklin Lakes, NJ. USA).
5. Mannitol salt agar (Oxoid; Oxoid Ltd., Basingtoke, Hampshire, UK).
6. Plate Count Agar-Tryptone Glucose Yeast agar (Oxoid; Oxoid Ltd., Basingtoke, Hampshire, UK).
7. MacConkey Agar (Fluka,Biochemika 70143, Switzerland).
8. Kligler Iron Agar (Oxoid; Oxoid Ltd., Basingtoke, Hampshire, UK).
9. Enterotube (Hy-Laboratories Ltd. Park Tamar, Rehovot 76 326, Israel)

2.2 Sampling

2.2.1 Microbiological sampling method:

Eighty five well and spring groundwater samples were collected during the period between May 2005 and January 2006 from Wadi AL-Arroub drainage Basin in the southern part of the West Bank. A total of five rounds were done. The first round (R1) was accomplished in May 2005. The second round (R2) was accomplished in June 2005. The third round (R3) was accomplished in August 2005. The fourth round (R4) was accomplished in November 2005. The fifth round (R5) was accomplished in January 2006.

Sampling for microbiological tests for wells was done as follows:
Since supply wells are equipped with permanently installed pumps and taps;
1- Before collecting the samples screens, filters, or other devices from the tap were removed.
2- Before sampling, taps were sterilized by flamer and water allowed to flow before and after sterilization for few minutes.
3- Samples were collected directly from the tap into sterile 1000 ml bottles without splashing or allowing the sample bottle to touch the tap.

Sampling for microbiological tests for springs was done as follows;

Sterile 1000 ml bottles were filled by direct flow at the end point of the spring water.

Water samples from both sources collected into sterile 1000 ml containers were immediately placed in a lightproof insulated box containing ice or ice-packs with water to ensure rapid cooling, and transported on ice to the Palestinian Water Authority (PWA) Central Lab. Samples were stored at 4 ºC and processed for microbial testing within 24 h of collection.

2.2.2 Sample Processing:

Water samples collected were analyzed for bacterial organisms (Total and Faecal coliform bacteria, Faecal streptococci, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and Total Viable Count), Protozoa (*Entamoeba histolytica* and *Giardia intestinalis*) and Nitrate (NO_3^-) levels (Fig 2.1) and differentiation of TC (Fig 2.2).

All bacterial tests, except Total Viable Counts were analyzed using the membrane filter technique; breifly, one hundred milliliters of each sample was vacuum-filtered through a sterile 0.45-µm Millipore filter. The filter was placed on the specific medium for each test followed by incubation at 37 ºC or 44.5 ºC as shown in (Table 2.1; APHA, 1995).

Table 2.1: Bacterial tests done, medium used and incubation temperature.

Bacterial test	**Medium**	**Incubation Temp**
Total coliform	M-Endo agar less	37 º C
Faecal coliform	M-FC agar base	44.5 º C
Faecal streptococci	KF streptococcus agar	37 º C
Pseudomonas aeruginosa	Pseudomonas isolation agar	37 º C
Staphylococcus aureus	Mannitol salt agar	37 º C
Total Viable Count (TVC)	Plate Count Agar Tryptone Glucose Yeast agar	37 º C
Differentiation of Total coliform	MacConkey Agar	37 º C
Identification of Total coliform groups	Kligler Iron Agar Enterotube	37 º C

Test for protozoa (*Entamoeba histolytica and Giardia intestinalis*) was done on the most polluted spring water samples based on results of presence of bacterial indicators and pathogens using the membrane filter technique (Oxoid, 1990).

Total Viable Count (TVC) tests were done for spring water and groundwater well samples collected before chlorination and after chlorination samples by using the spread plate method on solidified agar (Plate Count Agar). One ml of each sample was spread on the surface of the agar and the plates were incubated at 37 ºC for 48 h (APHA, *et al.*, 1998).

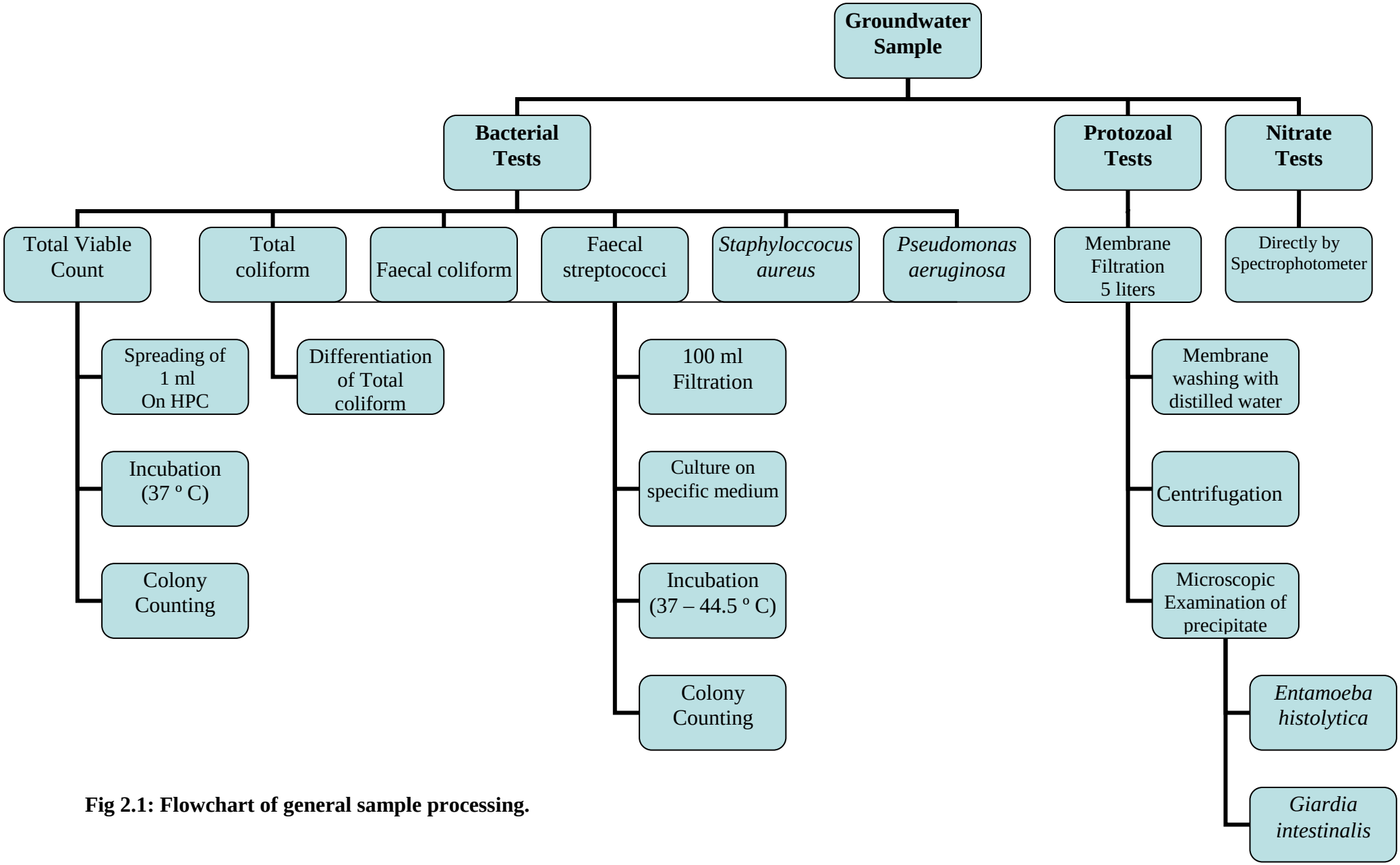

Fig 2.1: Flowchart of general sample processing.

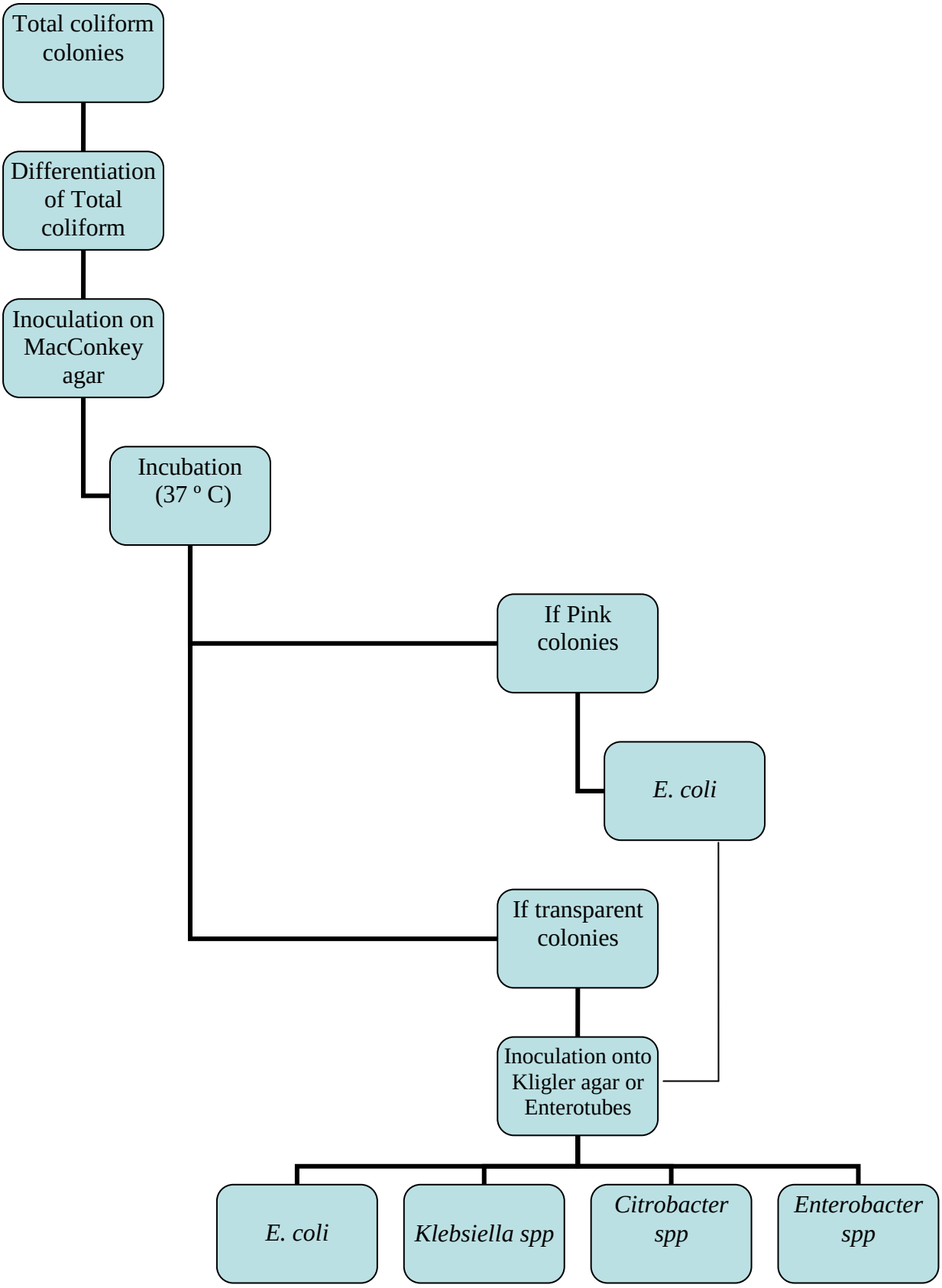

Fig 2.2: Flowchart of Total coliform Differentiation sample processing.

2.2.3 Membrane-Filter Technique:

1- 100 ml volumes selected for every sample analyzed
2- Petri dishes were labeled with station number, volume of sample filtered, date and time of sample collection
3- Sterile filter aseptically placed on sterile filtration apparatus.
4- Samples shacked and delivered to filtration apparatus aseptically
5- Vacuum applied; afterwards, filtration apparatus and cylinder were rinsed twice with sterile buffered water
6- Forceps were sterilized and filter removed and funnel replaced on filtration apparatus
8- Filter paper rolled onto media in Petri dishes and inverted Petri dishes placed in the incubator.

2.3 Microbiological analysis of water:

2.3.1: Detection of Total coliform (TC):

Total coliform*s* were detected in 100 ml samples of water. The procedure is based on the production of acid from lactose or the production of the enzyme B-galactosidase. The procedure involved membrane filtration followed by incubation of the membrane on selective media; M-Endo agar less at 35–37 °C and counting of representative green metallic sheen colonies after 24 h (APHA., 1998) (Fig 2.3).

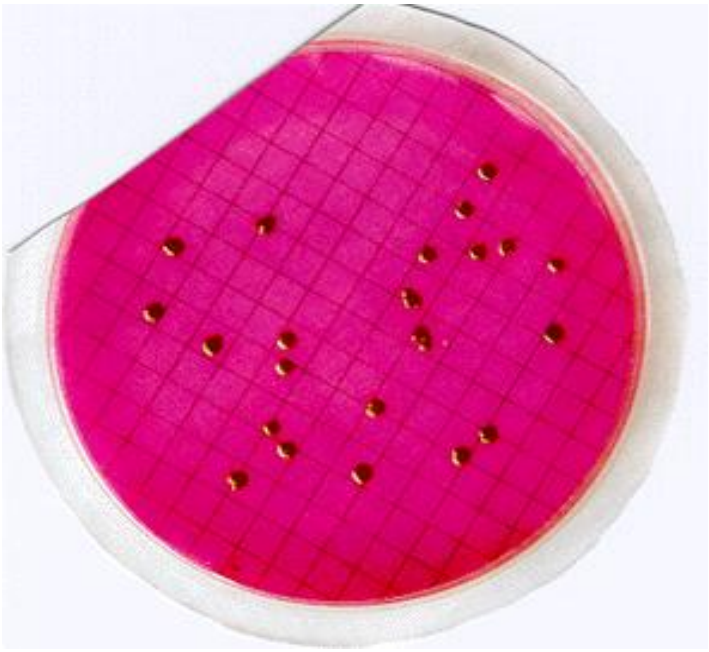

Fig 2.3: Total coliform colonies on selective media M-Endo agar less (Adapted from Microorganism photos, 1993)

2.3.2 Detection of Faecal coliform (FC):

Escherichia coli (or, alternatively, thermotolerant coliforms) were detected in 100 ml samples of water. The procedure is based on the production of acid and gas from lactose. The procedure involved membrane filtration followed by incubation of the membrane on selective medium M-FC agar base at 44-45 °C and counting of representative dark blue colonies after 24 h (APHA, 1995).

2.3.3 Detection of Faecal streptococci (F. streptococci):

Faecal streptococci were detected by membrane filtration followed by incubation of membranes on selective medium; KF streptococcus agar and counting of representative red or pink colonies after incubation at 35–37 °C for 24 h (APHA, 1995).

2.3.4 Detection of *Staphylococcus aureus*:

Staphylococcus aureus was detected by membrane filtration followed by incubation of membranes on selective medium Mannitol salt agar and counting of

representative white, gray or black colonies after incubation at 35-37 °C for 24 h (APHA, 1995).

2.3.5 Detection of *Pseudomonas aeruginosa* (*P. aeruginosa*):

Pseudomonas aeruginosa was detected by membrane filtration followed by incubation of membranes on selective medium; Pseudomonas isolation agar and counting of representative blue-green or brown pigmented colonies after incubation at 35-37 °C for 24h (APHA, 1995).

2.3.6 Detection of *Entamoeba histolytica* (*E. histolytica*):

Entamoeba histolytica was detected by membrane filtration method. The filter paper was transferred to a 100 ml beaker and repeatedly flushed with distilled water, then centrifuged for three minutes. A wet mount from the precipitate was examined under low power magnification for cysts and / or trophozoites (APHA, 1995).

2.3.7 Detection of *Giardia intestinalis* (*G. intestinalis*):

Giardia intestinalis was detected by membrane filtration method. The filter paper was transfered to a 100 ml beaker and repeatedly flushed with distilled water, then centrifuged for three minutes. A wet mount prepared from the precipitate was examined at 40X magnification for cysts and / or trophozoites (APHA, 1995).

2.4 Differentiation of Total coliform (TC):

Colonies with metallic sheen on M-Endo agar were analyzed further by subculturing on MacConkey agar for identification as *Enterobacter spp*., *Citrobacter spp., Klebsiella spp.,* or *E. coli*.

After incubation at 35°C for 24 hours, pink dry colonies growing on MacConkey were considered to be *E.coli*. Transparent (nonlactose - fermenting colonies) were cultured on Kligler Iron Agar slants and incubated for 18-24 hours. Genus

identification was based on carbohydrate utilization, gas reaction and H_2S production (APHA, 1995; Table 2.2).

Table 2.2: Identification of TC to genus based on carbohydrate utilization, gas and H_2S production (Oxoid, 1990).

Organism	Slope	Bottom	Gas	H_2S
Enterobacter spp	Red	yellow	+	-
Citrobacter spp	yellow	yellow	+	+
Klebsiella spp	yellow	yellow	+	-
E. coli	yellow	yellow	+	-

2.4.1 Enterotube Test:

Enterotubes (Hy-Laboratories) were used as confirmation tests for the differentiation of Total coliform in the fourth round (R4); October 2005 and fifth round (R5); January 2006.

2.5 Determination of Nitrate levels:

An ultraviolet (UV) technique (Ultraviolet Spectrophotometric Screening Method) was used for measuring the absorbance of NO_3 in the water samples analyzed (APHA, 1995). The Absorbance of water samples measured at 220 nm using Perkin-Elmer 5500 spectrophotometer.

Chapter 3: Geology and Hydrology

3.1 Geology

3.1.1 Stratigraphy and lithology of the West Bank:

The stratigraphy and lithology of the West Bank consist of several formations which were formed during the different geological ages; the outcrops are predominantly carbonate rocks of Cretaceous and Tertiary ages. The oldest exposed rocks belong to the Albian, overlain by younger strata of the Cenomanian, Turonian, Senonian and Eocene. These Albian rocks are exposed on both flanks of the anticlinal structure in the West Bank (Figure 3.1; Table 3.1) (Owaiwi and Awadallah, 2004).

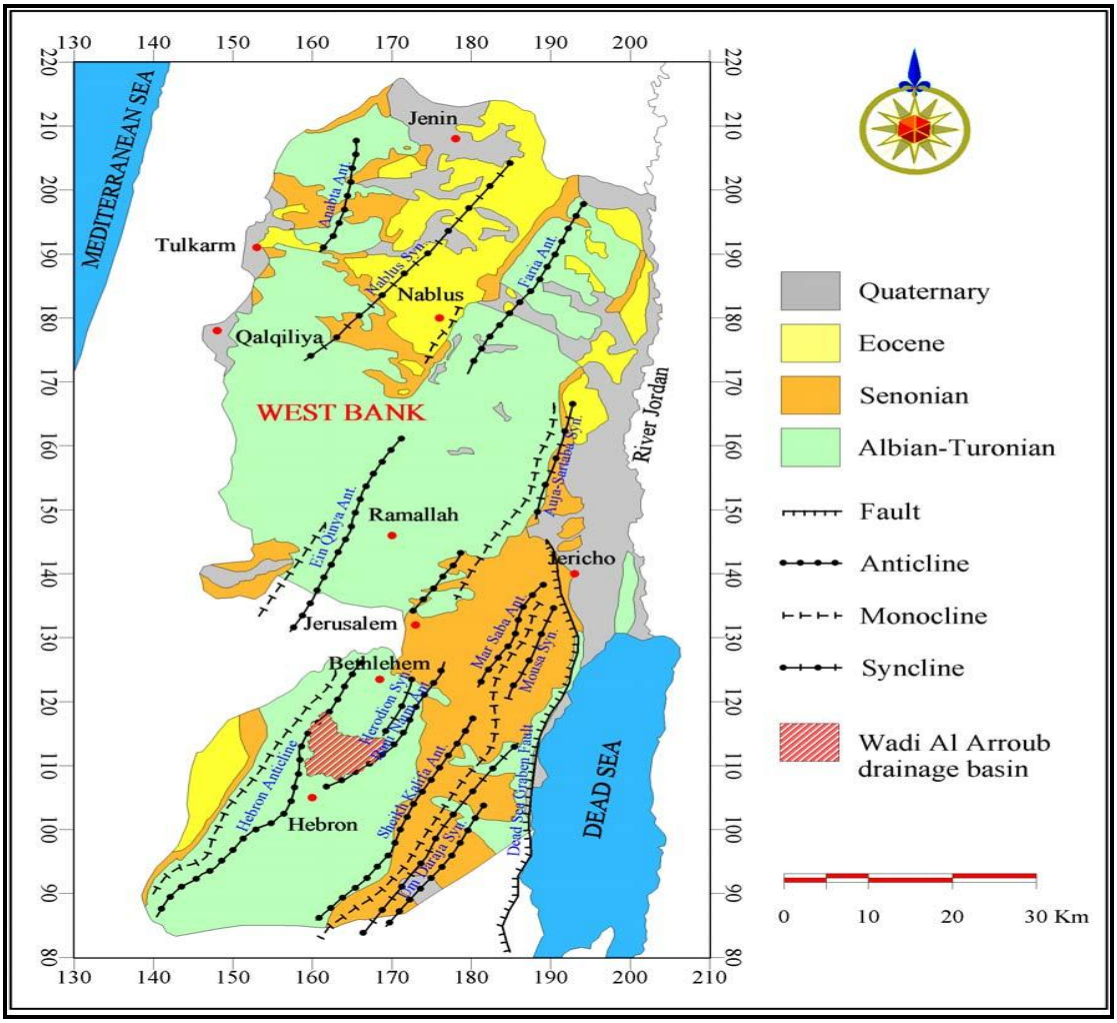

Fig 3.1: General geological and structural map of the West Bank (After Qannam, 2003)

Table 3.1: The general stratigraphy of the West Bank as well as their lithology, thickness and aquifer potentiality (modified after Braun and Hirsch 1994; Millennium Engineering Group *et al*. 2000, Guttman 2000; Guttman and Zuckerman, 1995 and Qannam, 2003).

Geological Time Scale				Group		Formation		Lithology	Thickness (m)	Hydrostatigraphy
Era	System		Epoch	Palestinian	Israeli	Palestinian	Israeli			
CENOZOIC	Quaternary		Holocene	Recent	Kurkar	Alluvium	Alluvium	Marl, alluvium, gravel	Variable	Aquifer
						Gravel	River gravel			Aquifer
			Pleistocene	Lisan		Dead Sea	Lisan	Lisan	Thinly laminated marl with gypsum bands	200+	
	Tertiary	Neogene	Pliocene-Miocene	Beida	Jenin Sub Series	Saqia	Beida	Bit Nir and Ziglag	conglomerate	0-200	Aquifer
		Paleogene	Eocene	Belqa		Avidat	Reef nummulitic limestone	Zora	Reef limestone, bedded limestone, chalk with limestone undifferentiated	100-500	Aquifer in limeston and aquiclude in chalk
			Paleocene			Mount Scopus	Nummulitic limestone	Taqiya	Marl, chalk and clay		Aquiclude
MESOZOIC	Cretaceous	Senonian	Mastrichtian				Khan Al Ahmar and Zerqa	Ghareb	Yellowish chalk		Aquiclude
			Campanian				Amman and Abu Dis	Mishash	Chalk with back chert		Aquiclude
			Santonian					Menuha	Chalk		Aquiclude
			Turonian	Ajlun		Judea	Jerusalem	Bina	Limestone and dolomite (karstic).	90-120	Aquifer
			Cenomanian				Bethlehem	Weradim	Hard gray porous dolomite	90-100	Aquifer
								Kfar Shaul	Chalky limestone, chalk and marl	30-40	Aquitard
							Hebron	Aminadav	Karstic limestone and dolomite	110-140	Aquifer
							Yatta	Moza	Marl, clay and marly limestone	10-20	Aquiclude
								Beit Meir	Limestone, chalky limestone and dolomite	120-140	Aquifer
									Limestone inter -bedded with marl		Aquiclude
			Albian				Upper Beit Kahil	Kesalon	Limestone inter-bedded with marl	30-50	Aquifer
								Soreq	Dolomite inter-bedded with marl	110-170	Aquifer
							Lower Beit Kahil	Giva't Yearim	Limestone, dolomite	20-70	Aquifer
								Kefira	Limestone, dolomite and marly limestone	120-180	Aquifer
				Kurnub			Kobar	Qatana	Marl and clay	50	Aquitard
								Ein Qinyia	Marl and marly limestone	60-70	Aquitard
								Tammun	Caly and marl	80-150	Aquitard
			Aptian					Ein Al Asad	Limestone		Aquifer
					Kurnub			Nabi Said	Limestone		Aquifer
			Neocomian				Ramali	Hatir	Sandstone	150	Aquifer
	Jurassic		Callovian-Bajocian	Zerqa	Arad		Upper Malih	Upper Malih	Marl interbedded with chalky	190	Aquitard
							Lower Malih	Lower Malih	Dolomitic limestone, jointed and karstic	55	Aquifer

3.1.2 Geology and stratigraphy of Wadi Al-Arroub drainage basin:

Carbonate rocks of Albian to Holocene age are the main components of the geology of Wadi Al-Arroub drainage basin and its immediate surrounding. The marls and clays of Qatana Formation and the marl and marly limestones of the Ein Qiniya Formation are the oldest formations, exposed 2-4 kilometers to the west of the study area (Millennium Engineering Group *et al*. 2000). The younger formations of lower Beit Kahil, Upper Beit Kahil, Yatta, Hebron, Bethlehem and Jerusalem crop out from west to east (Fig. 3.2). Holocene alluvial deposits cover the wadi floors of the area.

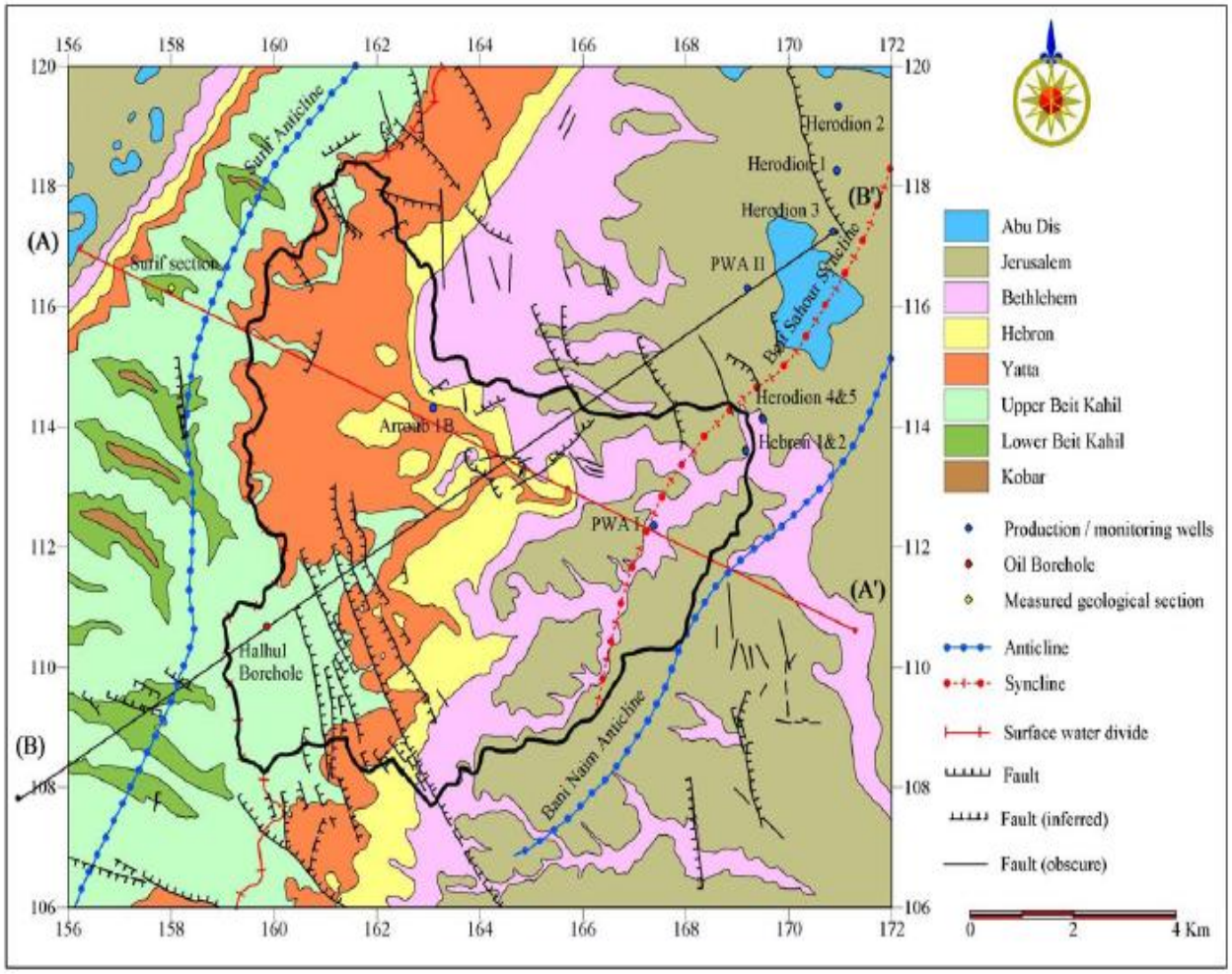

Fig 3.2: A geological and structural map of Wadi Al-Arroub drainage basin (After Qannam, 2003)

3.1.2.1 Cretaceous Rocks:

3.1.2.1.1 Kobar Formation (Aptian to Albian):

Kobar formation is expected to be exposed near Jala Village and to the West of Halhul City. It consists mainly of marl, marly limestone, and limestone. In some locations it is comprised of limestone and intercalation of marl (Shakhnai, 1969 and Millennium Engineering Group, *et al*., 2000).

3.1.2.1.2 Lower Beit Kahil Formation (Albian):

The Lower Beit Kahil Formation consists mainly of limestone, which is well bedded fine crystalline and highly karstic, and sometimes dolomitic in the upper part. This formation has sometimes-intermediate marl layers, and marl increases in downward direction. Its thickness ranges from 120-280 m. In some places the Lower Beit Kahil formation has dark grey dolomite, massively bedded fine crystalline and hard dolomite. It is highly fractured and karstic, though less than lower part (Guttman and Gotlieb, 1996).

3.1.2.1.3 Upper Beit Kahil Formation (Albian):

The Upper Beit Kahil contains light and yellowish limy to marly dolomite, fine crystalline and sometimes soft, which is interbedded with thin marly layers. Its thickness ranges between 40-220 m. In some places it is built up of dolomite and reef limestone, massively bedded to cliff forming, usually coarse crystalline. In the study area this formation is found in Beit Kahil, Jala, Taffuh, Halhul, Baqqar, and parts of Hebron area **(**Owaiwi and Awadallah, 2004).

3.1.2.1.4 Yatta Formation (Cenomanian):

The Yatta Formation consists of yellowish marly limestone, which is contains fine to medium crystalline dolomite and limestone, with marly intercalation, marly at the

bottom. In some places it consists of marly limestone, usually highly enriched with fossilized fauna. Yatta Formation, 86 m thick in Herodion 4 and 128 m in Herodion 3, (Rofe and Raffety, 1963).

3.1.2.1.5 Hebron Formation (Cenomanian):

Hebron formation consists of hard and massive dolomite or limestone. It is highly karstic. Its thickness ranges from 20-120 m. The Hebron Formation without doubt is the most important aquifer within the West Bank. Its Vertical thickness ranges between 70 in Herodion well 4 and 120 m in PWA I (Qannam, 2003).

3.1.2.1.6 Bethlehem Formation (Cenomanian):

The Bethlehem Formation consists of limestone and dolomite, chalky limestone, with marl and rich in faunal remains. In some places it is built up of dolomite, massive and coarse crystalline Limestone lenses, well bedded. The thickness is 80 - 270 m.
Bethlehem Formation is frequently highly jointed and fractured making this formation a good aquifer. Its vertical thickness ranges between 88 m in PWA I and 260 m in Herodion well 4 (Qannam, 2003).

3.1.2.1.7 Jerusalem Formation (Turonian):

The Jerusalem Formation consists mainly of limestone, soft, thin-bedded, dolomitic, chalky and marly limestone. In some place it consists of limestone, hard and massive in other places it consists of hard limestone, dolomitic limestone and marl. Its thickness ranges 90 - 130 m. The Jerusalem Formation has a thickness of about 90-100 m as in Herodion wells 3 and 4. Due to fractures and joints of this formation turns out to be a good aquifer (Qannam, 2003).

3.1.2.1.8 Abu Dis Formation (Senonian):

This formation is part of Senonian age. It consists of chalk and chert, the chalk usually white but in some areas dark colored due to the presence of bituminous

materials. In general chalk often appears to be a fracture aquifer but because of its clayey nature it is considered as an aquiclude (Qannam, 2003). According to ARIJ (1995) the thickness of this formation ranges between 40 and 150 m.

3.1.2.2 Quaternary:

Alluvial formations are of Pleistocene to Recent age, consisting of unconsolidated, laminated marls, clay, silt, gravel and conglomerate. The deposits of this formation cover the floors of all wadis in the study area with thicknesses ranging from less than one meter to 33 meters at the Hebron well 2 (Guttman, Gotlieb 1996 and Qannam, 2003).

3.1.2.3 Karstification:

Karstification is a result of the widening of the joints and fractures, through the dissolution of the carbonate rocks, by CO^{2-} rich percolating water. As the solubility of the dolomite is less and slower than that of the limestone, karst is less developed in the dolomite than in limestone and only minor developed in marl (Milanovic 1981). The dominance of the jointed and fractured carbonate rocks, limestone and dolomite, in the West Bank and the study area, suggests the possible existence of karst caves. According to Arkin (1980), fractures and karstification are common features of the West Bank. There several karst features are to be seen in Wadi Al Arroub drainage basin. Caves are common in the mountains, particularly in the Hebron Formation. One example is the caves from which the spring of El Bas-West is flowing. Such caves are also common between the houses and they are used for animal keeping. The loss of drill fluid during drilling of wells is another example of karstification phenomena (Qannam, 2003).

3.2 Hydrogeology

3.2.1 Introduction:

Groundwater is the main water resource of Palestine. The water table depth is tens of meters in the Pleistocene gravel, to hundreds of meters below the earth's surface in the Lower Cenomanian (Issar 1990). Based on the direction of hydraulic drainage of the Mountain Aquifer was divided into three main groundwater basins; Western, North-Eastern and Eastern Basins. The approximate boundaries between the three basins are shown in Fig.3.3.Only the Eastern Basin lies entirely in the West Bank while the other two are partially in the West Bank (WWS, 2002 and Libiszewski, 1995).

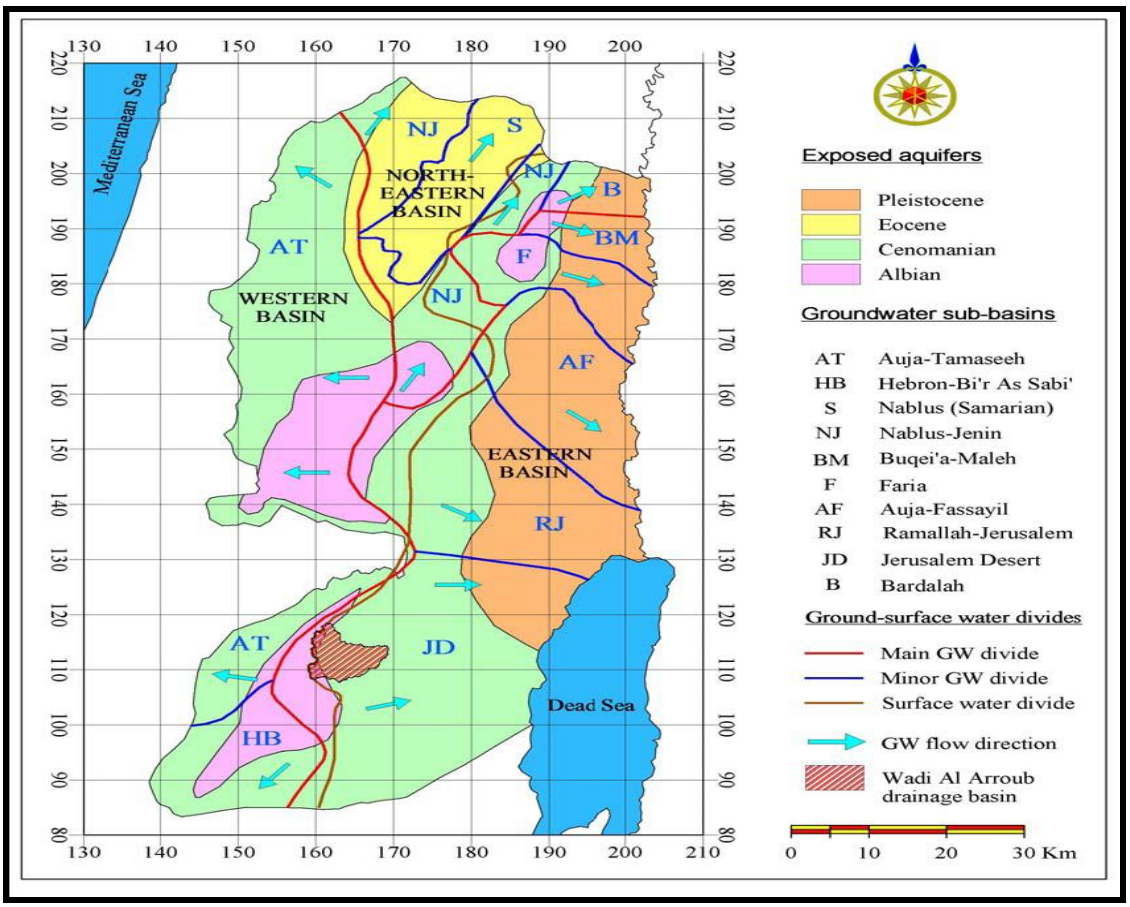

Fig 3.3: Groundwater basins and exposed aquifers in the West Bank / Palestine (After Qannam, 2003).

A combination of Israeli policies had three interconnected results: first, although Palestinian wells rarely reached a depth of more than 100 m, Mekorot's (the Israeli water company) wells were normally sunk to a depth from 200 to 750 m; second, whereas an Israeli well in Central Palestine yields, on average, 1 MCM/year, its Palestinian counterpart yields only 150 000 m^3 /year; and third, the total yield of Palestinian wells was kept at its pre-1967 level of about 36 MCM/year.

3.2.1.1 The western basin:

The western basin is the largest in the formation. Although more than 80% of its recharge area lies in Central Palestine, 80% of the basin itself lies within the Green Line (Israeli boundaries before the 1967 war). Estimates of the western basin's annual renewable yield vary between 310 MCM and 362 MCM. This basin has a recharge area of 1800 km^2 of which 1400 km^2 are in the West Bank, whereas its storage area is about 2500 km^2 (Sturm, *et al*., 1996). The flow direction in this basin is westward towards the Mediterranean Sea.

3.2.1.2 The northeastern basin:

The northeastern basin contains the second largest aquifer in Central Palestine; it yields between 131 and 145 MCM/year. There are several estimates for the recharge and storage capacity of this basin. According to Sturm *et al.*, 1996, this basin has a total area of 700 km^2 out of which 650 km^2 are within the boundaries of the West Bank, while Elmusa (1996) estimated 500-590 km^2. The dominant direction of water movement is northeastwards along the plunge of Nablus-syncline.

3.2.1.3 The eastern basin:

The eastern basin is not an international body of water; the whole basin lies within the boundaries of Central Palestine. Water in this basin flows eastward and discharges into the Jordan River and Dead Sea. The recharge area of this basin encompasses over 2200 km² and the storage area over 2000 km² (Gvirtzman 1994). The safe yield of this basin are not well determined; 100 MCM/year (Elmusa 1996 and Gvirtzman, 1994), 125 MCM/year (Wolf, 1995) and 172 MCM/year (Oslo 2 Accords, 1995).
According to the surface and subsurface hydrological divisions of the West Bank, Wadi Al-Arroub area is a part of the Jerusalem Desert sub-basin and accordingly part of the Eastern Basin (Fig 3.3).

3.2.2 Hydrogeology of Wadi Al-Arroub drainage basin:

The aquifer potentiality of the geological formations of the southern part of the Eastern Basin of the Mountain Aquifer part of which is Wadi Al-Arroub drainage basin is represented in (Table 3.2). The formations thickness and lithology presented in (Table 3.2) are based on the geological log of the wells of Hebron 1, Al-Arroub, Herodion 3 and 4, PWA 1 and 11 (Qannam, 2003).

Table 3.2: Hydrogeological formations at Wadi Al-Arroub drainage basin and its surrounding area (Millennium Engineering Group *et al*. 2000; Guttman and Zuckerman, 1995; Tahal, 1975; Guttman and Gotlieb, 1996, Guttman, 2000 and Qannam, 2003).

Formation name			**Thickness (m)**	**Simplified lithology**	**Classification**
Palestinian	**Israel**				
Jerusalem	Bina		90-130	Hard limestone, some dolomite and marl	Aquifer
Bethlehem	Weradim		70-200	Dolomite and some limestone	Aquifer
	Kfar Shaul		10-70	Soft limestone, chalky limestone and marl	Aquitard
Hebron	Aminadav		20-120	Dolomite and dolomitic limestone	Aquifer
Yatta	Moza	Upper	10-20	Limestone, marly limestone, marl and clay at the bottom,	Aquiclude
	Beit Meir	Upper	40-110	Limestone, dolomite and Marl at bottom	Aquifer
		Lower			Aquiclude
Upper Beit Kahil	Kesalon		10-80	Limestone, dolomite	Aquifer
	Soreq		30-140	Dolomite with marl	Aquifer
Lower Beit Kahil	Givat Yearim		20-80	Limestone, dolomite	Aquifer
	Kefira		100-200	Limestone and marls	Aquifer
kobar	Qatana		30-50	Marl and clay	Aquiclude

3.2.3 Groundwater resources:

3.2.3.1 Groundwater wells:

The wells of Hebron 1 and PWA I are the only deep production wells within the boundaries of Wadi Al-Arroub drainage basin, but these wells are part of a well field, the Herodion-Beit Fajjar well field, extending for a few kilometers to the northeast of the study area. These wells are the main water resource not only for this area but also for about 50 % of the southern West Bank. The water of these wells is being pumped to the area of Wadi Al-Arroub through a network of pipes. The major characteristics of the Herodion wells are summarized in (Table 3.3).

Table 3.3: The deep wells supplying the area of Wadi Al-Arroub and about 50 % of the southern West Bank with tap water (Guttman and Zuckerman, 1995 and CDM, 1997).

Well	Coordination (E/N)	Aquifer	Altitude of well head (masl)	Depth (m)	Pump. rate (m^3/hr)
Hebron 1	169156/113619	Albian	696	704	300
PWA I	167424/112301	Cen.-Tur. and Albian	746	601	250
PWA II	169186/116304	Albian	752	851	250
Beit Fajjar	169696/115208	Cen -Tur.	736	237	230
Herodion 1	170772/118220	Cen.-Tur.	580	351	128
Herodion 2	170958/119332	Albian	560	770	340
Herodion 3	170857/117241	Albian	610	800	400
Herodion 4	169485/114160	Albian	686	691.5	240
Arroub Nursery	162542/114974	ArrLPA	840	50	7

3.2.3.2 Springs:

The dryness of most springs and the decrease in the flow rate of some springs such as that of the El Bas-West and El Bas-East could be attributed to the dry climate that generally dominates the area since 1993. Expansion of the housing over the local surrounding of the springs reducing their recharge areas as well as the growing number of the agricultural dug wells (open pits) could be additional factors that kept these springs with little flow rate or to dryness even in the good rainy season of 1994-1995 and 2000-2001. The number of the shallow agricultural wells (open pits) increased by about 400 % in the last ten years. Especially on properties owned by the inhabitants who depend to 90 % on farming especially crops freshly eaten. The water of many of these springs and shallow wells was used for domestic purposes, especially in the case of interrupted tap water supply. Springs such as El Bas-West and El-Bas east are always used for domestic purposes.

3.2.4 Climate:

The West Bank is a part of Palestine has a typical Mediterranean climate with two distinct seasons: dry hot season from June to October, and cold wet season from November to May (Husary et al., 1995). And so according to PNAMO (1998 and 1999) the area of Wadi Al-Arroub as part of the West Bank has a warm-humid Mediterranean climate. Meteorological Station in the study area Wadi Al-Arroub is the climatological reference which is located on 162100 E/114700 N (PG) and at an elevation of 865 masl.

3.2.4.1 Rainfall:

The quantity of the annual rainfall from year to year is considerable variable, Although the average annual rainfall recorded at Al-Arroub Meteorological Station for the period 1953 –2001 is 607.1 mm, The maximum recorded annual rainfall was 1200 mm in the 1991- 1992 season, while the minimum was 212 mm in 1998-1999 (Fig. 3.4).

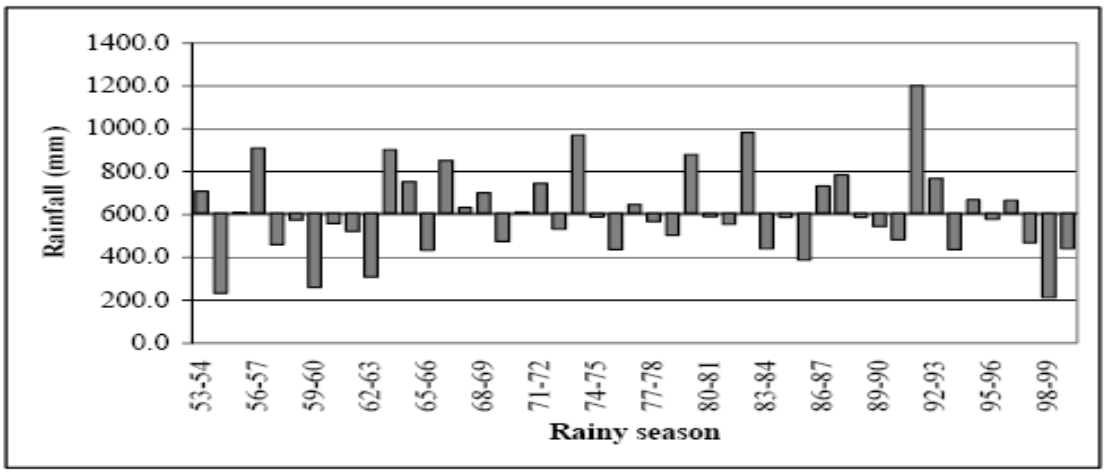

Fig. 3.4: The annual rainfall variations at the Arroub Meteorological Station during the period 1953-2001 (After Qannam, 2003).

In general the study area Wadi AL-Arroub wet season begin almost from October to May, but the most rainfall occurs during the period November to April. About two thirds of the rainfall amount falls between December and February (Fig. 3.3). Not only 1959-1960 and 1961-1962 were very special seasons as the rain started in September, but also 1991-1992 was very special as June appeared to be the end of that rainy season (Qannam, 2003).

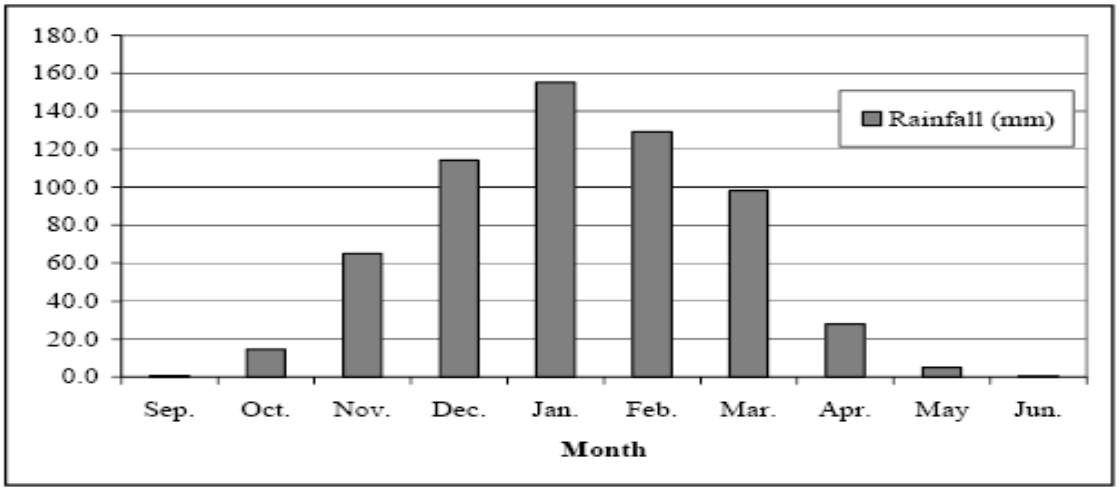

Fig. 3.5: The average monthly rainfall recorded at Al-Arroub Meteorological Station for the period 1953-2001 (After Qannam, 2003).

3.2.4.2 Temperature:

Summer average Temperature in the West Bank varies between 20 and 23 °C, reaching a maximum of 43 °C. The average long term winter temperature is 10 to 11 °C with a minimum of 3 °C. These variations are expected because of the differences in position, elevation, and distance from the coast and the environment around the stations (Ghanem, 1999). The temperature increases from north to south and from west to east on contrary to the altitude. The summer daily temperatures are relatively constant whereas they fluctuate in winter hence, warm and nice days are quickly followed by cold cloudy ones.

During the period (1965-1998), in Wadi Al-Arroub area, the long term average of the monthly mean temperature ranges between 7.5 °C in January and 22.6 °C in August. The average of the monthly maximum temperature varies between 11.6 °C in January and 29.6 °C in August, whereas the average monthly minimum temperature ranges between 3.4 in January and 15.7 °C in July. Fig. 6.4 shows clearly that January is the coldest month and August is the warmest.

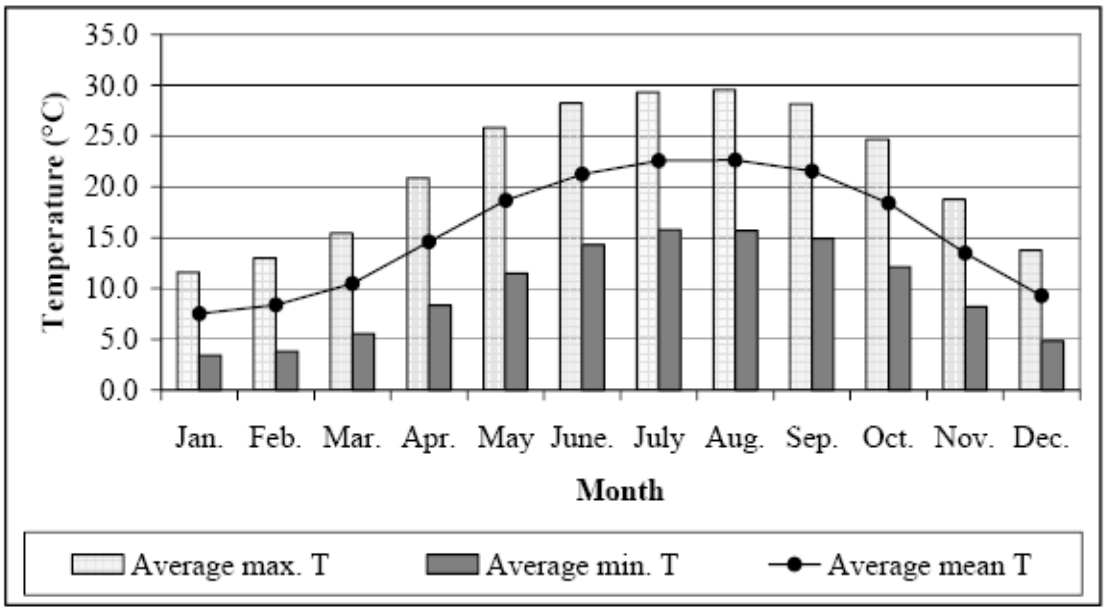

Fig.3.6: Monthly averages of the mean, maximum and minimum temperatures at Al-Arroub Meteorological Station (1965-1998) (Qannam, 2003).

3.2.4.3 Evapotranspiration (ET):

Evapotranspiration is the total water lost from a cropped land due to evaporation from the soil and transpiration by the plants, in other words, is the sum total of water transpired by the plant and that evaporated from the adjacent soil (Kumar and Arora, 1998). Potential evapotranspiration (PET) is the maximum evapotranspiration, which occurs if there is no deficiency of water available in the soil in a fully vegetative basin. The water available is usually less than required for potential evapotranspiration, thus the actual evapotranspiration is the less than potential evapotranspiration. The actual evapotranspiration (AET) represents the actual amounts of transpiration and evaporation (Kumar and Arora, 1998).

According to Qannam, 2003 the evapotranspiration has its highest value of 152 mm in June, and the lowest of 40.5 mm in January. In winter the average value is 125.5 mm/month and in summer the average value is 67.5 mm/month. The annual average of the potential evapotranspiration at Arroub was found to be 1108 mm.

Chapter 4

Results

4.1 Distribution of bacterial types in groundwater wells and springs

Eighty five water samples were collected during the period of the study; the TC bacteria isolated were identified in 17 (52%) out of 33 well water samples and 33 (94%) out of 35 spring water samples. Faecal coliform were not detected in any well water sample tested however 35 (80%) out of 44 spring water samples had FC. Faecal streptococci were detected in 5 (12%) out of 41 well water samples and 38 (86%) out of 44 spring water samples. *Staphylococcus aureus* was identified in 9 (22%) out of 41 well water samples and 36 (82%) out of 44 spring water samples. *Pseudomonas aeruginosa* was detected in 3 (7%) out of 41 well water samples and 8 (18%) out of 44 spring water samples (Tables 4.1, 4.2 and 4.3) and (Figures 4.1, 4.2 and 4.3).

Table 4.1: Distribution of TC, FC, F. streptococci, *S. aureus*, *P. aeruginosa* and TVC in groundwater wells before and after chlorination (Cl_2) in the period between May 2005 and January 2006.

Round # # of samples	**TC**	**FC**	**Faecal streptococci**	***S. aureus***	***P. aeruginosa***	**TVC Before Cl_2 : After Cl_2**
R 1 9 samples	4/9 44.4%	0/9 0%	1/9 11 %	0/9 0 %	1/9 11 %	6/9 : 0/9 67 % : 0
R 2 8 samples	ND	0/8 0 %	1/8 13 %	0/8 0 %	0/8 0 %	5/8 : 0/8 63 % : 0
R 3 9 samples	6/9 67 %	0/9 0 %	2/9 22 %	1/9 11 %	1/9 11 %	5/9 : 0/9 56 % : 0
R 4 6 samples	4/6 67 %	0/6 0 %	0/6 0 %	3/6 50 %	0/6 0 %	1/6 : 0/6 17 % : 0
R 5 9 samples	3/9 33	0/9 0%	1/9 11%	5/9 56%	1/9 11%	2/9 : 0/9 22 % : 0
Total 41 samples	17/33 52%	0/41 0%	5/41 12%	9/41 22%	3/41 7%	18/41 : 0/41 44% : 0%
WHO Guidelines	**0/100ml**	**0/100ml**	**0/100ml**	**0/100ml**	**0/100ml**	**≤500CFU/ ml**

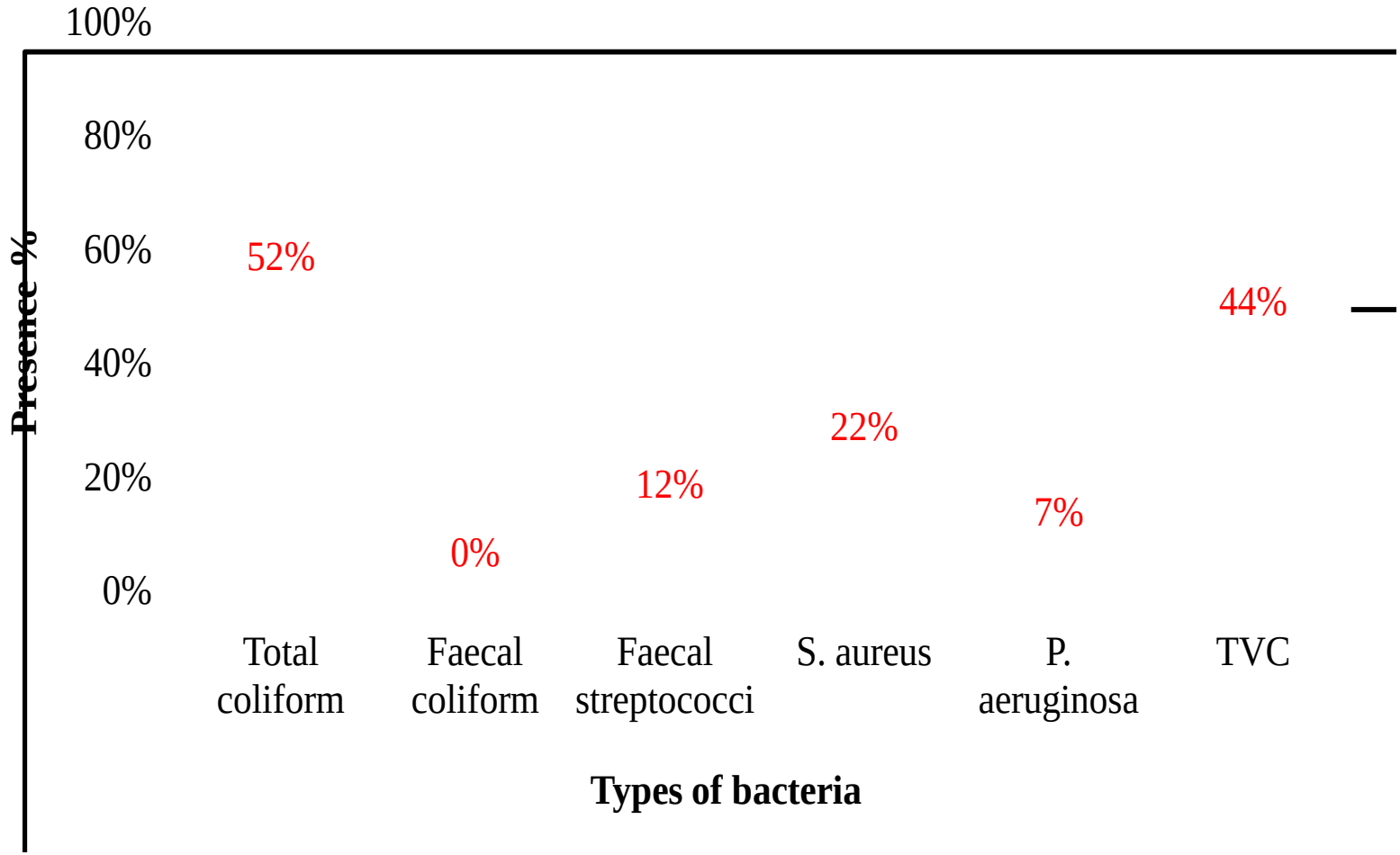

Fig 4.1: Distribution of bacterial types in groundwater wells in the period between May 2005 and January 2006.

Table 4.2: Distribution of Total coliform (TC), Faecal coliform (FC), Faecal streptococci, *Staphylococcus aureus, Pseudomonas aeruginosa* and TVC in groundwater springs before and after chlorination (Cl_2) in the period between May 2005 and January 2006.

Round # # of samples	TC	FC	Faecal streptococci	*S. aureus*	*P. aeruginosa*	TVC Before Cl_2 : After Cl_2
R 1 4 samples	4/4 100 %	3/4 75 %	2/4 50 %	2/4 50 %	0/4 0 %	4/4 : No Cl2 100 % :
R 2 9 samples	ND	7/9 80 %	8/9 90 %	4/9 44 %	0/9 0 %	9/9 : No Cl_2 100 % :
R 3 11 samples	10/11 91 %	10/11 91 %	11/11 100 %	11/11 100 %	3/11 27 %	11/11 : No Cl_2 100 % :
R 4 11 samples	10/11 91 %	9/11 82 %	9/11 82 %	10/11 91 %	2/11 18 %	8/11 : No Cl_2 73 % :
R 5 9 samples	9/9 100 %	6/9 67 %	8/9 90 %	9/9 100 %	3/9 33 %	9/9 : No Cl_2 100 % :
Total 44 samples	33/35 94 %	35/44 80 %	38/44 86 %	36/44 82 %	8/44 18 %	41/44 : No Cl_2 93 % :
WHO Guidelines	0/100ml	0/100ml	0/100ml	0/100ml	0/100ml	≤500CFU/ ml

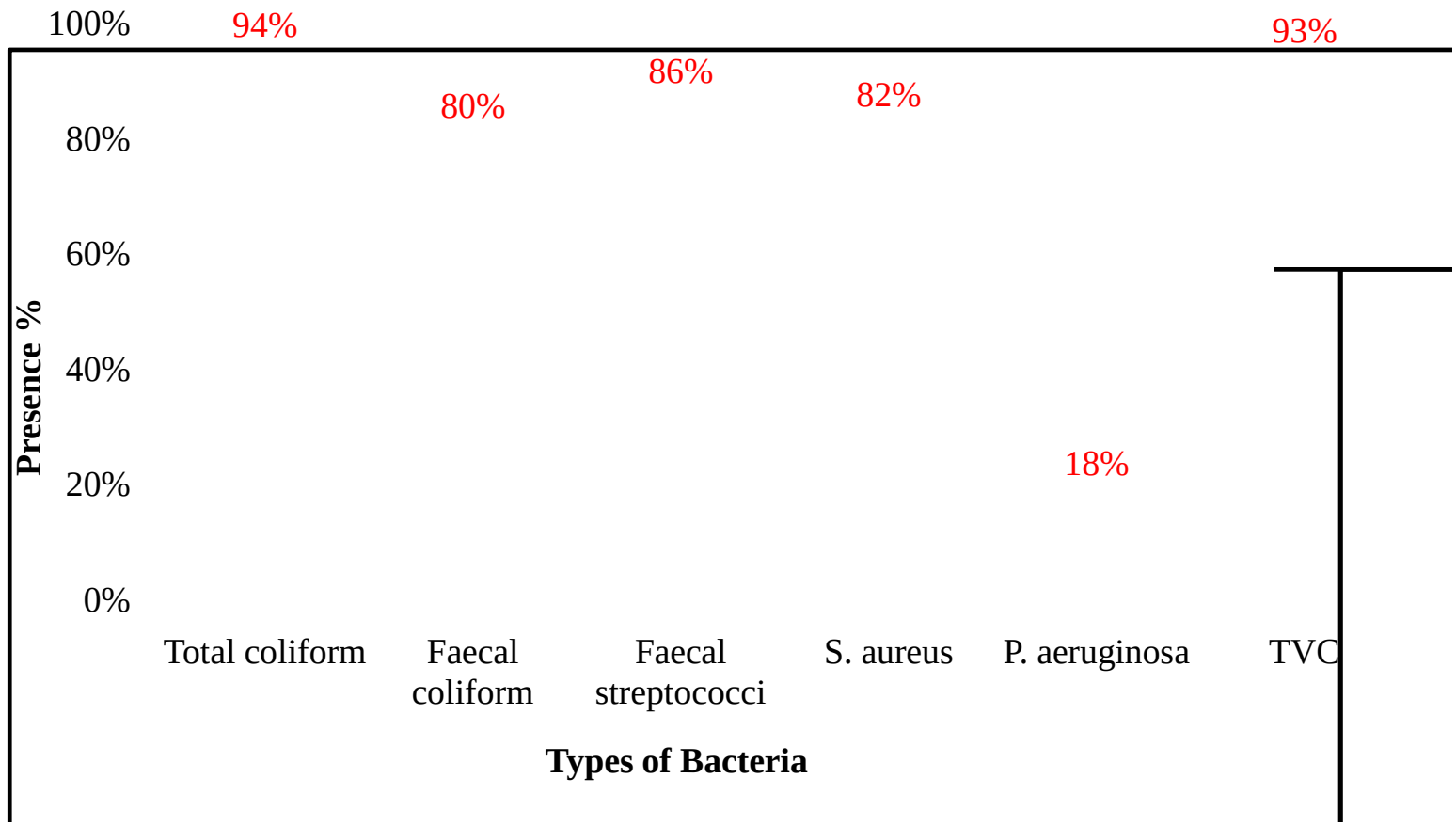

Fig 4.2: Distribution of bacterial types in groundwater springs in the period between May 2005 and January 2006.

Table 4.3: Distribution of bacterial types in all rounds samples of groundwater wells and springs in the period between May 2005 and January 2006.

Rounds # # of samples Months	T. coliform	F. coliform	Faecal streptococci	*Staphylococcus aureus*	*Pseudomonas aeruginosa*
Round # 1 13 samples (May 05)	8/13 (62%)	3/13 (23%)	3/13 (23%)	2/13 (15%)	1/13 (8 %)
Round # 2 17 samples (June 05)	Not Done (ND)	7/17 (41%)	9/17 (53%)	4/17 (24%)	0/17 (0 %)
Round # 3 20 samples (August 05)	16/20 (80%)	10/20 (50%)	13/20 (65%)	12/20 (60%)	4/20 (20%)
Round # 4 17 samples (November 05)	14/17 (82%)	9/17 (53%)	9/17 (53%)	13/17 (77 %)	2/17 (12 %)
Round # 5 18 samples (January 06)	12/18 (67%)	6/18 (33%)	9/18 (50%)	14/18 (78%)	4/18 (22 %)
Total # of samples (85)	50/68 (74%)	35/85 (41%)	43/85 (51%)	45/85 (53%)	11/85 (13%)
WHO Guidelines	0/100ml	0/100ml	0/100ml	0/100ml	0/100ml

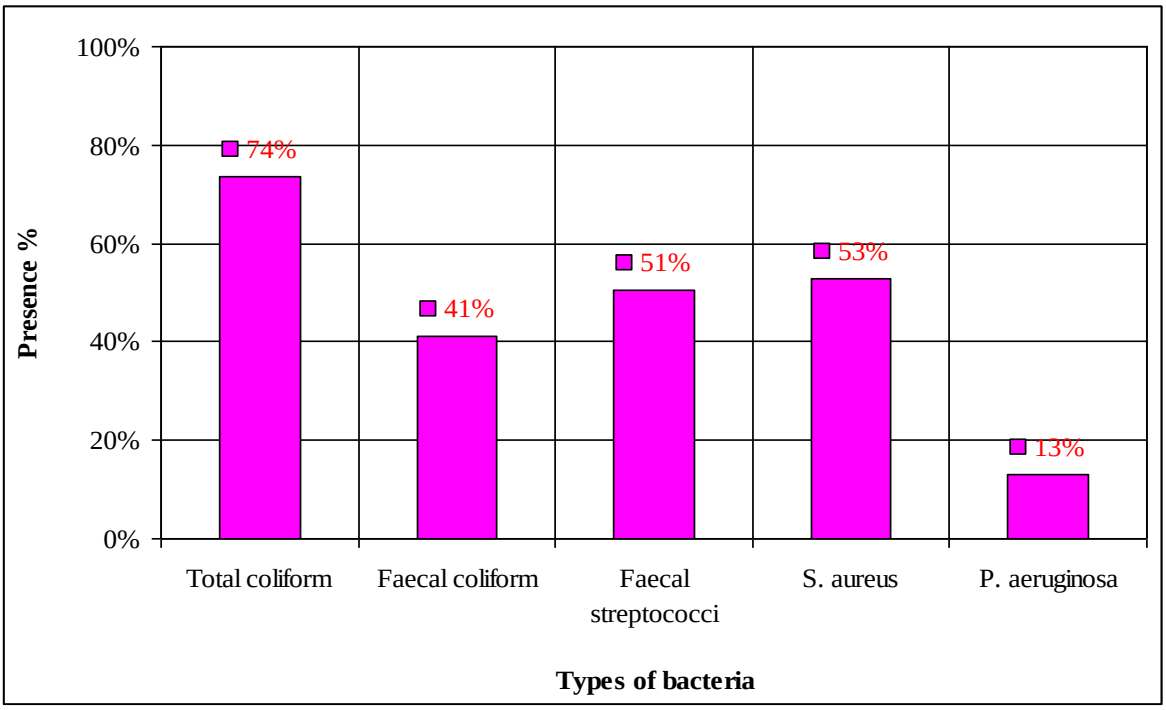

Fig 4.3: Distribution of bacterial types in groundwater wells and springs in the period between May 2005 and January 2006.

4.2 Distribution of indicator bacteria in groundwater wells and springs

4.2.1 Distribution of Total coliform (TC):

Total coliform bacteria isolated were identified in 17 out of 33 (52%) well water samples and 33 out of 35 (94%) spring water samples (Tables 4.1 and 4.2)

The percentage of positive samples for Total coliform bacteria were always higher in groundwater springs than groundwater wells (Figures 4.4 and 4.5).

The percentage of water samples from groundwater wells positive for Total coliforms was lowest (33%) in January and highest (67%) in August and November. The percentage of water samples from groundwater springs positive for Total coliforms varied between 100% and 91% in the period between May 2005 and January 2006.

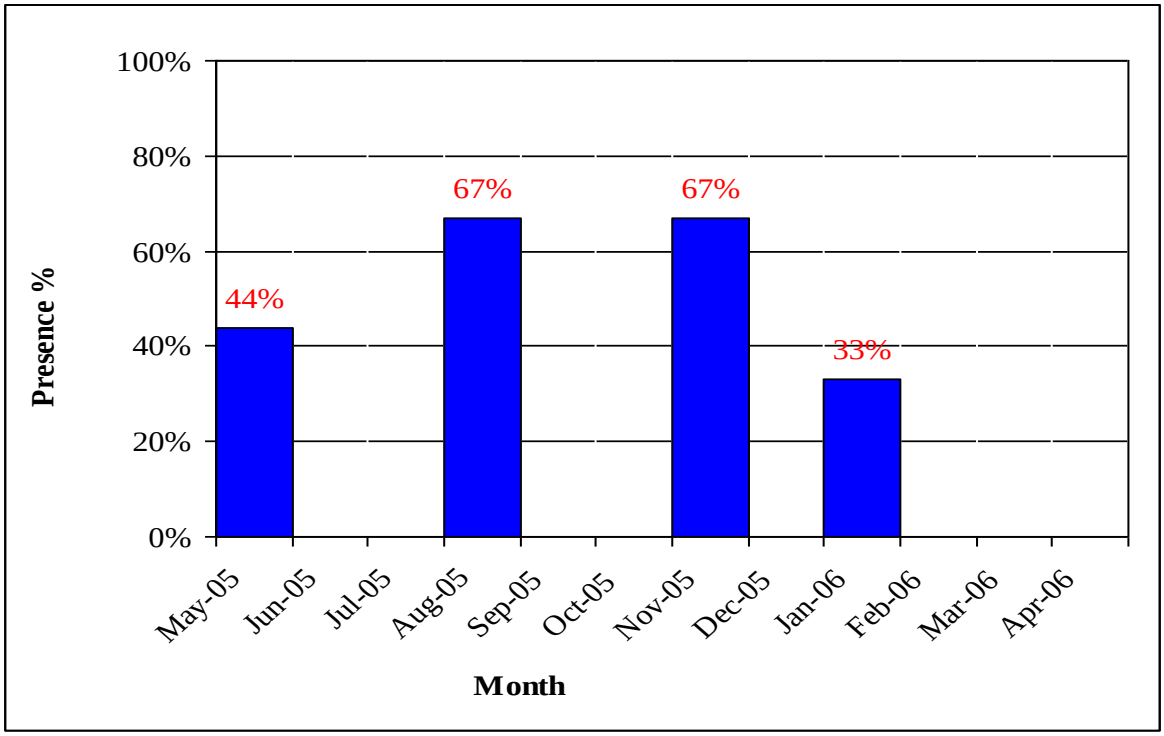

Fig 4.4: Distribution of Total coliform in groundwater wells in the period between May 2005 and January 2006.

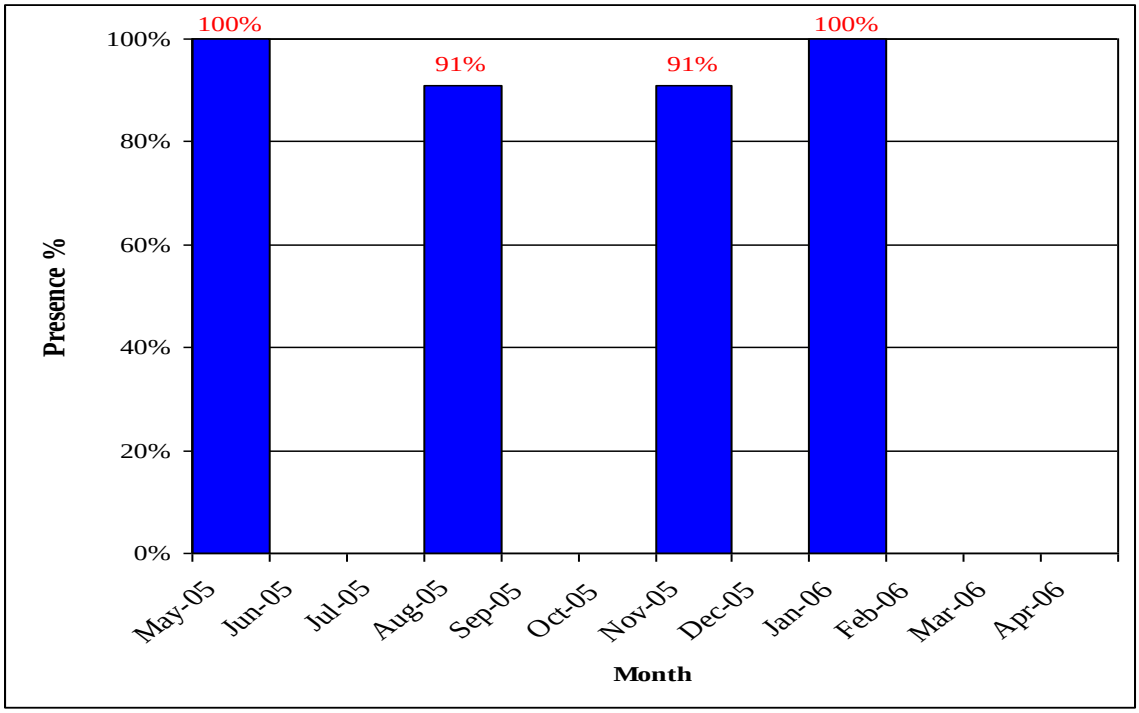

Fig 4.5: Distribution of Total coliform in groundwater springs in the period between May 2005 and January 2006.

4.2.2 Distribution of Faecal coliform (FC):

Faecal coliforms (FC) were not detected in all well water samples for all rounds in different months, but 35 out of 44 spring water samples (80 %) were positive for Faecal coliform (Tables 4.1 and 4.2, and Fig 4.6).

The presence of Faecal coliforms in groundwater springs varied between 67% in January and 91% in August. The higher presence is very likely related to the decrease in the amount of spring water during the summer months.

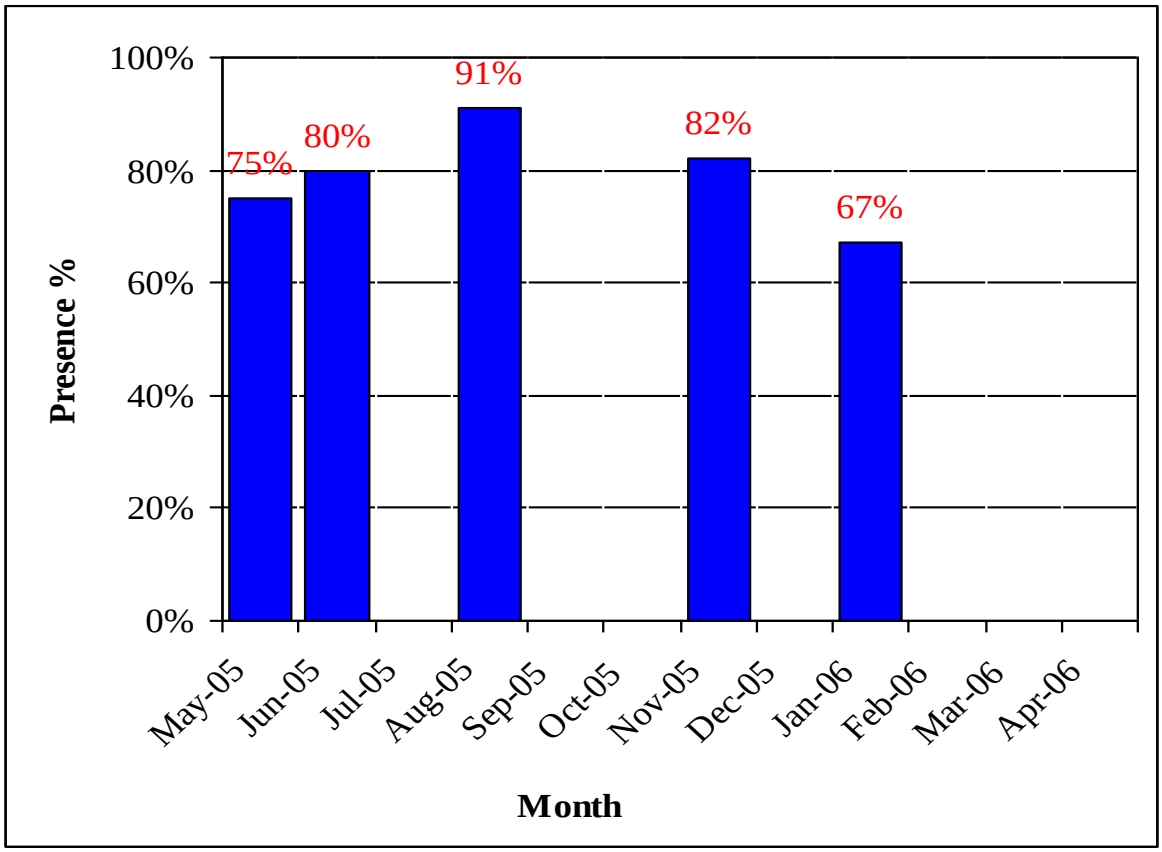

Fig 4.6: Distribution of Faecal coliforms in groundwater springs in the period between May 2005 and January 2006.

4.2.3 Distribution of Faecal streptococci:

Faecal streptococci were detected in 5 (12%) out of 41 well water samples and 38 (86%) out of 44 spring water samples (Tables 4.1 and 4.2). The distribution of Faecal streptococci in groundwater wells (Fig 4.7) was lower than Faecal streptococci in groundwater springs (Fig 4.8) in the period between May 2005 and January 2006. The percentage of water samples from groundwater wells positive for Faecal streptococci varied between 0% in November and 22% in August. Whereas the percentage of water samples from groundwater springs positive for Faecal streptococci varied between 50% in May and 100% in August. The higher presence in August is very likely related to the decrease in the amount of water in the spring and to the increase in temperature during the summer months.

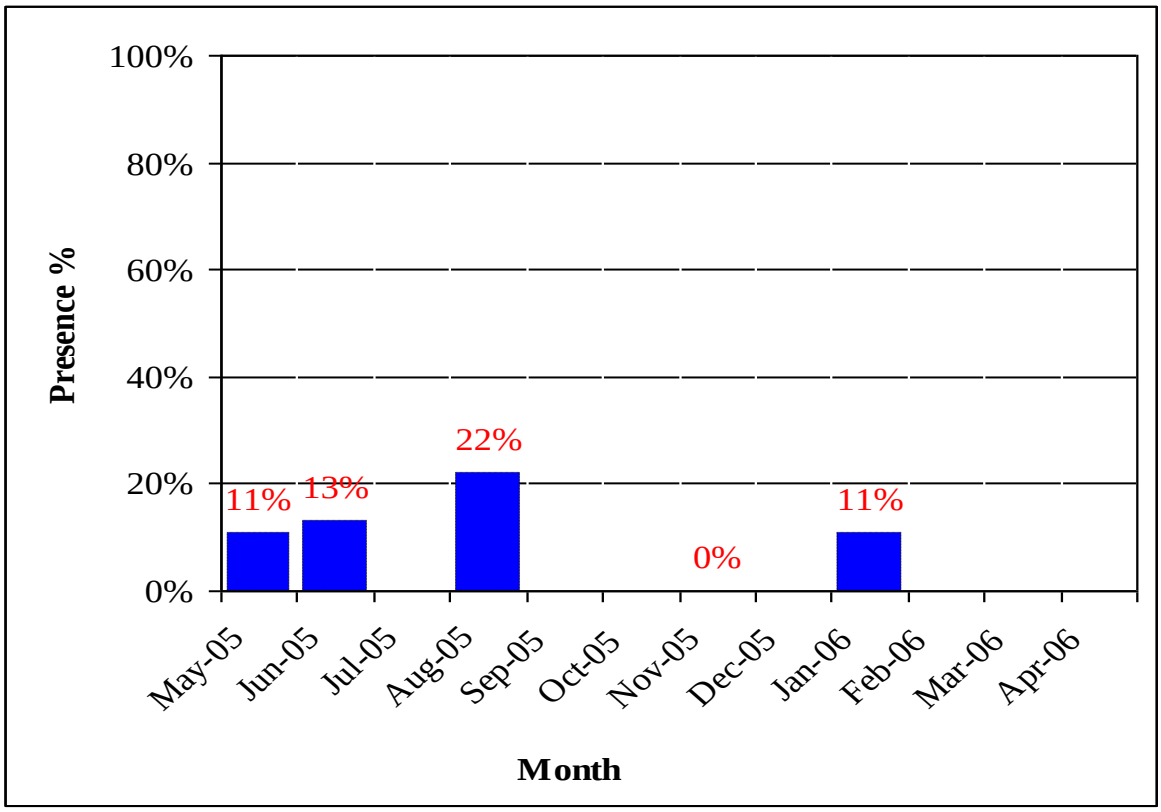

Fig 4.7: Distribution of Faecal streptococci in groundwater wells in the period between May 2005 and January 2006.

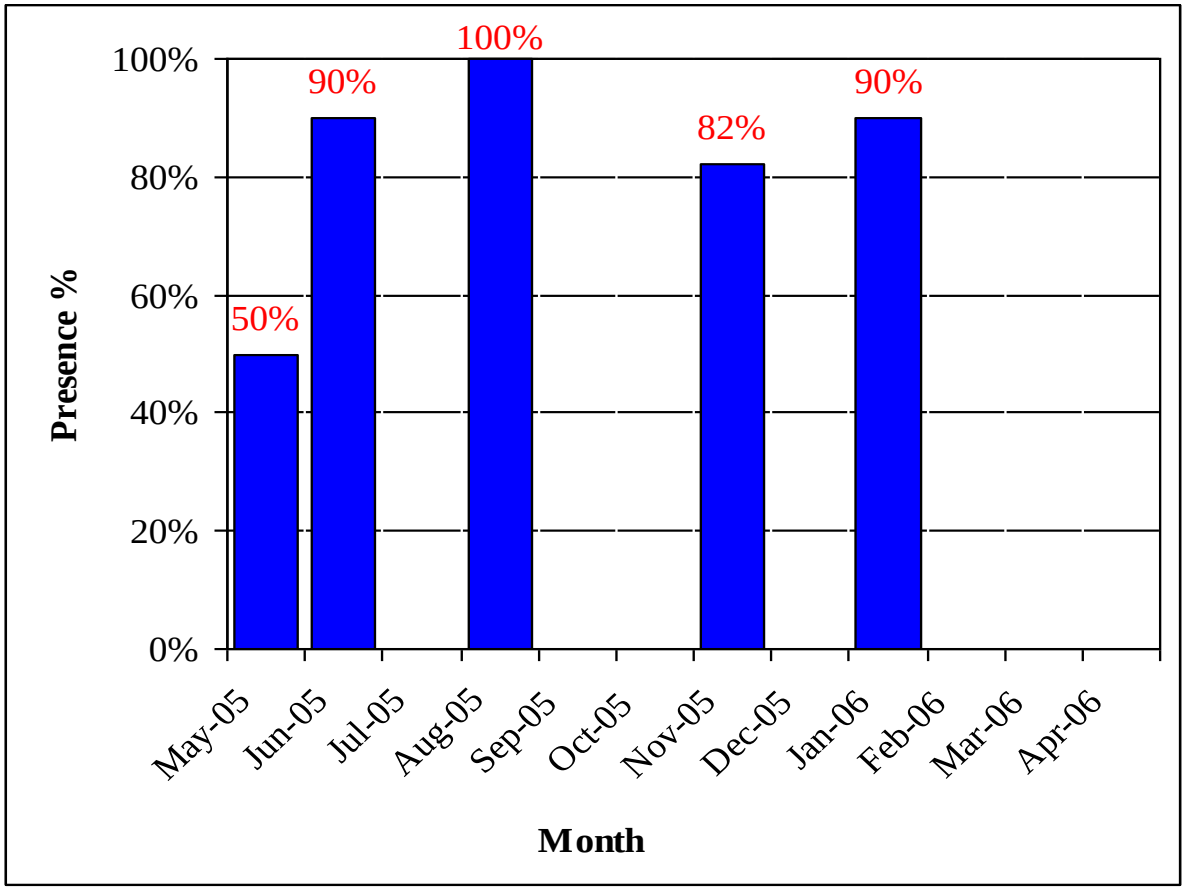

Fig 4.8: Distribution of Faecal streptococci in groundwater springs in the period between May 2005 and January 2006.

4.2.4 Distribution of Total Viable Count (TVC):

Forty four percent of groundwater well samples had a Total Viable Count (TVC) more than 500 CFU/1 ml before chlorination. Whereas 41 (93 %) out of 44 spring water samples had a TVC more than 500 CFU/1 ml. The TVC in 44 % well and 93 % spring water samples exceeds the limit of World Health Organization (WHO) guidelines for drinking water, where the number of TVC permitted is ≤ 500 CFU/ ml (Tables 4.1 and 4.2). The well water is always chlorinated before being pumped to be distributed. However spring water is consumed without chlorination.

4.3 Distribution of Pathogenic bacteria in groundwater wells and springs

4.3.1 Distribution of *Staphylococcus aureus:*

Staphylococcus aureus was detected in 9 (22%) out of 41 well water samples and 36 (82%) out of 44 spring water samples (Tables 4.1 and 4.2). *S. aureus* was detected in groundwater wells less frequently than in springs in the period between May 2005 and January 2006 (Figures 4.9 and 4.10). The presence of *S. aureus* in groundwater wells varied between 11% in August and 56% in January. The presence of *S. aureus* in groundwater springs was 100% in August and in January.

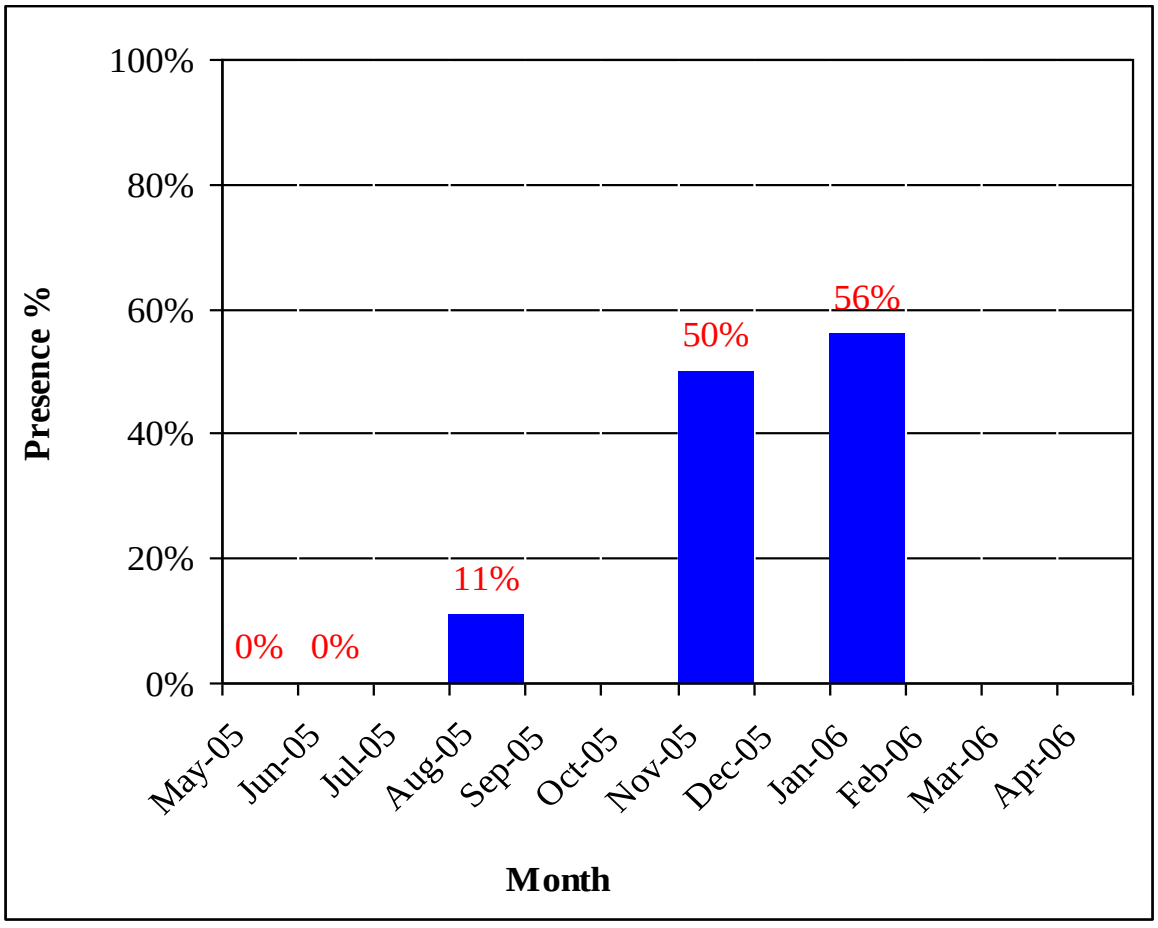

Fig 4.9: Distribution of *Staphylococcus aureus* in groundwater wells in the period between May 2005 and January 2006.

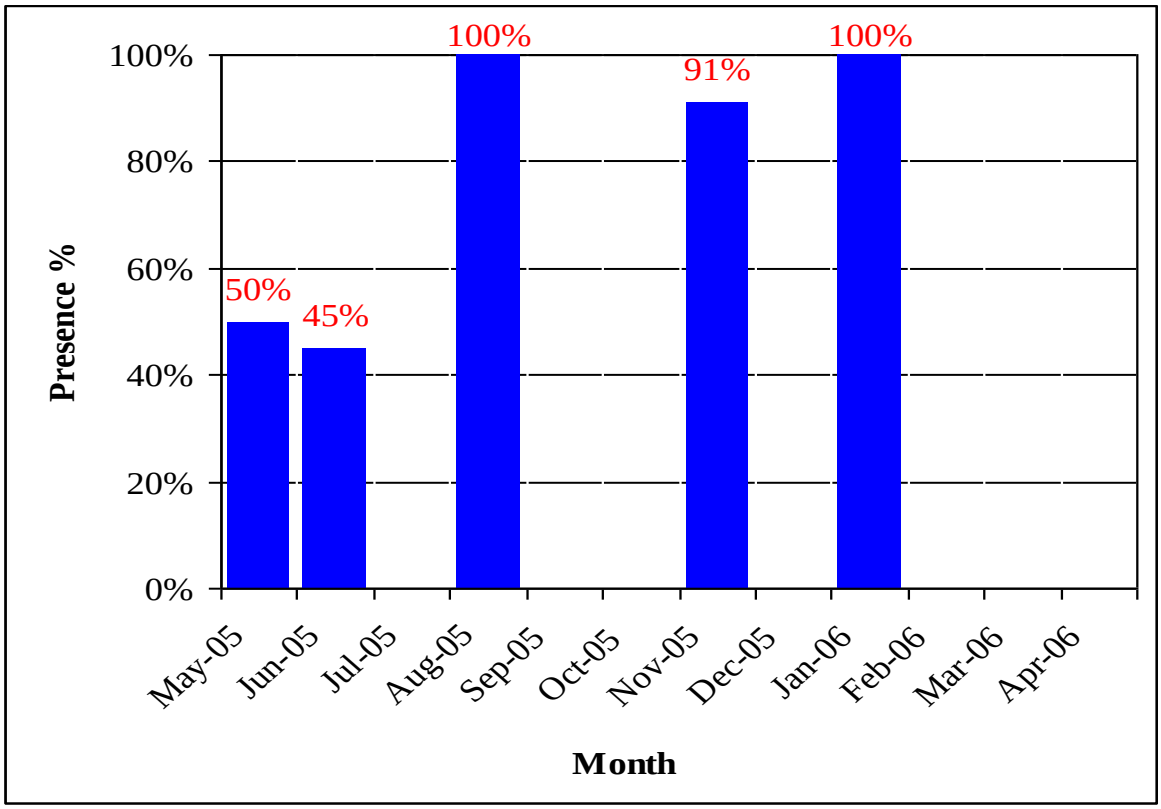

Fig 4.10: Distribution of *Staphylococcus aureus* in groundwater springs in the period between May 2005 and January 2006.

4.3.2 Distribution of *Pseudomonas aeruginosa*:

Pseudomonas aeruginosa was detected in 3 (7%) out of 41 well water samples and 8 (18%) out of 44 spring water samples (Tables 4.1 and 4.2).

P. aeruginosa was detected in groundwater wells less frequently than in groundwater springs in the period between May 2005 and January 2006 (Figures 4.11 and 4.12). The distribution varied between 0% and 11% in groundwater wells. Whereas the distribution of *P. aeruginosa* varied between 0% and 33% in springs in the period between May 2005 and January 2006.

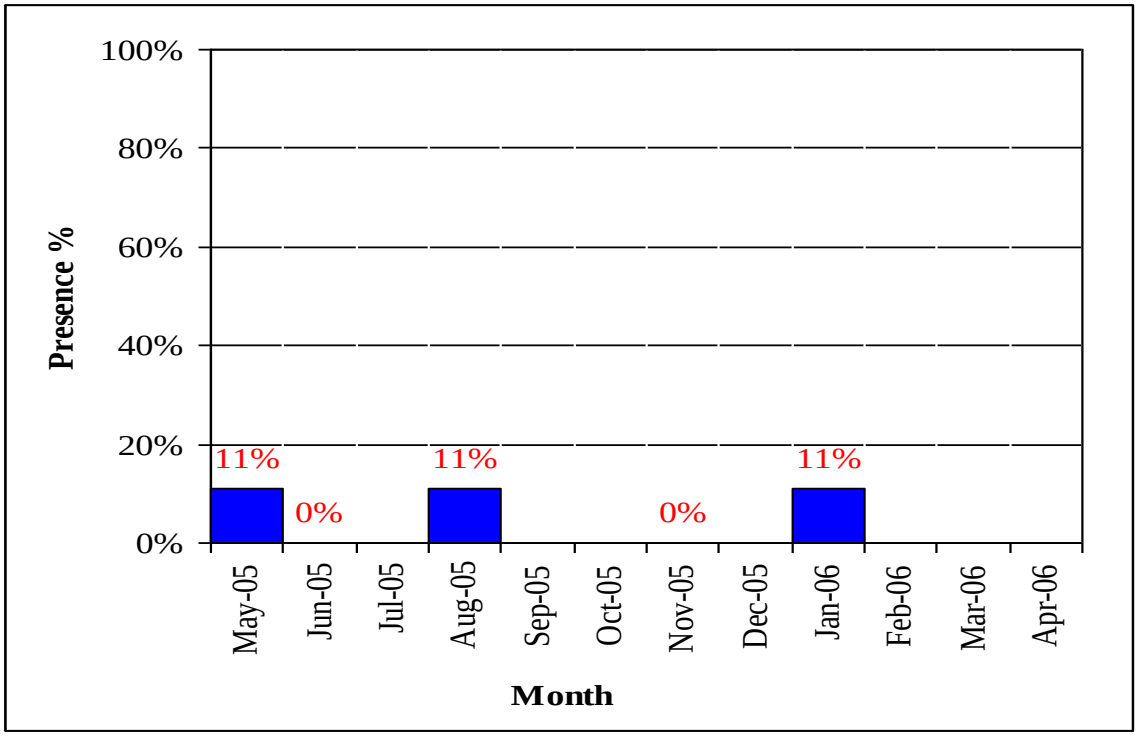

Fig 4.11: Distribution of *Pseudomonas aeruginosa* in groundwater wells in the period between May 2005 and January 2006.

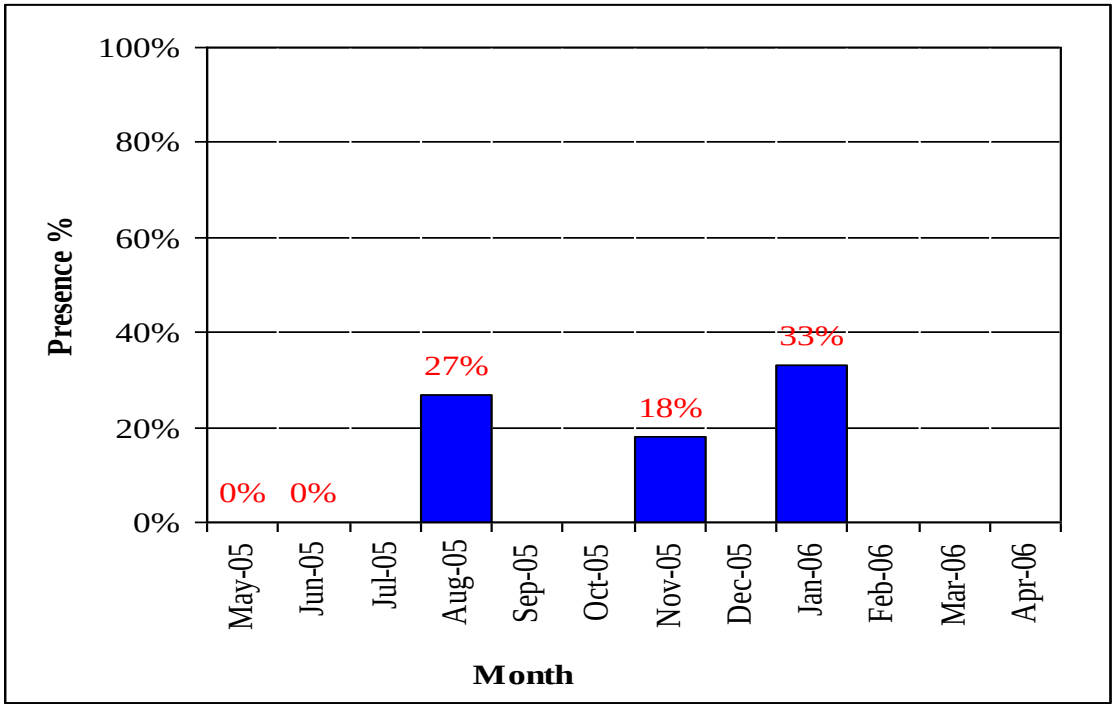

Fig 4.12: Distribution of *Pseudomonas aeruginosa* in springs in the period between May 2005 and January 2006.

4.4 Distribution of Parasite Pathogens

4.4.1 *Entameoba histolytica*:

Entameoba histolytica was not detected in any sample tested from groundwater springs (Table 4.4).

4.4.2 *Giardia intestinalis*:

Giardia intestinalis pathogen was not detected in any sample tested from groundwater springs (Table 4.4).

Table 4.4: Distribution of parasite pathogens in groundwater springs of Wadi Al-Arroub drainage basin.

Sample No	Site Name	Location	Sampling Date	*Entamoeba histolytica*	*Giardia intestinalis*
1	Sa"ir	Hebron	6/11/2005	-ve	-ve
2	Al-Therweh	Halhoul	6/11/2005	-ve	-ve
3	Ein Al-bus (east)	Al-Arroub	6/11/2005	-ve	-ve
4	Ein Al-Dilbi	Halhoul	1/21/2006	-ve	-ve
5	Ein Al-Shinnar	Halhoul	1/21/2006	-ve	-ve
6	Ein Almarj	Al-Arroub	1/21/2006	-ve	-ve
7	Ein Al-Arroub	Al-Arroub	1/21/2006	-ve	-ve

4.5 Differentiation of Total coliform

The Total coliform bacterial isolates were identified in 17 samples (52%) out of the 33 well water samples analyzed in this study. Total coliform differentiation into *Enterobacter, Klebsiella, Citrobacter, Escherichia coli* and Faecal coliform was identified as 48 %, 9%, 12%, 15% and 0% respectively. However the Total coliform bacterial isolates were identified in 33 samples (94%) out of the 35 spring water samples. Total coliform differentiation into *Enterobacter, Klebsiella, Citrobacter, Escherichia coli* and Faecal coliform was identified as 46%, 54%, 46%, 71%, and 80% respectively which indicates more serious springs water contamination (Table 4.5), (Figures 4.13 and 4.14). The identification of *E. coli* in 5/33 (15%) of well water samples and in 35/44 (80%) of spring water samples is an indication of feacal contamination of these water sources.

These results were obtained by using Kligler's iron agar (APHA, 1995) and were confirmed by using the Enterotube in the fourth and fifth rounds.

Table 4.5: Overall distribution of *T. coliform* differentiation in both wells and springs of Wadi Al-Arroub drainage basin in the southern part of the West Bank in Palestine.

Sources	Wells	Springs	Total (%)
Total Samples	33	35	68
Total coliform No. (%)	17/33 (52 %)	33/35 (94 %)	50/68 (74 %)
Enterobacter spp No. (%)	16/33 (48 %)	16/35 (46 %)	32/68 (47 %)
Klebsiella spp No. (%)	3/33 (9 %)	19/35 (54 %)	22/68 (32 %)
Citrobacter spp No. (%)	4/33 (12 %)	16/35 (46 %)	20/68 (29 %)
E. coli No. (%)	5/33 (15 %)	25/35 (71 %)	30/68 (44 %)
Faecal coliform No. (%)	0/41 (0 %)	35/44 (80 %)	35/85 (41 %)

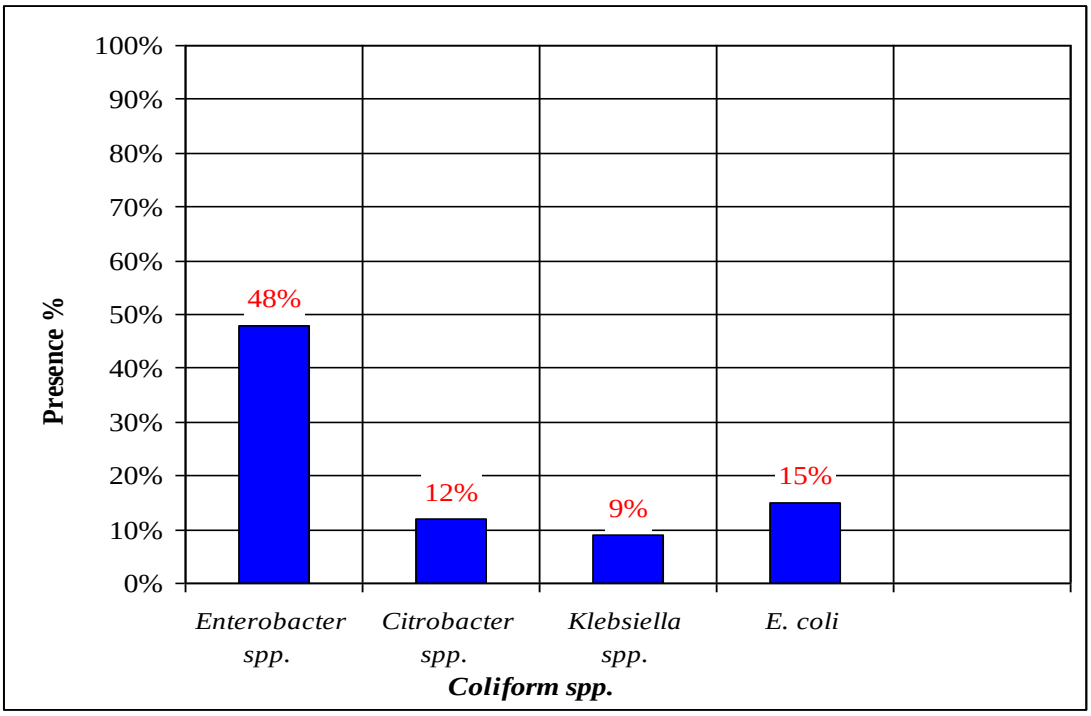

Figure 4.13: Differentiation of Total coliform in groundwater wells in the period between May 2005 and January 2006.

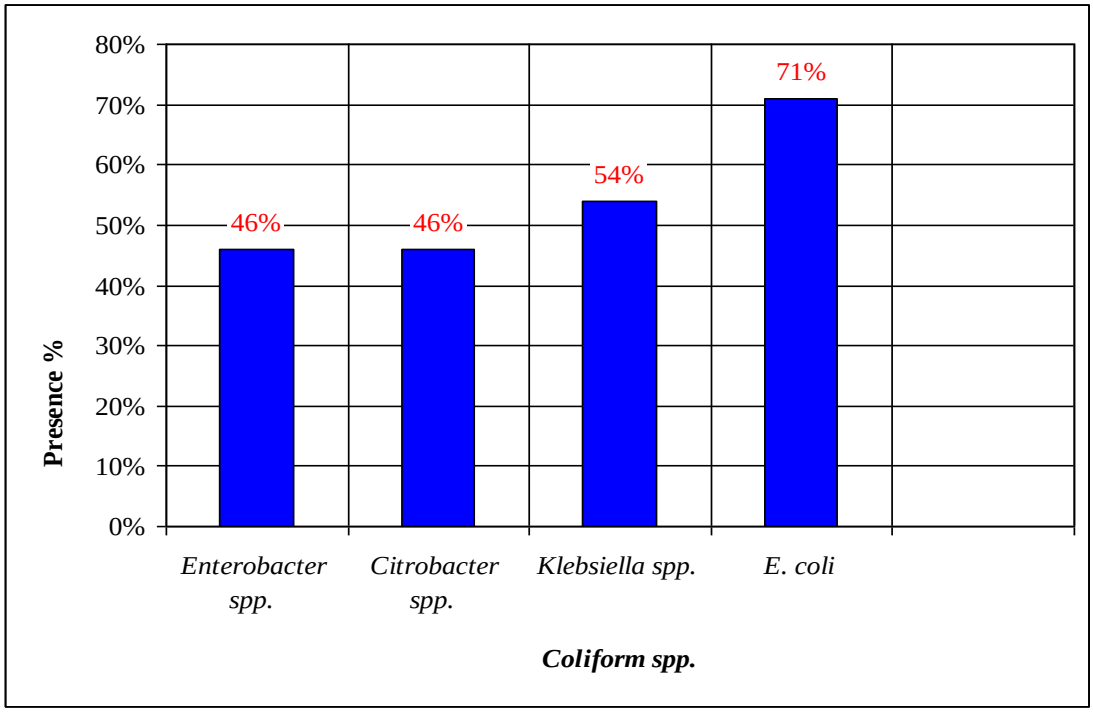

Figure 4.14: Differentiation of Total coliform in groundwater springs in the period between May 2005 and January 2006.

4.6 Nitrate

The nitrate (NO_3) level was higher in spring water samples than well water samples (Tables 4.6 and 4.7) and (Figures 4.15 and 4.16). None of the groundwater well samples tested had nitrate levels above 45 mg/liter recorded in November. However in January 3/9 (33 %) of the samples had nitrate levels above 45 mg / liter. Whereas 15/20 (75%) of spring water samples had nitrate levels above 45 mg/liter in the two months (November and January).

The presence % of nitrate above WHO guidelines in groundwater spring samples was only slightly higher in January 78% than in November 73%.

Table 4.6: Nitrate level in groundwater well samples of Wadi Al-Arroub drainage basin.

Round No. Month	No. of samples	No. of samples with $NO_3 > 45$ mg/l	No. of samples with $NO_3 < 45$ mg/l
Round No.4 November 05	6	0/6 (0 %)	6/6 (100 %)
Round No. 5 January 06	9	3/9 (33 %)	6/9 (67 %)
Total	15	3/15 (20 %)	12/15 (80 %)
WHO Acceptable level	$NO_3 < 45$ mg/l		

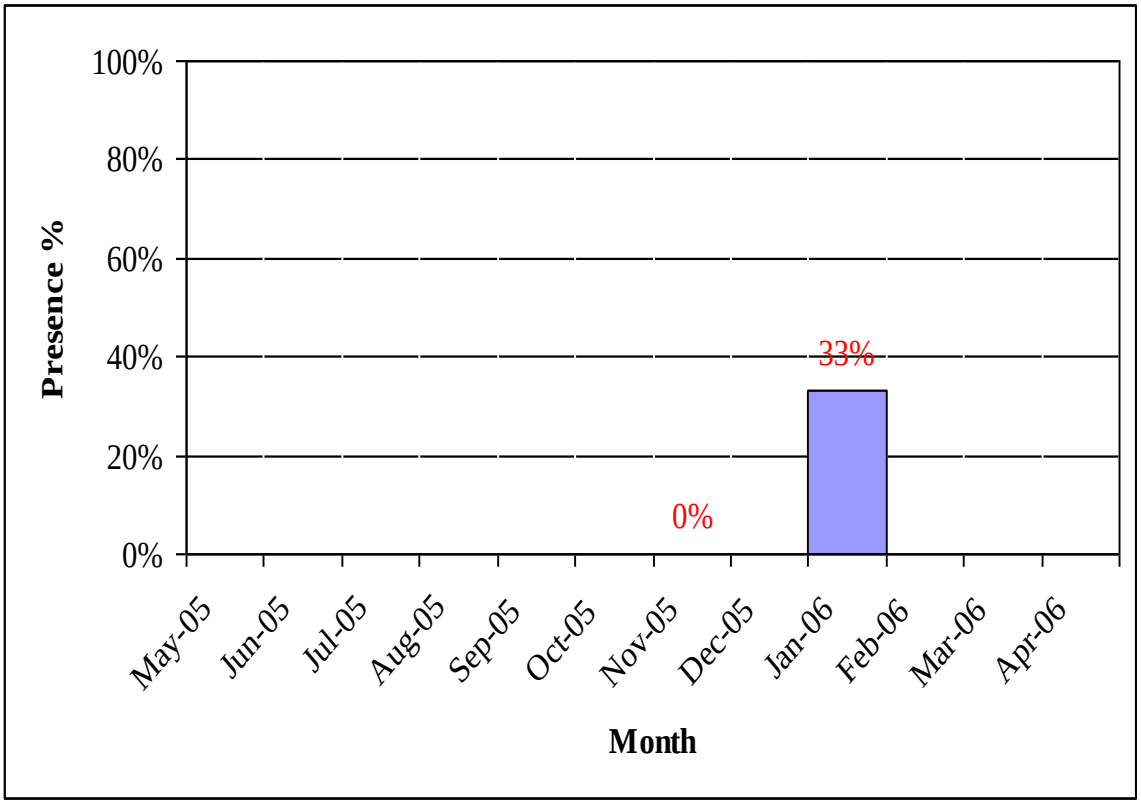

Fig 4.15: Nitrate levels in groundwater well samples that exceed WHO guidelines in November 2005 and January 2006.

Table 4.7: Nitrate level in groundwater springs samples of Wadi Al-Arroub drainage basin

Round No. Months	No. of samples	No. of samples with NO_3 > 45 mg/l	No. of samples with NO_3 < 45 mg/l
Round No.4 November 05	11	8/11 (73 %)	3/11 (27 %)
Round No. 5 January 06	9	7/9 (78 %)	2/9 (22 %)
Total	20	15/20 (75 %)	5/20 (25 %)
WHO Acceptable level	NO_3 < 45 mg/l		

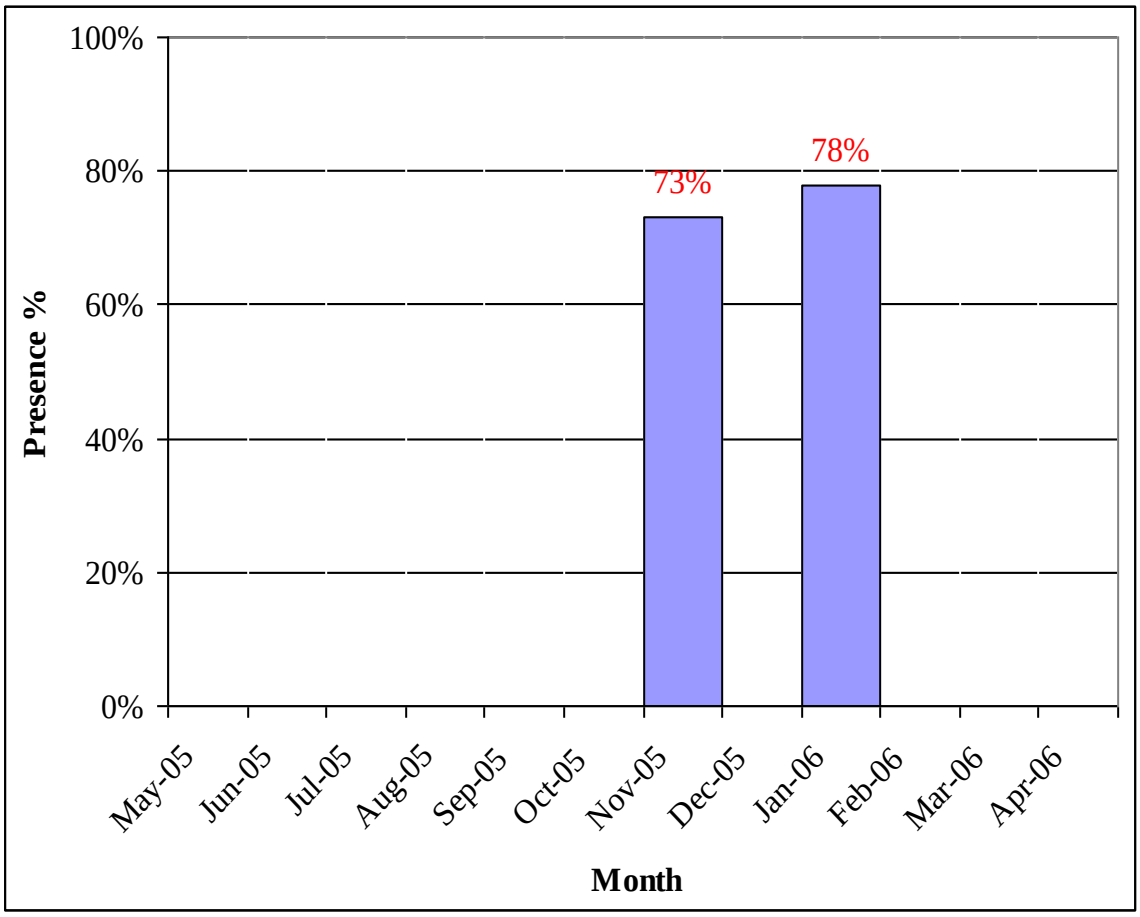

Fig 4.16: Nitrate levels in groundwater spring samples that exceed WHO guidelines in November 2005 and January 2006.

4.7 The efficiency of chlorination systems

Chlorinated water samples of groundwater well had no colony forming units (CFU) when analyzed for Total coliform (TC) and Faecal coliform (FC) bacteria. Also all of these groundwater well samples had a Total Viable Count (TVC) less than 500 CFU/ ml after chlorination which agrees with the WHO guidelines for drinking water ≤ 500 CFU/ml (Table 4.7). The chlorination system used for groundwater wells in Wadi Al-Arroub drainage basin is efficient.

Table 4.8: Total coliform (TC), Faecal coliform (FC) bacteria and Total Viable Count (TVC) before and after chlorination system of groundwater wells of Wadi Al-Arroub drainage basin in the period between May 2005 and January 2006.

Round # # of samples	**Before chlorination**			**After chlorination**		
	TC	FC	TVC≥500 CFU/ ml	TC	FC	TVC≥500 CFU/ ml
R 1 9 samples	4/9 (44 %)	0/9 (0%)	6/9 (67 %)	0/4 (0%)	0/9 (0%)	0/9 (0 %)
R 2 8 samples	Not done (ND)	0/8 (0%)	5/8 (63 %)	0/0 (0%)	0/8 (0%)	0/8 (0 %)
R 3 9 samples	6/9 (67 %)	0/9 (0%)	5/9 (56 %)	0/6 (0%)	0/9 (0%)	0/9 (0 %)
R 4 6 samples	4/6 (67 %)	0/6 (0%)	1/6 (17 %)	0/4 (0%)	0/6 (0%)	0/6 (0 %)
R 5 9 samples	3/9 (33%)	0/9 (0%)	1/9 (11 %)	0/3 (0%)	0/9 (0%)	0/9 (0%)
Total 41 samples	17/33(52%)	0/41(0%)	18/41 (44%)	0/41(0%)	0/41(0%)	0/41 (0 %)
WHO Guidelines	0/100ml	0/100ml	≤500CFU/ ml	0/100ml	0/100ml	≤500CFU/ ml

4.9 Microbiological quality of groundwater wells and springs

The number of indicator bacteria such as Total coliform (TC), Faecal coliform (FC) and Faecal streptococci were always higher in springs water than in deep wells in all rounds in different months.

Also the number of pathogenic bacteria *Staphylococcus aureus* (*S. aureus*) and *Pseudomonas aeruginosa* (*P. aeruginosa*) were less prevalent in groundwater wells than groundwater springs in all rounds at different months.

All groundwater well samples analyzed for Total Viable Count TVC, Total coliform (TC) and Faecal coliform (FC) after chlorination system agreed with WHO guidelines standards. There is no chlorination system applied for spring's water.

Chapter 5

Discussions and Recommendations

5.1 Discussion

Microbiological analysis of water is used to monitor the microbiological content and its suitability for drinking water and domestic use. In general, microbiological analysis includes determining the Total Viable Count (TVC) as well as detecting indicator organisms or potential pathogens. Bacteria and viruses are the most important microorganisms in water microbiology (APHA, 1995).

This study focused on determining Total Viable Count, before and after chlorination, Total coliform bacteria (TC), Faecal coliform and Faecal streptococci which are sensitive and commonly used indicators of bacterial pathogen contamination of natural waters (Schaffter and Parriaux, 2002). Total coliform bacteria were further differentiated into *Enterobacter, Klebsiella, Citrobacter* and *Escherichia coli*. Differentiation of Total coliform helps in the identification of *Escherichia coli* a very strong indicator of faecal contamination of water; similarly, the detection of Faecal streptococci. The presence of these organisms makes the presence of potential pathogens very likely such as *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The presence of two important protozoan pathogens *Entameoba histolytica* and *Giardia intestinalis* was also tested.

Total coliform bacteria are found in animal and human faeces, but not all total coliform bacteria are necessarily of faecal origin. These indicator bacteria have been found to be useful for monitoring safe drinking and recreational waters (Greenberg *et al.,* 1992). The Faecal coliform group is a subset of the Total coliforms. These bacteria are thermotolerant and can grow at 44.5°C. Faecal coliform bacteria are used to determine faecal contamination of water, because they have an excellent positive correlation with faecal contamination (Greenberg *et al.,* 1992). If one Faecal coliform per 100 ml of water is detected, the water source is considered unsafe for drinking (Greenberg *et al.*, 1992 and USEPA, 1998).

The nitrate level in the groundwater wells and springs tested was also monitored during the period of study May 2005 till January 2006, an important indicator of contamination of the water source with materials, originating primarily from fertilizers, septic systems or manure storage or spreading.

More than half of the well water samples tested had Total coliform bacteria.Whereas 94% of the spring water samples had Total coliform. The identification of these Total coliforms showed that 15% were *E. coli* in groundwater wells and 71% were *E. coli* in spring water samples. The presence of *E. coli* is a very strong indicator of feacal contamination. Obviously the spring water has a much higher degree of faecal contamination which may be directly related to the hydrogeological factors that influence groundwater pollution such as infiltration mechanisms, length of transport (Matthess, 1981), type of soil, filtration, (Lance and Gerba, 1984), adsorption of microorganisms (Matthess, 1985), and permeability of the aquifer which decrease the traveling time of microorganism to the groundwater (Gerba, 1984).

The depth of the wells varies between 50-851m. As water infiltrates to the aquifer, microorganisms, especially bacteria, are filtered or adsorbed by soil and rocks. The filtration of spring water is less due to the short lengths of transport of spring water. The concentration of bacteria is decreasing with increasing depth of wells, TVC taken as an example in round number five (Fig 5.1). This relationship is not applicable to Safi 1 well where the well depth is 704 m, but it has 800 CFU/1 ml; this due to geological characteristics like karstification features with high porosity and permeability facilitating smooth infiltration into the aquifer.

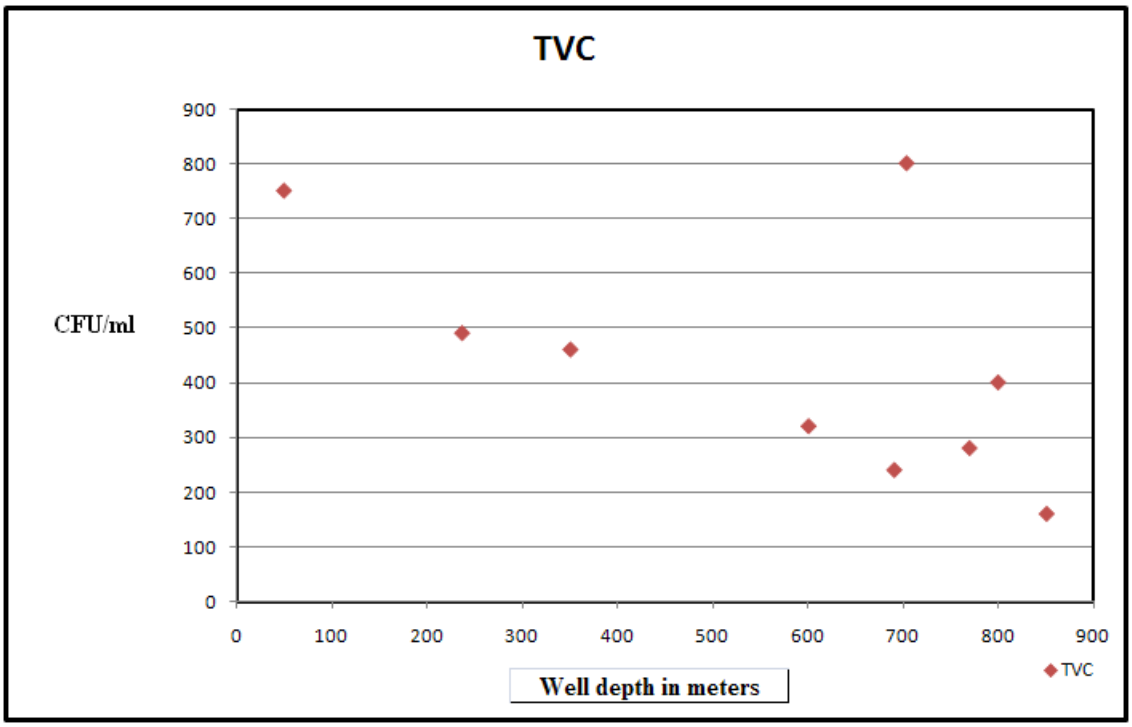

Fig 5.1: The relationship between Total Viable Count (TVC) and Well depth.

Faecal coliform bacteria another important indicator of faecal contamination was not found in groundwater deep well samples (Table 4.1). In contrast, 80% of spring water samples contained Faecal coliform (Table 4.2). Another indicator of higher levels of contamination of spring water. These results comply with previous studies (Celico, *et al*., 2003) done in Italy.

The contamination of groundwater aquifer may be due to many reasons; infiltration of bacteria by wastewater from cesspits that are built for houses next to springs. Infiltration by wastewater stream along the valley. Low depth of groundwater table of aquifer that allows bacterial infiltration. The nature and characteristic of soil and rocks with several karst features seen in the study area and high porosity and permeability facilitating smooth infiltration into the aquifer.

The source of contamination by different types of bacteria especially FC are mainly wastewater from cesspits and wastewater stream. In addition wastes from animals and

different landfill-leachates that are spread in many places in the study area contribute to water contamination.

The presence of indicator bacteria (TC, FC and F. streptococci) in groundwater was higher in shallow springs than deep wells (Tables 4.1 and 4.2). This is directly related to the water flow through the aquifer, shorter time and less opportunity for the bacteria to contact and adhere to sediments and rocks. Contamination of surface flow and groundwater from animal waste has been well documented (Mallin, 1997). These results agree with the study by (James, et al., 2000).

Forty four percent of groundwater well samples had a Total Viable Count (TVC) above the acceptable level before chlorination. However Total Viable Count for groundwater well samples was less than 500 CFU/ ml after chlorination which agrees with the WHO guidelines for drinking water (WHO, 2004). This indicates that the chlorination system is efficient. Chlorine is a strong disinfectant that is effective in inactivating bacteria and viruses and under certain circumstances Giardia (USEPA, 1989).
Human pathogens like *Pseudomonas aeruginosa* and *Staphylococcus aureus* were less prevalent in groundwater wells than groundwater springs in all rounds in different months. The presence of F. streptococci, *S. aureus*, and *P. aeruginosa* in groundwater well samples indicates that the concentration of these bacteria in rainfall is very high. Water flow through the aquifer decreases the number of bacteria that reaches these deep wells however it does not allow significant self purification of polluted water.

These results show that well water is a safer source of water. Spring water tested had Faecal coliform indicating faecal contamination, also higher rate of human pathogens like *Pseudomonas aeruginosa* and *Staphylococcus aureus* in all rounds in different months during this study making it unsuitable for human consumption and farming crops that are eaten fresh.

The nitrate (NO_3) concentration (< WHO guideline 45 mg/l) is higher in spring water samples (75%) than in well water samples (20%) (Tables 4.4 and 4.5). Nitrates are a major component of fertilizer and wastewater. They also result from the breakdown of organic matter buried in the soil and from Nitrogenfixation. Excess nitrates in drinking water could be the result of a number of things: the overuse of fertilizers in farming in the recharge area of the well; the presence of septic effluent in the groundwater supplies caused by failed or failing septic system or runoff containing animal wastes. Drinking water that has high levels of nitrate can cause serious illness in infants under the age of six months. This condition is called Maethaemoglobinemia or "blue baby" syndrome, and can result in death (Shearer, *et al.*, 1972 and Drinking water and health, 1977). Nitrate is not readily removed by filtration or other common domestic water treatment systems. The best method for limiting nitrate in well water is by controlling nearby sources of nitrate (Drinking water and health, 1977).

These results agree with previous studies done on drinking water and swimming pool water in Tulkarem area where nitrate levels were found to exceed WHO guidelines (Kan'an, 2005)

Protozoal pathogens such as *E. histolytica* and *G. intestinalis* were not detected in spring water samples (Table 4.4) in spite of the presence of bacterial indicators and pathogens. This may be related to the size of protozoal pathogen that filtered and adhered along the distance soil and rocks through infiltration of water to the aquifer (Schijven and Hassanisadeh, 2000).

In general, the percentage of Total coliform (TC), Faecal coliform (FC), Faecal streptococci, *Staphylococcus aureus* and *Pseudomonas aeruginosa* increased in groundwater well and spring samples during the period between May 2005 (dry season) and January 2006 (wet season) (Tables 4.1 and 4.2).
The transport of microorganisms in groundwater is influenced by many factors. A critical factor is water flow, it is the driving force of transport and pathogen transport velocities appear to be proportional to the water flow. Under unsaturated flow conditions, water fills the small pores. This increases the soil microorganism contact and adsorption (Gerba and Bilton, 1984 and Hassanisadeh, 2000)

Groundwater well and spring samples contamination by bacteria during the period of May 2005 and January 2006 showed a series of peaks that were irregularly distributed (Figures 4.1 and 4.2) with several increasing/decreasing phases of bacterial pollution, these may be related to rainfall events that produced effective infiltration. During winter, the percentage of contamination decreased for Faecal streptococci in groundwater wells and springs but not for *Staphylococcus aureus*. This phenomenon may be a function of dilution (significant increase in discharge) (Kibbey and others 1978). These results comply with previous studies (Celico, et al., 2003).

Wadi Al-Arroub drainage basin in the southern part of the West Bank has a population estimated to be about 69,000 inhabitants (Palestinian Central Bureau of Statistics, 2006) It is a densely populated region of Palestine .The results of this study show that the water source from springs for a large number of the inhabitants of this region is not safe for consumption especially that they use it without chlorination. However water from the wells which reach the inhabitants inconsistently is chlorinated through a system established by the Palestinian Authority.

Due to water shortage in this region people use spring water very often. This creates health hazards which results in severe cases of diarrhea as evidenced by the large number of diarrhea cases seen in the health centers in Wadi Al-Arroub area. Mostly affected, are children less than three years, Approximately 65 cases / month are reported compared to 10-12 cases / month for people above three years (UNRWA, 2006). The numbers of cases reported by the main UNRWA health center does not reflect the real number of patients who suffer from diarrhea as many cases are not reported.

Clearly, the present study provided essential information that we believe will be very helpful in evaluating the water quality in Wadi Al-Arroub drainage basin, and sheds light on the health hazards which spring water predisposes the inhabitants of this area to. This study is the first to look extensively into the microbiology of viable organisms in a deep groundwater resource in the West–Bank. Further studies are needed to investigate the microbiology of remaining groundwater resources in the West Bank and Gaza. This research also showed the need for regular water-quality monitoring of the groundwater resources in Palestine.

5.2 Recommendations

- Long-term regular monitoring of the water quality, especially indicator bacteria (Total coliform, Faecal coliform, Faecal streptococci *and* Total Viable Count) and NO_3 concentrations in springs.

- Since spring water is not chlorinated, inhabitants of the region should be advised to boil water before use.

- It is recommended to supply pregnant women and infants with low nitrate bottled water in areas where springs are the major source for domestic purposes.

- Replacement of the present poorly designed cesspits and open waste water conduit with a proper sewage and sanitation system and treatment plant. Where that is not feasible construction of well-designed cesspits (i.e. double-walled and two chambered cesspits). This could provide treated waste water to be used for irrigation rather than utilizing water resources of high drinking water quality. This will also alleviate health hazards to which the present population is subjected.

- Prevention of uncontrolled disposal of waste water in the nearby fields and valleys.

- Prevention of uncontrolled disposal of solid wastes and establishment of infrastructure for environmentally-safe solid waste disposal (i.e. of central sanitary landfills).

- Controlling urban expansion in the study area, which is a recharge area for the aquifers, as it reduces the area of recharge and human activities will pollute the underlying aquifers in the long run.

- Rain water harvesting by means of house cisterns, small scale dams and infiltration basin is recommended in order to increase groundwater recharge in periods with surplus of rain water.

- Rehabilitation of springs (i.e. construction of storage facilities). This offers protection against contaminating activities such as watering of livestock and domestic laundering. Such storage facilitates in-situ water treatment such as disinfection.

- Legislative and executive measures are recommended to control the water quality and the hygiene of the tankers.

- It is not recommended to use water from springs especially those adjacent to the sewage conduit or in the vicinity of houses for drinking purposes unless it is treated properly.

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Appendices

Appendix I: Number of colony forming units (TVC) for TC, FC, TVC, Faecal streptococci, *Staphylococcus aureus* and *Pseudomonous aeruginosa* of groundwater wells and springs in the period between May 2005 and January 2006.

Lab No.	Site ID.	Site Name	Location	Sampling Date M/D/Y	HPC CFU/ ml	T. coliform CFU/100 ml	F. coliform CFU/100 ml	F. streptococci CFU/100ml	*S. aureus* CFU/100ml	*P. aeruginosa* CFU/100ml
1	17-11/001	Herodion # 1	Bethlehem	5/2/2005	2540	0	0	0	0	1
2	17-11/002	Herodion # 2	Bethlehem	5/2/2005	200	0	0	0	0	0
3	17-11/003	Herodion # 3	Bethlehem	5/2/2005	1860	0	0	0	0	0
4	16-11/006	Herodion # 4	Bethlehem	5/2/2005	980	0	0	0	0	0
5	16-11/001A	Beit Fajjar	Bethlehem	5/2/2005	1700	121	0	TMTC	0	0
6	16-11/008	Safi 1	Bethlehem	5/2/2005	3200	6	0	0	0	0
7	16-11/001	PWA 1	Bethlehem	5/2/2005	220	0	0	0	0	0
8	16-11/-11	PWA 11	Bethlehem	5/2/2005	120	1	0	0	0	0
9	CB/50	Sa"ir	Hebron	5/2/2005	TMTC	TMTC	77	0	242	0
10	CB/52	Al-Therweh	Halhoul	5/2/2005	2000	108	140	170	0	0
11	CB/47b	Ein Al-Bas (west)	Al-Arroub	5/2/2005	680	15	0	0	0	0
12		Ein Al-Bas (east)	Al-Arroub	5/2/2005	3200	TMTC	40	16	22	0

13	16-11/004	Arroub-Nursery	Al-Arroub	5/2/2005	TMTC	8	0	0	0	0
14	17-11/001	Herodion # 1	Bethlehem	6/11/2005	2650	N.D	0	0	0	0
15	17-11/002	Herodion # 2	Bethlehem	6/11/2005	180	N.D	0	0	0	0
16	17-11/003	Herodion # 3	Bethlehem	6/11/2005	2640	N.D	0	2	0	0
17	16-11/006	Herodion # 4	Bethlehem	6/11/2005	620	N.D	0	0	0	0
18	16-11/001A	Beit Fajjar	Bethlehem	6/11/2005	1244	N.D	0		0	0
19	16-11/008	Safi 1	Bethlehem	6/11/2005	320	N.D	0	0	0	0
20	16-11/001	PWA 1	Bethlehem	6/11/2005	280	N.D	0	0	0	0
21	16-11/-11	PWA 11	Bethlehem	6/11/2005	TMTC	N.D	0	0	0	0
22	CB/50	Sa"ir	Hebron	6/11/2005	TMTC	N.D	TMTC	0	64	0
23	CB/52	Al-Therweh	Halhoul	6/11/2005	2600	N.D	190	220	0	0
24	CB/47b	Ein Al-Bas (west)	Al-Arroub	6/11/2005	780	N.D	52	96	10	0
25		Ein Al-Bas (east)	Al-Arroub	6/11/2005	1940	N.D	46	27	0	0
26		Ein Al-Warasnah	Al-Arroub	6/11/2005	1650	N.D	6	27	0	0
27		Ein Mesleh	Halhoul	6/11/2005	860	N.D	0	5	0	0
28		Ein Al-Dilbi	Halhoul	6/11/2005	TMTC	N.D	TMTC	TMTC	6	0
29		Ein Ayoub	Halhoul	6/11/2005	2460	N.D	38	52	0	0
30		Ein Al-Shinnar	Halhoul	6/11/2005	2320	N.D	0	6	62	0
31	17-11/001	Herodion # 1	Bethlehem	8/20/2005	800	7	0	0	0	0

32	17-11/002	Herodion # 2	Bethlehem	8/20/2005	1950	14	0	0	0	1
33	17-11/003	Herodion # 3	Bethlehem	8/20/2005	3900	23	0	0	0	0
34	16-11/006	Herodion # 4	Bethlehem	8/20/2005	80	0	0	0	0	0
35	16-11/001A	Beit Fajjar	Bethlehem	8/20/2005	1600	9	0	32	18	0
36	16-11/008	Safi 1	Bethlehem	8/20/2005	550	0	0	0	0	0
37	16-11/001	PWA 1	Bethlehem	8/20/2005	10	0	0	1	0	0
38	16-11/-11	PWA 11	Bethlehem	8/20/2005	10	1	0	0	0	0
39	CB/50	Sa'ir	Hebron	8/20/2005	4300	TMTC	170	360	90	0
40	CB/52	Al-Therweh	Halhoul	8/20/2005	900	370	68	180	40	0
41	CB/47b	Ein Al-Bas (west)	Al-Arroub	8/20/2005	2800	122	68	92	34	0
42		Ein Al-Bas (east)	Al-Arroub	8/20/2005	3700	TMTC	TMTC	TMTC	TMTC	98
43		Ein Al-Warasnah	Al-Arroub	8/20/2005	3200	TMTC	152	380	TMTC	0
44		Ein Mesleh	Halhoul	8/20/2005	3800	0	0	3	13	0
45		Ein Al-Dilbi	Halhoul	8/20/2005	TMTC	340	76	130	110	0
46		Ein Ayoub	Halhoul	8/20/2005	3800	TMTC	76	136	60	0
47		Ein Al-Shinnar	Halhoul	8/20/2005	3600	TMTC	3	TMTC	TMTC	0
48	16-11/004	Arroub-Nursery	Al-Arroub	8/20/2005	130	3	0	0	0	0
49		Ein Almarj		8/20/2005	TMTC	TMTC	TMTC	TMTC	TMTC	1
50		Ein Al-Arroub		8/20/2005	TMTC	TMTC	62	TMTC	TMTC	4

51	17-11/002	Herodion # 2	Bethlehem	12/11/2005	130	0	0	0	0	0
52	17-11/003	Herodion # 3	Bethlehem	11/12/2005	380	32	0	0	0	0
53	16-11/006	Herodion # 4	Bethlehem	11/12/2005	40	120	0	0	38	0
54	16-11/001A	Beit Fajjar	Bethlehem	11/12/2005	620	48	0	0	1	0
55	16-11/-11	PWA 11	Bethlehem	11/12/2005	30	8	0	0	26	0
56	CB/50	Sa"ir	Hebron	11/12/2005	TMTC	TMTC	280	386	120	0
57	CB/52	Al-Therweh	Halhoul	11/12/2005	460	TMTC	3	6	TMTC	0
58	CB/47b	Ein Al-Bas (west)	Al-Arroub	11/12/2005	400	0	0	0	80	0
59		Ein Al-Bas (east)	Al-Arroub	11/12/2005	TMTC	TMTC	164	42	TMTC	0
60		Ein Al-Warasnah	Al-Arroub	11/12/2005	TMTC	TMTC	52	3	TMTC	0
61		Ein Mesleh	Halhoul	11/12/2005	80	15	0	0	18	0
62		Ein Al-Dilbi	Halhoul	11/12/2005	TMTC	TMTC	346	360	TMTC	0
63		Ein Ayoub	Halhoul	11/12/2005	TMTC	TMTC	87	1	TMTC	0
64		Ein Al-Shinnar	Halhoul	11/12/2005	800	TMTC	TMTC	14	TMTC	0
65	16-11/004	Arroub-Nursery	Al-Arroub	11/12/2005	40	0	0	0	1	0
66		Ein Almarj	Al-Arroub	11/12/2005	TMTC	TMTC	420	380	166	24
67		Ein Al-Arroub	Al-Arroub	11/12/2005	TMTC	TMTC	TMTC	16	0	3
68	17-11/001	Herodion # 1	Bethlehem	1/21/2006	460	0	0	0	0	0
69	17-11/002	Herodion # 2	Bethlehem	1/21/2006	280	0	0	0	0	0

70	17-11/003	Herodion # 3	Bethlehem	1/21/2006	400	0	0	0	6	0
71	16-11/006	Herodion # 4	Bethlehem	1/21/2006	240	0	0	0	0	0
72	16-11/001A	Beit Fajjar	Bethlehem	1/21/2006	490	8	0	8	11	4
73	16-11/008	Safi 1	Bethlehem	1/21/2006	800	45	0	0	32	0
74	16-11/001	PWA 1	Bethlehem	1/21/2006	320	0	0	0	0	0
75	16-11/11	PWA 11	Bethlehem	1/21/2006	160	0	0	0	0	0
76	CB/52	Al-Therweh	Halhoul	1/21/2006	2900	350	0	24	16	0
77	CB/47b	Ein Al-Bas (west)	Al-Arroub	1/21/2006	2600	26	0	4	20	0
78		Ein Al-Bas (east)	Al-Arroub	1/21/2006	TMTC	20	7	110	76	0
79		Ein Al-Warasnah	Al-Arroub	1/21/2006	3200	80	4	8	20	6
80		Ein Mesleh	Halhoul	1/21/2006	760	32	0	0	3	0
81		Ein Ayoub	Halhoul	1/21/2006	2600	260	30	16	22	6
82		Ein Al-Shinnar	Halhoul	1/21/2006	3000	50	29	12	64	2
83	16-11/004	Arroub-Nursery	Al-Arroub	1/21/2006	750	30	0	0	4	0
84		Ein Almarj	Al-Arroub	1/21/2006	3800	250	32	82	66	0
85		Ein Al-Arroub	Al-Arroub	1/21/2006	2680	60	23	32	110	0
Total samples		85 samples								
WHO Guidelines					≤500CFU/ ml	0/100ml	0/100ml	0/100ml	0/100ml	0/100ml

Appendix II: Number of colony forming units (TVC) for TC, FC and TVC before and after chlorination systems built on groundwater wells.

Lab. No.	Well / Spring No.	Well / Spring Name	Sampling date M/D/Y	Before Chlorination			After Chlorination		
				T. coliform CFU /100ml	F. coliform CFU/ 100ml	TVC CFU /1ml	TVC CFU /1ml	T. coliform CFU /100ml	F. coliform CFU/ 100ml
1	17-11/001	Herodion # 1	5/2/2005	0	0	2540	150	0	0
2	17-11/002	Herodion # 2	5/2/2005	0	0	200	30	0	0
3	17-11/003	Herodion # 3	5/2/2005	0	1860	0	0	0	0
4	16-11/006	Herodion # 4	5/2/2005	0	1860	0	0	0	0
5	16-11/001A	Beit Fajjar	5/2/2005	121	0	1700	60	0	0
6	16-11/008	Safi 1	5/2/2005	6	0	3200	0	0	0
7	16-11/001	PWA 1	5/2/2005	0	0	220	0	0	0
8	16-11/11	PWA 11	5/2/2005	1	0	120	80	0	0
9	16-11/004	Arroub Nursery	5/2/2005	8	0	TMTC	NF	NF	NF
10	17-11/001	Herodion # 1	6/11/2005	ND	0	2650	300	ND	0

11	17-11/002	Herodion # 2	6/11/2005	ND	0	180	90	ND	0
12	17-11/003	Herodion # 3	6/11/2005	ND	0	2640	340	ND	0
13	16-11/006	Herodion # 4	6/11/2005	ND	0	620	20	ND	0
14	16-11/001A	Beit Fajjar	6/11/2005	ND	0	1244	30	ND	0
15	16-11/008	Safi 1	6/11/2005	ND	0	320	250	ND	0
16	16-11/001	PWA 1	6/11/2005	ND	0	280	0	ND	0
17	16-11/-11	PWA 11	6/11/2005	ND	0	TMTC	0	ND	0
18	17-11/001	Herodion # 1	8/20/2005	7	0	800	300	0	0
19	17-11/002	Herodion # 2	8/20/2005	14	0	1950	400	0	0
20	17-11/003	Herodion # 3	8/20/2005	23	0	3900	10	0	0
21	16-11/006	Herodion # 4	8/20/2005	0	0	80	20	0	0
22	16-11/001A	Beit Fajjar	8/20/2005	9	0	1600	240	6	0
23	16-11/008	Safi 1	8/20/2005	0	0	550	0	0	0
24	16-11/001	PWA 1	8/20/2005	0	0	10	0	0	0
25	16-11/11	PWA 11	8/20/2005	1	0	10	0	0	0
26	16-11/004	Arroub Nursery	8/20/2005	3	0	130	NF	NF	NF

27	17-11/002	Herodion # 2	11/12/2005	0	0	130	0	0	0
28	17-11/003	Herodion # 3	11/12/2005	32	0	380	0	0	0
29	16-11/006	Herodion # 4	11/12/2005	120	0	40	0	0	0
30	16-11/001A	Beit Fajjar	11/12/2005	48	0	620	10	0	0
31	16-11/11	PWA 11	11/12/2005	8	0	30	0	0	0
32	16-11/004	Arroub Nursery	11/12/2005	0	0	40	NF	NF	NF
33	17-11/001	Herodion # 1	1/21/2006	0	0	460	20	0	0
34	17-11/002	Herodion # 2	1/21/2006	0	0	280	0	0	0
35	17-11/003	Herodion # 3	1/21/2006	7	0	400	10	0	0
36	16-11/006	Herodion # 4	1/21/2006	0	0	240	0	0	0
37	16-11/001A	Beit Fajjar	1/21/2006	8	0	490	10	1	0
38	16-11/008	Safi 1	1/21/2006	45	0	800	40	0	0
39	16-11/001	PWA 1	1/21/2006	0	0	320	10	0	0
40	16-11/11	PWA 11	1/21/2006	0	0	160	0	0	0
41	16-11/004	Arroub Nursery	1/21/2006	30	0	750	NF	NF	NF
	WHO Guidelines			**0/100ml**	**0/100ml**	**≤500CFU/ ml**	**≤500CFU/ ml**	**0/100ml**	**0/100ml**

Appendix III: Differentiation of Total coliform into *Enterobacter, Klebsiella, Citrobacter and Escherichia coli* in groundwater well and spring samples in the period between May 2005 and January 2006.

Lab No.	Site Name	Location	Sampling date M/D/Y	Total coliform CFU/1 ml	*Enterobacter spp*	*Citrobacter spp*	*Klebsiella spp*	*E. coli*
1	Beit Fajjar	Bethlehem	5/2/2005	121	+	+	-	+
2	Safi 1	Bethlehem	5/2/2005	6	+	-	-	-
3	PWA 11	Bethlehem	5/2/2005	1	+	+	-	+
4	Sa'ir	Hebron	5/2/2005	TMTC	-	+	+	+
5	Al-Therweh	Halhoul	5/2/2005	108	+	+	-	+
6	Ein Al-Bas (west)	Al-Arroub	5/2/2005	15	-	+	-	+
7	Ein Al-Bas (east)	Al-Arroub	5/2/2005	TMTC	+	+	+	+
8	Arroub-Nursery	Al-Arroub	5/2/2005	8	-	+	-	+
9	Herodion # 1	Bethlehem	8/20/2005	7	+	-	-	-
10	Herodion # 2	Bethlehem	8/20/2005	14	+	-	-	-
11	Herodion # 3	Bethlehem	8/20/2005	23	+	+	-	-
12	Beit Fajjar	Bethlehem	8/20/2005	9	+	-	-	-
13	PWA 11	Bethlehem	8/20/2005	1	+	-	+	+
14	Sa'ir	Hebron	8/20/2005	TMTC	-	+	+	+

15	Al-Therweh	Halhoul	8/20/2005	370	-	-	+	+
16	Ein Al-Bas (west)	Al-Arroub	8/20/2005	122	+	-	-	-
17	Ein Al-Bas (east)	Al-Arroub	8/20/2005	TMTC	+	+	+	+
18	Ein Al-Warasnah	Al-Arroub	8/20/2005	TMTC	-	+	-	+
19	Ein Al-Dilbi	Halhoul	8/20/2005	340	-	-	+	+
20	Ein Ayoub	Halhoul	8/20/2005	TMTC	+	+	-	+
21	Ein Al-Shinnar	Halhoul	8/20/2005	TMTC	+	-	-	-
22	Arroub-Nursery	Al-Arroub	8/20/2005	3	+	-	-	-
23	Ein Almarj	Al-Arroub	8/20/2005	TMTC	+	-	-	-
24	Ein Al-Arroub	Al-Arroub	8/20/2005	TMTC	+	+	+	+
25	Herodion # 3	Bethlehem	11/12/2005	32	+	-	+	-
26	Herodion # 4	Bethlehem	11/12/2005	120	+	-	-	-
27	Beit Fajjar	Bethlehem	11/12/2005	48	+	-	+	+
28	PWA 11	Bethlehem	11/12/2005	8	+	-	-	-
29	Sa'ir	Hebron	11/12/2005	TMTC	-	-	+	+
30	Al-Therweh	Halhoul	11/12/2005	TMTC	-	+	-	+
31	Ein Al-Bas (east)	Al-Arroub	11/12/2005	TMTC	-	+	-	+
32	Ein Al-Warasnah	Al-Arroub	11/12/2005	TMTC	+	-	-	+
33	Ein Mesleh	Halhoul	11/12/2005	15	+	+	-	-
34	Ein Al-Dilbi	Halhoul	11/12/2005	TMTC	-	+	+	+

35	Ein Ayoub	Halhoul	11/12/2005	TMTC	-	+	+	+
36	Ein Al-Shinnar	Halhoul	11/12/2005	TMTC	-	-	+	-
37	Ein Almarj	Al-Arroub	11/12/2005	TMTC	-	-	+	
38	Ein Al-Arroub	Al-Arroub	11/12/2005	TMTC	-	+	-	+
39	Beit Fajjar	Bethlehem	1/21/2006	8	+	-	-	-
40	Safi 1	Bethlehem	1/21/2006	45	+	-	-	-
41	Al-Therweh	Halhoul	1/21/2006	350	-	-	+	+
42	Ein Al-Bas (west)	Al-Arroub	1/21/2006	26	+	-	+	-
43	Ein Al-Bas (east)	Al-Arroub	1/21/2006	20	+	-	+	+
44	Ein Al-Warasnah	Al-Arroub	1/21/2006	80	-	-	+	+
45	Ein Mesleh	Halhoul	1/21/2006	32	+	-	-	-
46	Ein Ayoub	Halhoul	1/21/2006	260	+	-	+	+
47	Ein Al-Shinnar	Halhoul	1/21/2006	50	+	-	+	+
48	Arroub-Nursery	Al-Arroub	1/21/2006	30	+	-	-	-
49	Ein Almarj	Al-Arroub	1/21/2006	250	-	-	+	+
50	Ein Al-Arroub	Al-Arroub	1/21/2006	60	+	+	-	+

Appendix VI: Nitrate concentration in groundwater well and spring samples of Wadi Al-Arroub drainage basin.

Lab No.	Site Name	Location	Sampling Date M/D/Y	NO_3 mg/l	Sampling Date	NO_3 mg/l
1	Herodion # 1	Bethlehem	21/1/2006	44		
2	Herodion # 2	Bethlehem	21/1/2006	38	11/12/2005	17
3	Herodion # 3	Bethlehem	21/1/2006	25	11/12/2005	16
4	Herodion # 4	Bethlehem	21/1/2006	22	11/12/2005	20
5	Beit Fajjar	Bethlehem	21/1/2006	46	11/12/2005	36
6	Safi 1	Bethlehem	21/1/2006	48		
7	PWA 1	Bethlehem	21/1/2006	10		
8	PWA 11	Bethlehem	21/1/2006	12	11/12/2005	32
9	Sa"ir	Hebron	21/1/2006		11/12/2005	144
10	Al-Therweh	Halhoul	21/1/2006	66	11/12/2005	161
11	Ein Al-Bas (west)	Al-Arroub	21/1/2006	30	11/12/2005	41
12	Ein Al-Bas (east)	Al-Arroub	21/1/2006	56	11/12/2005	31
13	Ein Al-Warasnah	Al-Arroub	21/1/2006	65	11/12/2005	62
14	Ein Mesleh	Halhoul	21/1/2006	60	11/12/2005	70
15	Ein Al-Dilbi	Halhoul	21/1/2006		11/12/2005	228
16	Ein Ayoub	Halhoul	21/1/2006	64	11/12/2005	470
17	Ein Al-Shinnar	Halhoul	21/1/2006	41	11/12/2005	31
18	Arroub-Nursery	Al-Arroub	21/1/2006	53	11/12/2005	41
19	Ein Almarj	Al-Arroub	21/1/2006	66	11/12/2005	77
20	Ein Al-Arroub	Al-Arroub	21/1/2006	65	11/12/2005	82
	WHO Acceptable level		**NO_3 < 45 mg/l**		**NO_3 < 45 mg/l**	

Appendix V: The Springs of the Wadi Al-Arroub drainage basin.

No	Spring	Coordinates (PG)		Elevation (masl)	Well depth (mbgl)
		North	East		
1	Ein Almarj	114065	163584	812	7.5
2	Ein Al-Arroub	113926	163705	814	175
3	Ein Al-Bas (east)	113575	163143	856	4.5
4	Ein Al-Bas (west)	113664	163049	855	3.2
5	Ein Al-Dilbi	113088	163921	819	1.4
6	Ein Al-Shinnar	111256	161217	892	2.3
7	Ein Al-Therweh	110498	159824	934	0
8	Ein Al-Warasnah	113899	163967	797	14
9	Ein Ayoub	113288	163193	825	1.3
10	Ein Mesleh	111063	160290	907	0
11	Ein Sa"ir	110217	163847	863	4