Integrated Concepts for Reuse of Upgraded Wastewater

AQUAREC

EVK1-CT-2002-00130
Work package 2
Deliverable D15

Guideline for quality standards for water reuse in Europe

May 2006
Edited by Miquel Salgot and Esther Huertas, University of Barcelona

WP partners

M. Salgot & E. Huertas, Soil Science Laboratory. Faculty of Pharmacy. University of Barcelona. Avda. Joan XXIII, s/n. 08028, Barcelona, Spain.

J. Hollender, W. Dott & S. Weber. Inst. Hygiene & Environmental Medicine, RWTH. Pauwelsstraβe 30, D-52076 Aachen, Germany.

A. Schaefer & S. Khan, Environmental Engineering, University of Wollongong, NSW 2522 AUSTRALIA

B. Bis. Faculty of Biology & Environmental Protection. University of Lodz. Ul. Pilarskiego, 14. 90-231 Lodz, Poland.

A. Aharoni & H. Chikurel, Mekorot Ltd - 9 Lincoln St. PO Box 20128 Tel-Aviv 61201, Israel.

R. Messalem. The Institutes for Applied Research, Ben-Gurion University of the Negev, Beer-Sheva 84105, Israel.

CONTENTS

1. EXECUTIVE SUMMARY

Reclamation and reuse hazards are usually defined accordingly to standards issued or recommended by local authorities or international agencies. When trying to study the rationale of such approach, we can find several inconsistencies, namely the adequacy of control parameters, a certain lack of definition of the appropriate sampling points, the number of samples and analysis necessary, and the cost of the analytical work.

In any case, if standards are to be met, it appears that planning steps, economic calculations, and social tasks are to be performed for successful reclamation and reuse practices. Then, it seems obvious that scenarios must be built for the correct comparison of the different possible alternatives, including all the data needed to reach a correct decision.

From the zero scenario (no reuse) to the theoretically more expensive one (reclamation using Reverse Osmosis), adequate tools are to be employed in order to help stakeholders to decide the best option for the increase of available water resources. Among the tools, decision support systems are the most useful for gaining good knowledge, but other studies must be previously undertaken to adequately know the necessary technologies and schemes. As health hazards are one of the main constraints for reuse, it appears that risk assessment, based on hazard calculations, is basic for several definitions in reclamation and reuse projects.

There are different possible applications for reclaimed water reuse. Seven microbial water qualities, according to the Spanish recommended regulation, and four chemical water qualities are required by Reclaimed water irrigation of crops consumed raw or cooked.

Risk evaluation in relation with wastewater reclamation and reuse became at the end of the last century one of the more interesting tools to establish relationships between reclaimed water quality and the health hazards associated to the practice of reuse.

In this sense, there are several steps to develop in any facility, initially devoted to establish the quality of reclaimed water. Then it is to consider that the application of the risk related tools starts with risk assessment, is followed by risk calculation, and ends with the management and communication.

The basis of it all is the availability of reliable data on the analytical parameters which exert a noticeable impact on the whole risk. Those data need to be elaborated in such a way that allows ranking the parameters and giving a figure, a magnitude, of the risk associated to a unique chemical or microorganism.

Apart from it, and making the analysis more difficult, there are antagonisms and agonisms (synergies) which can exert in the real world a final effect on the target: the human being, or alternately the animals or the whole ecosystem.

The number of microorganisms is relatively high, but it is nothing compared to the huge amount of chemicals, mainly xenobiotics, which have an effect on the target.

At this level, there are several sciences implied in the procedure: toxicology, ecotoxicology, physico-chemistry, microbiology, parasitology… All of them can offer tools to calculate the risks derived from

any practice where the organisms and chemicals enter into contact with nature, and more specifically men.

The studies to be undertaken are varied and derive from the mentioned sciences, plus the studies on the degradation of the component in the environment. Then, the solubility, the decay rate, the capability to be degraded by biological or physico-chemical means, and the calculation of several constants appear to be paramount.

The final results are complicated formulae, which can give us an idea of the difficulties to reach an objective determination of the risk associated to the practice. It is to indicate that the figures are always approximate, because the ecosystems, and especially the biocenosis in the ecosystem, never act in the same way. The main cause is the variability inherent to any system in nature. Bioavailability, modifications, and any circumstance differ between ecosystems and in the same ecosystem also vary depending on the existing conditions of temperature, exchanges, and the like.

2. INTRODUCTION

Throughout the last decades, municipal wastewater reuse has emerged as an important and viable means of supplementing dwindling water supplies in a large number of regions throughout the world. In many instances, reuse is also promoted as a means of limiting wastewater discharges to aquatic environments.
Reclamation and reuse hazards are usually defined according to standards issued or recommended by local authorities or international agencies. When trying to study the rationale of such approaches, we can find several inconsistencies, namely the adequacy of control parameters, a certain lack of definition of the appropriate sampling points, the number of samples and analysis necessary, and the cost of the analytical work.
In any case, if standards are to be met, it appears that planning steps, economic calculations, and social tasks are to be performed for successful reclamation and reuse practices. Then, it seems obvious that scenarios must be built for the correct comparison of the different possible alternatives, including all the data needed to reach a correct decision.
From the zero scenario (no reuse) to the theoretically more expensive one (reclamation using Reverse Osmosis), adequate tools are to be employed in order to help stakeholders to decide the best option for the increase of available water resources. Among the tools, decision support systems are the most useful for gaining good knowledge, but other studies must be previously undertaken to adequately know the necessary technologies and schemes. As health hazards are one of the main constraints for reuse, it appears that risk assessment, based on hazard calculations, is a basis for several definitions in reclamation and reuse projects.
Through integrated water resources planning, the use of Reclaimed water may provide sufficient flexibility to allow water authorities, as well as individual users, to respond to short-terms needs and to increase sustainable, long-term water supply reliability, without constructing additional storage or conveyance at substantial economic and environmental expenditures. Significant progress has been made in developing sound technical approaches to producing reliable sources of quality water by wastewater reclamation processes. As a result, continued research, progress in innovative conceptual and methodological approaches and testing efforts will result in further progress in the advanced development of water reuse applications – directly related to cost-effective strategies. Some key topics involved are: (1) assessment of human health associated with trace organic substances, (2) environmental risk assessment, (3) improvements of monitoring approaches to evaluate chemical and microbiological water and related materials quality. In this study part (2) will be developed.
We define risk as the probability of injury, disease or death under specific circumstances. In quantitative terms, risk is expressed in values ranging from zero (representing the certainty that harm will not occur) to one (representing the certainty that harm will occur). When dealing with the risk associated to reclaimed water reuse, we must take into consideration not only the risk associated to the human health but the one that is related to the environment.

2.1. Water reuse benefits

The risk and disadvantages of water reuse against the background of water reuse advantages are compiled in Table 1.

Table 1 Advantages, disadvantages and possible risks of wastewater reuse [modified from 1]

Advantages	Disadvantages	Risks
ECONOMY • Improvement of the economic efficiency of investments in wastewater disposal, irrigation and other uses • Conservation of freshwater resources • Recharge of aquifers through infiltration water (natural treatment) • Increase and ensure resources availability	Wastewater is normally produced continuously throughout the year, whereas wastewater reuse for irrigation is mostly limited to the growing season: additional disposal or storage facilities are needed	Potential harm to natural waters due to pathogens, heavy metals, nitrate, organic matter and other chemicals which will make water not useful for the initially existing purposes
AGRICULTURE • Use of the nutrients of the wastewater (e.g. nitrogen and phosphorus) • Reduction of the use of synthetic fertilizer • Improvement of soil properties soil fertility; higher yields	Some substances that can be present in wastewater in such concentrations that they are toxic for plants	Potential harm to human health by spreading pathogenic germs and chemicals
TREATMENT/RECLAMATION • Reduction of treatment costs: Soil treatment of the pre-treated wastewater via irrigation (no tertiary treatment necessary, highly dependent on the source of wastewater)	Increase costs of treatment (especially if sensible uses are implemented)	Environmental impact of treatment by-products and sludge
ENVIRONMENT • Beneficial influence of a small natural water cycle • Reduction of environmental impacts (e.g. eutrophication and minimum discharge requirements)	Some substances that can be present in wastewater in such concentrations that they lead to environmental damage	Potential harm to environment matrices by spreading pathogenic germs and chemicals

Water reuse include benefits, which can be classified as:

a) global:

- reduced diversion costs, and
- value of a secure "drought proof" supply of reclaimed water,

b) agricultural:

- increased farm production, and
- value of reclaimed water nutrients = savings in fertilizer applications;

c) for urban water supply:

- savings in the capital cost of diversion structures, drought storage, transfer systems and water treatment, and
- savings in operation and maintenance costs including pumping energy and treatment chemicals;

d) related to urban wastewater:

- savings in discharge pump stations and pipelines, and
- savings in treatment and nutrient removal costs required for discharge to sensitive waters;

e) for environmental water quality:

- reduction in freshwater diversions

- more river flow for downstream users
- better downstream water quality
- reduction in pollutant discharges
- better downstream water quality
- reduced environmental impact and improved river aesthetics
- reduced impacts on fisheries and aquatic life
- improved public health for downstream users
- lower water treatment costs for downstream users
- improved recreational values of waterways.
- Nevertheless, several inconveniences can be described, as:
- river flow reduction
- need for additional structures
- health and environment hazards (include toxicity)
- additional treatment costs

It is to consider that in rivers from arid and semiarid areas the existing flow can be mainly provided by wastewater (reclaimed or not) disposed of into the river. Thus, reuse can reduce the flow, then compromising existing previous uses.

2.2. Reclamation Treatments

Different wastewater reclamation treatments, depending on the origin and reuse application, are available. The most important for irrigation purposes are secondary and advanced tertiary treatment. Advanced tertiary treatment includes ammonia removal, secondary water plus microfiltration and reverse osmosis, softening treatment, reverse osmosis with adequate pretreatment, stabilization ponds, UV irradiation and disinfection. It is to consider that any additional treatment has a cost, which must be beared by the end-users or by the water administration. There is then the need to optimizing costs, trying to implement the best available technology.

Usually, the treatments include further elimination of organic matter and suspended solids, although in some cases this is not necessary (when using several extensive technologies). After this kind of pretreatment, the disinfection step follows. There are several potential new hazards associated to this reclamation treatment, namely the appearance of disinfection by-products (DBP), especially when using chemicals for disinfection or reliability problems.

2.3. Uses of Reclaimed Water

Reclaimed water is adequate for a large variety of uses, being the main one:

- agricultural irrigation
- non-agricultural irrigation
- urban domestic
- urban not domestic
- livestock
- recreational uses (fountains)
- bathing water
- water-related sports, leisure activities
- industry
- groundwater recharge
- stream and water bodies recharge
- thermoelectric power use
- aquaculture
- drinking water production

The different reclamation purposes need specific microbial and chemical requirements that are described below in more details and are related to rules and regulations, including standards. A comparison of international standards might help to develop guidelines for the reference area within each particular project.

Agricultural Irrigation

The main possibilities of reclaimed water use are related to irrigation: agricultural, urban, private and public gardens. Nevertheless, the most important user of reclaimed water for irrigation – and generally of all reclaimed water uses – has been the agriculture. As a consequence, agricultural irrigation with reclaimed water has been fully studied and has been the basis for issuing standards and all types of regulations

For agricultural use, the water quality should be sufficient to protect human health when consuming food produced from reclaimed water irrigation. Furthermore, the soil environment specifically, and all related environments generally, should also be protected from contamination by reclaimed water irrigation. In terms of agricultural concerns, the irrigation water must mainly require low salinity. In addition, bioaccumulation of organic and inorganic contaminants in plants has to be considered for human health protection. Growth inhibition by several components of reclaimed water, such as boron, should be taken into account, and also toxic effects from reclaimed water constituents.

In agricultural reuse the health protection measures should be considered and recommended in the main strategic areas (management practices to interrupt the flow of pathogens or the presence of harmful chemicals), selected to suit local circumstances, including:

crop selection

reclaimed water application measures/control

human exposure control.

For which respects to the main irrigation-related parameters that could affect the plants, there is to mention Chlorides, Electrical Conductivity, Sodium, Potassium, and Boron. Some components of the irrigation water (reclaimed) can be recycled/treated by the soil-plant system: e.g. nutrients, or several contaminants difficult to be degraded. Soil salinity could be overwhelmed if good quality water is not available; e.g. by leaching practices. In the Mediterranean, medium and sometimes high salinity water can be used for years without further damage if the system management is correct.

Contamination of crops by pathogens can be:

1. Exterior: deposition of organisms which have been transported through irrigation water. In this way, crops can be contaminated:
   a) directly, through the contact with irrigation water,
   b) indirectly, through the contact of leaves with soil or contaminated material,
   c) indirectly, through deposition of aerosols containing viable pathogens,
2. Interior: organisms enter the plant through injuries and continuity solutions.

The irrigation method conditions crop contamination: irrigation by periodic cycles of inundation/drying presents an advantage in front of irrigation by sprinkling. Sprinkler irrigation is a watering method which needs to be performed frequently. It is to note that the prolongation of the time elapsed between subsequent irrigations favours the action of environmental conditions (light and desiccation) against

pathogens. When the periodicity is shorter, the environmental inactivation is more difficult; periodic desiccation is substituted by a wet environment that favours the survival of pathogens.

Drip irrigation allows the use of reclaimed water without contaminating most of the crops. This irrigation method has negative (no contamination) results for bacterial, viral and parasitic contamination of not underground crops.

Bacteria survival in crops is influenced by the kind of crop. Several plant structures (like lettuce) give protection against the action of ultraviolet rays of solar light. The survival is bigger in dense and leafy plants. Other factors which influence the survival in the crop are: temperature, humidity and rain, that drags organisms to the soil.

The risk of goods of animal origin as transmitters of pathogenic agents is often neglected or can be underestimated. Animals can accumulate in their bodies many pathogenic agents (e.g. *Taenia* species) or toxic substances that they consume with fodder irrigated with raw wastewater and which are transmitted later to humans.
The infection hazard for workers using reclaimed water for irrigation purposes without sufficient self protection should be avoided.

Groundwater recharge

To recharge groundwater is a powerful tool for recovering water levels of streams, aquifers or other waterbodies.
Groundwater recharge can be performed in two ways, through soil and subsoil passage or by direct entrance (injection) into the aquifer. Water recovered from aquifers directly recharged with reclaimed water should not be used for drinking water production. For human health protection and maximum chemical clarity preservation, drinking water supplementation should only be permitted after advanced tertiary treatment (membrane processes, reverse osmosis) and other barriers such as percolation through the soil.
The purpose of indirect aquifer recharge using reclaimed water may be a) to establish saltwater intrusion barriers in coastal aquifers, b) to provide further treatment for future reuse, c) to augment potable or nonpotable aquifers, d) to provide storage of reclaimed water for subsequent retrieval and reuse, and e) to control or prevent ground subsidence.
The soil acts as a barrier retaining contaminants, but it is necessary to have a soil isotropic, without great discontinuities. For this reason, the reclaimed water applied to such systems could have less quality than the water directly injected into the aquifer. There is a classical system which uses the favourable characteristics of soil, subsoil and aquifer for further treating applied water. It is the SAT (Soil Aquifer Treatment) developed by Bouwer [2]. The Dan Region project in Israel is a big scale application of the SAT principle.
Obviously, the water quality depends also on the use that water will have once recovered from the aquifer. If water has to be used for drinking purposes the quality must be high. On the contrary, if recharged water is used to fight against seawater intrusion, water quality could be of not so high quality.
Usually, the hydraulic retention time of reclaimed water in the aquifer must be long, exceeding usually one month and reaching easily several years. The quality of such water is suitable for unrestricted irrigation purposes.

Residential uses

For private uses (urban domestic) like house garden irrigation, toilet flushing, home air conditioning and car washing, with delivered reclaimed water to households, a satisfactory water quality for all intended uses should be ensured. Otherwise, several pipelines with variable water qualities would compound the hazards associated with improper handling by the owners and plumbing professionals. Additional attention has to be paid to avoid cross connections of potable and non potable water, and also concerning children playing in the nearby ground of private reclaimed water irrigation. The risk of infection by pollutant inhalation of reclaimed water aerosols, for example in home air conditioning, should be limited.
To implement reclaimed water reuse for private use in different scale only one additional pipeline should be installed. The water distributed by this pipeline should meet the highest water quality that is required for private uses.

Urban and recreational uses

Urban uses (urban not domestic) include street cleaning, fire fighting, ornamental impoundments and decorative fountains (recreational uses) as well as impoundments, water bodies and streams for recreational use, and sewerage management. Reclaimed water can also be used for irrigation purposes (public parks and gardens, cementeries).
The most important issues for reclaimed water used for recreation of impoundments, water bodies and streams should be the protection of the aquatic environment. The water quality should ensure no adverse effects on the aquatic organisms and ecosystems. Therefore fish toxic compounds have to be limited as well as endocrine disrupters. Contact, direct and indirect, with people needs to be fully controlled.

Aquaculture

Reclaimed water used for aquaculture should not harm or affect the aquatic environment when disposed. In addition, the human as consumer of the produced fish must be protected from adverse effects. Several substances are fish-toxic or accumulate in fish's lipid tissue. Therefore the water quality needs special limits concerning the bioaccumulation, toxic substances and endocrine disruptors.
The risk of goods of fish origin as transmitters of pathogenic agents is often neglected or can be underestimated. Fish, and especially shellfish, can accumulate in their bodies many pathogenic agents which are transmitted later.

Industrial cooling water

Among the variety of uses that reclaimed water may have in the industry (process water, heavy construction,boiler feed, etc.), industrial cooling water is the most popular.
The industrial cooling water is generally circulated in a closed system and does not have direct contact with humans or the environment. Therefore the issues of concern should prevent corrosion and calcareous crusts to protect pipelines and steel bins. For example high amounts of dissolved organic carbon (DOC > 5 mg/L) inhibit the corrosion protective coating of copper [3, 4]. The precipitation of mineral salts and silica during the heating process has also to be prevented.

Drinking water production

Although prohibited in several countries, reclaimed water could be used for driking purposes. The most important experience is in Namibia [5], although in California reclaimed water is used for drinking water production through aquifer recharge.

California's draft revision of Title 22 (devoted to reuse) remains very specific as to how reclaimed water quality objectives are to be achieved. It requires several barriers and controls, and a whole system of quality assurance is established.
There are several pilot facilities dealing with treatment and quality, but so far there are no additional experiences.

Bathing water

Reclaimed water used for bathing water purposes - swimming-pools, water-related sports or e.g. leisure activities - needs special quality requirements concerning protection of human health, i.e. disinfection. The quality requirements are set by the European Bathing Water Quality Directive 2006/7/EC [6].

Thermoelectric power use

Reclaimed water can be a useful source for power generation. This water doesn't need quality rules, but resources management are essential.

2.4. Recommended Guidelines for Wastewater Reuse

Several guidelines concerning reclaimed water reuse have been adopted in the last years, either for drinking water production or agricultural and other uses.

California title 22

In California, with the adoption of the Porter-Cologne Act in 1969, the Legislature declared its intent to "undertake all possible steps to encourage development of water recycling facilities". The California Water Code articulates a clearly-defined strategy favouring the beneficial reuse of water to the maximum extent practical. California Department of Health Services (DHS) establishes water quality standards and treatment reliability criteria for water recycling under Title 22, Chapter 4, of the California Code of Regulations. Requirements for a use of recycled water not addressed by the uniform statewide criteria are established by DHS on a case-by-case basis. The table called "Recycled Water Uses Allowed in California" illustrates the wide variety of successful reuse applications and the level of treatment required. In addition to establishing recycled water quality standards, Title 22 specifies the reliability and redundancy for each recycled water treatment and use operation. Treatment plant design must allow for efficiency and convenience in operation and maintenance and provide the highest possible degree of treatment under varying circumstances. For recycled water piping, DHS has requirements for preventing backflow of recycled water into the public water system and for avoiding cross-connection between the recycled and potable water systems.

World health organization

To protect public health and facilitate the rational use of wastewater and excreta in agriculture, in 1973, WHO developed "Reuse of Effluents: Methods of Wastewater Treatment and Public Health Safeguards." After a thorough review of epidemiological studies and other information, the guidelines were updated in 1989, discussing two approaches for developing guidelines on wastewater reuse for irrigation purposes: the setting of numerical standards based on technical decisions and the epidemiological approach. These

guidelines have been very influential and many countries have adopted or adapted them for their wastewater and excreta-use practices.
WHO is promoting a new perspective, using the DALYs instead of the numericao standards. This new way seems promising, but needs further understanding, devoted to research and a good communication policy.

Approaches in the mediterranean region

A very limited number of European countries have guidelines or regulations on wastewater reclamation and reuse. Usually, they are the southernmost countries that have regulations. Most of the northern states do not have any specific legislation on wastewater reuse because they do not have the necessity and their rivers have a sufficient dilution factor.
The only reference to wastewater reuse at European level is article 12 of the European Wastewater Directive 91/271/CEE: "Treated wastewater shall be reused whenever appropriate". In order to make this statement reality, common definitions of what is "appropriate" are needed.
Then the EU countries developed by themselves, if needed, their own regulations. A discussion on part of those items is following.
The 12th of June 2003, a Decree (number 185) was approved by the Italian Environmental Ministry, where minimum quality criteria of treated wastewater was established for its reuse. The different recommendations from Italian regions – not more enforceable - were a combination between the WHO and the California guidelines. The main actual problem is that the requirements are even more restrictive than the ones for tap water. France enacted a national code of practice, under the form of recommendations from the Conseil Superieur d'Hygiene Publique de France (CSHPF, 1991). These recommendations use the WHO guidelines as a basis, but complement them with strict rules of application. CSHPF (1991) calls for a strict observation of these restrictions to ensure the best possible protection of the public health. There is a revision theoretically under way, but did not reach an end up to now. In Greece a preliminary study for establishing quality criteria is in progress, although important projects are being carried out. Portuguese legislation allows reuse of correctly reclaimed water reuse in a large number of crops by the Law 74/90 (art. 32) since 1990. However, microbiological criteria and criteria related to the irrigation systems are still needed. In Cyprus, provisional standards related to the use of treated wastewater effluent for irrigation purposes are under establishment. Those standards are stricter than the WHO ones with the aim to cover the specific conditions of the island. Additionally, there is a practical code for irrigation with reclaimed water.

In Spain, the existing laws only state that:

(a) Every potential water user should ask for administrative concession of a given amount of water, even reclaimed water

(b) For reclaimed water, before giving the permission, a "compulsory" report should be issued by the Health Authorities. The conclusions given in this report are to be forcefully implemented in order to obtain the concession.

(c) The Government must develop a Reclamation and Reuse Regulation (not yet appeared, but under preparation).

Because of the lack of any additional indication, the Health Authorities of some of the Spanish Autonomous Regions were forced to develop their own guidelines in order to face the demands for

approval. Three of such guidelines have been published up to now. It is to indicate that the Canary Islands have their own water law and regulations. In these Islands, the Government does not own water resources, as it is the case in the rest of Spain [7].

Water stress in Southern Mediterranean countries is usually more pronounced than in Northern Mediterranean countries. There are several causes for it, and nowadays water demands are continuously increasing because of the economic development, tourism rise and demographic growth. Moreover, in these countries water resources are limited because of the droughts they have been suffering for decades. As a result it is necessary to exploit all the water resources, including wastewater.

Outside of Europe, Morocco does not have yet any specific regulation with regard to wastewater reuse. Reference is usually made in the projects to WHO recommendations. Agricultural wastewater reuse in Tunisia is regulated by the 1975 Water Law, and by a 1989 Decree. This Water Law provides a legal framework for reclaimed water use. This code prohibits the use of raw wastewater in agriculture, and irrigation with reclaimed water of any vegetable to be eaten raw. This law stipulates that wastewater for irrigation must have a quality which does not allow the transmission of diseases. The 1989 Decree specifically regulates reuse of wastewater in agriculture.

In Jordan the standards for wastewater treatment and reuse were introduced in 1982 by a Martial Law. In 1989 a more liberal version of the Martial Law was enacted. In Lebanon wastewater treatment and reuse are regulated by a legislation of 1930.

In Israel about 76% of the wastewater collected by municipal sewers is used for irrigation either by tertiary treatment (filtration and disinfection, or polishing reservoirs and disinfection) or by Soil Aquifer Treatment (SAT). In 1978 Israel published a sanitary regulation based on the California standards. In 1999 new regulations based on multiple barrier principle (Halperin committee regulations) were issued. A new recommended regulation (Inbar committee regulations) was published by Israel administration in 2003, which present more quality parameters to control in comparison with 1978 and 1999 Regulations [8]. The Inbar committee was composed of representatives from all government ministries in order to evaluate thresholds values for parameters needed for unrestricted irrigation.

In Israel, local, regional and national authorities must approve the use of reclaimed water. The authorities demand the farmers to get an “irrigation permit” based on the effluent quality, location of their irrigated land and the type of crop, in order to ensure that there is no danger to human health or water resources.

Guidelines in preparation: USEPA and Australia

Recently the US-EPA published extensive Guidelines for Water Reuse (2004) which summarize water reuse supporting information and cover water reclamation for nonpotable urban, industrial and agricultural reuse, as well as augmentation of potable water supplies through indirect reuse. Direct potable reuse is also covered, although only briefly, since it is not practiced in the U.S. [9]. This document is intended to be solely informational and does not impose legally-binding requirements on EPA, states, local or tribal governments. In states where standards do not exist, are being revised or expanded, the Guidelines can assist in developing reuse programs and appropriate regulations. In the U.S. water reclamation and reuse standards are the responsibility of state agencies.

The Australian guideline for water reuse is currently in preparation. The principles have been agreed and were adopted in the Australian Drinking Water Guidelines (ADWG, National Water Quality Management Strategy 2004). The principles include:

- Protection of water sources and treatment are of paramount importance and must never be compromised.
- The system must continually maintain robust multiple barriers appropriate to the level of potential contamination facing the raw water supply.
- Any sudden change in water quality, flow or environmental conditions should arouse suspicion that drinking water might become contaminated.
- System operators must be able to respond quickly and effectively to adverse monitoring signals
- System operators must be dedicated to providing customers with safe water and never ignore a consumer complaint about water quality.
- Ensuring drinking water safety requires application of a considered risk management approach.
- The framework incorporates principles of established systems such as HACCP, ISO 9001 (2000) and AS/NZS 4360 (1999). ADWG also extends beyond single water utilities and takes into account the need for multi-agency involvement where responsibilities for different components of the entire water supply system from catchment/aquifer to tap are dispersed between organizations, such as water supply utilities, catchment management boards, water resources, environment protection, agriculture and planning departments, local government, and community-based interest groups. The new code foster innovation to enable improved practices and use of resources while managing risks within a Hazard Analysis and Critical Control Points strategy to meet the first objective.

In conclusion, standards have reached a point were new developments could not be expected, and new working directions are needed. The WHO seems to have started such a new way of working, as the implementation on risk-related considerations is being suggested (Kamizoulis, 2005 [10] and 2006), as is detailed later on.

3. RECLAIMED WATER QUALITY REQUIREMENTS

The risks concerning Reclaimed water reuse can be classified in microbiological and chemical as well as ecological risks. The microbiological and chemical parameters of interest are described in more detail as follows.

3.1. Microbiological parameters

The increase of health hazards and concerns in relation with wastewater, the growing number of wastewater treatment facilities, and the need of obtaining additional water resources through wastewater reuse, forced the necessity of using more precise and sophisticated biological control tools for wastewater recycling. Due to the costs and complexity of analysing actual pathogens it should be considered [11] that wastewater professionals and regulators have relied for decades on traditional faecal indicators to predict potentially high pathogen levels.

The scientific community have been always aware that detection and quantification of *E. coli* is not enough to entirely define the quality of a certain wastewater treated, reclaimed, or discharged of in the environment. Some pathogens are more resistant than usual indicators to conventional wastewater treatments (including chlorination) and its sources are not warm blooded animal faeces. Therefore *E. coli* is an insufficient tool to reflect the microbiological quality changes due to wastewater treatment processes, conventional or advanced, extensive or intensive. In addition it does not fully permit the control of wastewater disinfection.

Otherwise, the long period of time needed to produce results is a negative feature in the analytical work, but not only related to *E. coli* determination. However, new molecular biological methods are under development, which enable a faster determination of specific microorganisms [12].

It is necessary to define more suitable indicators in order to establish the entire biological quality of different types of wastewater (Table 2) and do it in delays shorter than the present ones. For example *Enterococcus faecalis* and even spores of *Clostridium perfringens* are known to be more resistant against disinfection than *E. coli* and therefore may be used for reclaimed water control. Otherwise, the current analysis using bacterial indicators detects only the quality of a single wastewater sample and not the constant quality of such water during a given time.

Campos (1999) stated also the difficulty of defining the microbiological quality of the media that receive Reclaimed water, particularly the agricultural systems, where reclaimed water is used for irrigation [13]. Plants, soils, groundwater, runoff and atmosphere can and must be controlled separately from the Reclaimed water. Campos also indicates the need to establish and realise joint studies of all media implied in wastewater reuse systems. An additional difficulty is the lack of reference figures for pathogen organisms contents in other media than water.

Until now, indicators and sometimes index-based standards have been used to define a suitable reclaimed water quality. Nevertheless, it must be stated that the health risk related to reuse is defined by several aspects: the microbial agent, the human host, and the environment in which the infection process is mediated [14]. As it is the interaction of these components which produces human disease, risk is dependent on defining the exposed population, the microbial characteristics, and the environmental setting in which the exposure occurs. The human health risks associated with the ingestion of waterborne pathogens could be theoretically and numerically defined by using statistic models that calculate the

probability of individual infection or disease as a result of a single exposure event. This type of calculations [15] could be a good tool when fully developed in the future.

Table 2 Types of waterborne pathogens and used indicators [modified from 16]

Types of waterborne pathogens	Indicators	Observations
Bacteria	*E. coli*, Faecal coliforms, Total coliforms, *Enterococcus faecalis*, *Staphylococcus aureus*, *Salmonella spp.*, *Clostridium perfringens*, *Pseudomonas aeruginosa*, *Legionella pneumophila*	Faecal coliforms determination is the more usual; *E. coli* determination is slowly substituting it. Other bacteria are used for bathing waters, groundwater, etc.
Viruses	Enterovirus Hepatitis A virus Bacteriophages	An accepted indicator still does not exist. Bacteriophages are being studied in this sense and somatic ones are being suggested for general use.
Helminth - Nematode	Nematode eggs (*Ascaris*, *Trichuris*, *Ancylostoma* as indicated by the W.H.O.)	Discouraging: a lot of negative results in a lot of countries. Eggs viability is not asked for.
Other helminths (i.e. *Taenia*)	Not known	In some cases important for risk related to animal health
Protozoa (include *Giardia*, *Cryptosporidium*, *Amoeba*, *Balantidium*, etc.)	Not known The presence of one of them could indicate the presence of the other	Analytical tools not well developed until now
Fungi, algal toxins	Not known	Really few cases detected

Note: When the organism is detected directly, it is known as Index organism

Otherwise, it was suggested in a WHO report [17] that the epidemiological method must be employed for determining health risks associated with reuse practices. Nevertheless, it seems not to be a good tool, as stated by Cooper and Olivieri [18]. It appears that traditional epidemiological methods are not sensitive enough to "tease out" cases that might be associated with recycled water from the background incidence of these ailments in the community.

The hazards related to microbiological quality of Reclaimed water are due to the presence of pathogen organisms, as indicated before. The hazards/risks are generated when the pathogen reaches the host, mainly through contact or ingestion. Then, there is a need to work on the pathogens presence and on the contact/ingestion mechanisms.

There are several difficulties when trying to determine the presence of pathogens, mainly the high number of different organisms present, its different infectivity and the differences of susceptibility among the populations in the several countries. The hygienic habits, strongly related to the socio-economic status, are also related to the prevalence of organisms in the whole population.

Then, a compromise should be reached and *E. coli* determination is to be maintained for several reasons (e.g. the huge amount of data existing on that organism) but alternative indicators or determinations are needed. In order to overcome the problem, there are several solutions:

- The substitution/complement of *E. coli* by other indicators or indexes.
- The implementation of other control solutions, like the HACCP (systems of Hazard Assessment and Critical Control Points) or the GRP (Good Reuse Practices).

- The implementation of indirect technologies for the determination of microorganisms, or real-time determinations.

In which respects to the mechanisms to reduce contact/ingestion, it is also useful to refer to reuse practices and education/communication policies; which are similar to the tools to reduce the hazards/risks related to chemicals.

3.2. Chemical parameters

Depending on the origin of the wastewater and on the sewage treatment process different chemical contamination will be expected. It can be inorganic (salts, nutrients, heavy metals) or organic (bulk organic matter, detergents, organic trace pollutants, etc.). The specific reuse application determines its specific quality parameters. The high number of parameters and samples to be determined may be reduced by risk assessment approaches, which are useful to identify the most critical parameters. Furthermore, to minimize analytical costs, suitable indicator substances or indicator effects should be identified and a suitable monitoring strategy is to be developed.

Inorganic compounds

Many inorganic species may be of particular concern for reuse applications involving irrigation of farmland (see Table 3). These may include salts of common ions such as sodium, potassium, calcium, chloride and bromide, as well as trace heavy metals including mercury and cadmium. Highly saline irrigation water can severely degrade soils over time. High boron concentrations (>0.4 mg/L) and sodium concentrations should be avoided as detrimental effects occur on e.g. citrus crops.

Some compounds such as heavy metals may accumulate in soils, later into plants and consequently foodstuffs. In dry climates, much of the irrigation water can evaporate and the concentration of salts in the drainage can be much higher than in the water itself, posing potential threats to groundwater quality [19]. Inorganic chemicals are among the most recalcitrant during most processes of wastewater treatment. They are often highly water-soluble and they are not susceptible to biological degradation. Similarly, advanced oxidation processes are ineffective and these small ions are among the most difficult contaminants to treat by membrane processes. For heavy metals, which have been included in some new and recommended guidelines [8, 20], multi-element or multi-component analysis determination methods like ICP-MS may simplify the required analytical effort.

Table 3 Reported occurrence of selected inorganic pollutants in water [µg/L]

Chemical pollutant	Influent STP; Raw sewage water (mean)	Effluent STP (mean)	River water; Surface water (mean)	Groundwater; Bank filtrate	Ref.
B	500 – 3000	300 - 400			[21]
Heavy metals:					
Cr	100 – 300		0.2 – 650	0.8 – 50	[22]
Pb	n.d. – 1000	1	0..2 – 1560	2 – 100	[22-23]
Hg	0.2 – 3	28			[22-23]

Organic pollutants

Organic bulk compounds, determined by COD, BOD, TOC, and indirectly by TSS and turbidity, not necessarily present a hazard in themselves but may be significant to particular uses of reclaimed water. They are mostly used as indicators of sewage treatment plant effectiveness as they can indicate how successfully the treatment process has removed organic matter from the wastewater. High turbidity can reduce the effectiveness of disinfection involving chlorine, UV light or other disinfectants. High BOD may reduce dissolved oxygen to levels that are inadequate to support aquatic organisms, thus causing their death.

A wide range of organic trace chemical contaminants persist in municipal wastewater after conventional treatment processes. A selection of such representative contaminants is given in Table 4. These include inorganic compounds, heavy metals, persistent organic pollutants, pesticides, disinfection byproducts, endocrine disruptors, pharmaceutically active compounds and complex-forming compounds. Many of these chemicals are known or suspected of deleterious implications to human health or the environment.

Much attention has been devoted to natural and synthetic steroidal hormones, which are shown to induce biological effects on some organisms at part per trillion concentrations. Some steroidal hormones are poorly removed in conventional water treatment processes [24, 25]. Other chemicals exhibiting similar effects at higher concentrations that are known to be present in sewage include some plasticizers, pesticides and degradation products of some detergents. Further widespread attention has been focused on the broad range of pharmaceutically active compounds which have been reported in municipal wastewaters in many parts of the world [26-27]. At this point there are no indications for limitations to water reuse caused by these compounds, although its effects are largely unknown.

Table 4 Reported occurrence of selected organic pollutants in water [µg/L]

Chemical pollutant	Influent STP; Raw sewage water (mean)	Effluent STP (mean)	River water; Surface water (mean)	Groundwater; Bank filtrate	Ref.
PAH:					
Pyrene			n.d. – 0.33 (0.05)		[30-31]
Pesticides:					
Diuron		0.62	n.d. – 0.43	n.d. – 0.004	[32, 33]
2,4-D (herbicide)		2.94	n.d. – 0.194		[32, 34]
Organic solvents:					
1,1,2-Trichloro-ethane			0.004 – 0. 9		[35, 36]
Endocrine disrupting compounds:					
Ethinylestradiol	< 0.001 – 0.01 (0.005)	< 0.0003 – 0.004 (0.0005)	n.d. – 0.0004		[37-38]
Pharmaceutically active compounds:					
Diclofenac	3.0; 0.47 – 1.9	n.d. – 0.93	n.d. – 1.2	n.d. – 0.37	[26-39]
Carbamazepine	0.2; 1.7	0.1 – 2.1	0.025 – 6.3	0.9 – 1	[26, 27]
Iopamidol		1.1 ± 0.1		0.4 - 1.4	[39, 40]
Complex-forming compounds:					
EDTA		700 [c]	8.0 – 19		[41]
Chlorination byproducts:					
N-nitroso-dimethylamine (NDMA)	< 0.001 – 0.009 (< 0.001)	< 0.001 – 0.063 (0.001)			[42]
Chloroform	1100 [d]	< 1 – 20 (3) [e]; 106 [d]	n.d. – 3.8 (0.5) [e]	198	[43, 44]

n.d. = not detectable (< limit of detection); [a] unpolluted soil, [b] sediment, [c] industrial sewage treatment effluent, [d] USA, [e] Europe

As in drinking water, byproducts of disinfection processes may yet prove to be among the chemicals of greater concern in reclaimed water. Recent attention in the USA and Canada has been given to the

detection of the potent carcinogen nitrosodimethylamine (NDMA) in chlorinated sewages. It is known that disinfection of wastewater effluents, especially those with relatively high organic loads, can lead to significant concentrations of NDMA [28, 29].
However, these compounds of concern are not specifically regulated by any current wastewater reuse guidelines of which we are aware. This situation is now on the very verge of change as scientists and regulators are grappling with how to best address the issues presented by a wide range of individual chemical contaminants.

3.3. Quality categories for reclaimed water reuse

There are different possible fields of application for reclaimed water reuse; therefore different water quality categories are needed. In Table 5 use-depending water qualities concerning the microbiological and chemical parameters are presented. Seven microbial water qualities, according to the Spanish recommended regulation [9], and four chemical water qualities are proposed. In contrast to the microbial risks, no explicit differences in chemical water qualities are required by reclaimed water irrigation of crops consumed raw or cooked. Other categories (e.g. microbial category V) include uses that need the same microbial quality but different chemical requirements. The uses of reclaimed water are so different from each other, except from the varying irrigation categories, that theoretically separate distribution water networks with different water quality must be implemented, although it is economically non feasible.

3.4. Monitoring key indicators and analytical cost calculation

The determination of general physical-chemical parameters such as pH, COD, BOD, TSS, turbidity, etc., defined in most guidelines concerning water and wastewater, can be easily performed following EN/ISO or national standards, whereas for heavy metals, included in some new and recommended guidelines [8, 20], multi-element or multi-component analysis determination methods like ICP-MS or AAS could simplify the analytical effort. That is also true for pesticides (EN ISO 6468:1996; EN 12918:1999).
Specific organic micropollutants will depend on the origin of the wastewater. They form a diverse group of chemicals and their environmental behaviour will vary significantly. Analytical detection limits are often close to or above concentrations that may be significant in the environment, and it is to say that chemical determination at such low concentrations is extremely challenging. Most persistent organic micropollutants are not regularly controlled and therefore standardised methods for the analysis of these trace compounds are not generally available. In the scientific literature, sophisticated analytical methods using GC-MS(MS) or LC-MS(MS) have been published [26, 37, 39, 45]. The methods described so far are often time-consuming and cost-intensive. Bioassays with specific effect orientation may provide one solution for some organic pollutants. An example is the estrogen cell bioassays like E(strogen)-screen assay for measuring (estrogenic) endocrine effects. Therewith whole effects (contributed by numerous individual chemicals) can be measured [46, 47]. For routine screening, bioassays may prove suitable if detection limits and analysis times can be optimised for practical application.

Table 5 Microbial and chemical water quality categories for different final uses of Reclaimed water [adapted from CEDEX, [7]]

Microbial category	Chemical category	Specific final use
I	1	- Residential uses: private garden irrigation, toilet flushing, home air conditioning systems, car washing. - Aquifer recharge by direct injection into the soil for irrigation purpose.
II	1	- Bathing water.
III	1	- Urban uses and facilities: irrigation of open access landscape areas (parks, golf courses, sport fields …). Street cleaning, fire-fighting, ornamental impoundments and decorative fountains. - Greenhouse crops irrigation. - Irrigation of raw consumed food crops. Fruit trees sprinkler irrigated. - Unrestricted irrigation.
IV	1	- Irrigation of pasture for milking or meat animals. - Irrigation of industrial crops for canning industry and crops not raw-consumed. Irrigation of fruit trees except by sprinkling. - Irrigation of industrial crops, nurseries, fodder, cereals and oleaginous seeds.
	2	- Impoundments, water bodies and streams for recreational use in which the public's contact with the water is permitted (except bathing).
V	1	- Irrigation of forested areas, landscape areas and restricted access areas. Forestry.
	2	- Aquaculture (plant or animal biomass).
	3	- Aquifer recharge by localised percolation through the soil.
VI	2	- Surface water quality, impoundments, water bodies and streams for recreational use, in which the public's contact with the water is not permitted
VII	4	- Industrial cooling, except for the food industry.

The calculated analytical costs concerning the microbial parameters are presented in Table 6. The determination of *Legionella*, *Giardia*, *Cryptosporidium* and enteroviruses are the most expensive parameters. On the other hand, *E.coli* analysis present a very low price for their quantification (less than 6 € per analysis).

In relation with control frequency of microbiological parameters (see Table 6), *Legionella*, *Giardia*, *Cryptosporidium* and enteroviruses are determined with low frequency due to the high determination cost. Otherwise, the most habitual determination parameters are the *E. coli*, enterococci and bacteriophage.

Concerning the control frequency of the specified chemical parameters (Table 7), the simple and cheap parameters such as E.C., turbidity and COD can be measured in frequent time intervals, whereas the expensive indicator compounds of micropollutants or heavy metals need rare monitoring, e.g. once a year. In addition, the analytical effort could depend on the reclamation system. Large plants or even demonstration projects should perform a greater monitoring program including several micropollutants than small reclamation facilities.

Table 6 Cost calculation and frequency of microbiological analysis

Parameter	Frequency	Cost
Legionella	+	High
E. coli and similar	+++	Very low
Enterococci (*Salmonella*)	+++	Low
Nematode eggs	++	Medium
Taenia	++	Medium
Giardia and *Cryptosporidium*	+	High
Bacteriophage	+++	Low
Enterovirus	+	High

Frequency: +++, permanently – weekly; ++, monthly – once per year; +, once per 1 – 5 year. Costs per analysis: very high, > 200 €; high, 60 - 200 €; medium, 20 - 60 €; low, 6 - 20 €; very low, < 6 €

Table 7 Cost calculation and proposed measuring frequency of physical-chemical and chemical quality parameters

Parameter	**Example / indicators**	**Frequency**	**Costs**
Physical-chemical	pH, EC, Turbidity, TSS, Colour	+++	very low
Organic sum parameters	COD (TOC, DOC), BOD, DO	+++	low
	AOX	+++	medium
Nutrients	Total-N, NH^{4+}-N	+++	low
Residual chlorine	Cl_2 (if chlorination)	+++	low
Physical-chemical	Sodium absorption ratio (SAR), UV 254	++	very low
Minerals	NO_3^-, SO_4^{2-}, Cl^-, Total-P	++	low
(Heavy) metals	As, Cd, Cr(III,VI), Hg, Pb, B	+	medium
Organic micropollutants	Surfactants, Mineral oil	+	medium
(Heavy) metals	Al, Ba, Be, Co, Cu, Fe, Li, Mn, Mo, Ni, Se, Sn, Th, V, Zn	□	medium
Organic micropollutants	Aldehyde, Phenols	□	medium
Pesticides	Diuron; 2,4-D	□	high
Complex-forming substances	EDTA	□	high
Chloride solvents	if AOX > limit, e.g. TCE	□	high
Aromatic organic solvents	Benzene	□	high
PAHs	Benzo(a)pyrene	□	high
Pharmaceuticals	Carbamazepine, X-ray contrast media, Sulfamethoxa-zole	□	very high
Endocrine disrupters	E-Screen	□	very high
Disinfection (by)products	NDMA	□	very high

Frequency: +++, permanently - weekly; ++, 1 – 2 per month; +, 2 per month - once per year; □, once per 1 – 5 year. Costs per analysis: very high, > 200 €; high, 60 - 200 €; medium, 20 - 60 €; low, 6 - 20 €; very low, < 6 €

4. PROPOSED LIMITS FOR RECLAIMED WATER REUSE

According to recently appeared guidelines (e.g. Italy, Israel, Catalonia in Spain) and recent developments, the most important microbiological parameters with limits or limit ranges are compiled in Table 8 (bacteria) and Table 9 (other microorganisms) related to the final use of Reclaimed water. In accordance to guidelines [8, 20] and industrial principles [48] the following chemical limits for Reclaimed water reuse are set in Table 10. Important values not mentioned in the guidelines are also estimated including ecological effect concentrations described in the literature.

Table 8 Overview of the compiled and estimated microbiological limits for reclaimed water reuse I (bacteria, cfu = colony forming units, abs = absent)

Use	Total bacteria (cfu/mL)	Faecal coliforms (cfu/100mL)	*Clostridium perfringens* (cfu/mL)	*Legionella* (cfu/100mL)	Enterococci (cfu/100mL)	*Salmonella* (cfu/mL)
I	<1,000 - <10,000	abs	abs - 20	<100	abs	abs - 1,000
II	< 1,000	< 20 - < 1,000	abs - 10	-	<1,000	abs - 1,000
III	<10,000	abs - < 1,000	<10	<100	<20	abs - 1,000
IV	<10,000 - <100,000	abs - < 10,000	<10	abs	<1,000	<1
V	<100,000	abs - < 10,000	<100	-	<10,000	<0.1
VI	<10,000	< 200 - < 10,000	<1	-	<20	abs - 1,000
VII	<10,000	abs - < 10,000	<10	abs - < 100	<1,000	<1

Table 9 Overview of the compiled and estimated microbiological limits for reclaimed water reuse II (other than bacteria, pfu = plaque forming units, abs = absent)

Use	Enteroviruses (pfu/L)	Coliphages (pfu/L)	*Cryptosporidium and Giardia* (cyst/50 mL)	Nematode eggs (eggs/L)	*T. saginata* (egg/L)	*T. solium* (egg/L)
I	abs - 10	<1	< 1	<1 - 10	-	-
II	abs - 10	<1	<1	< 1	-	-
III	<1- <100	<1,000	<10	< 1	-	-
IV	-	-	-	< 1	-	-
V	-	-	-	< 1	<1	<1
VI	<100	<1,000	<10	< 1	-	-
VII	<1 - 0.04	-	-	< 1	-	-

Table 10 Overview of the compiled chemical limits for reclaimed water reuse from existing guidelines and proposed chemical limits depending on the specific use [mg / L]

	1	2	3	4
Parameter / chemical category	**Private, urban and irrigation**	**Environmental and aquaculture**	**Indirect aquifer recharge****	**Industrial cooling**
Parameters of very high analytical frequency (daily - weekly)				
pH	6.0 – 9.5	6.0-9.5	7 – 9	7.0-8.5
BOD	10 - 20	10 - 20		
COD	100	70 – 100	70-100	70
(or TOC)		(10 – 15)		(10 – 15)
Dissolved oxygen	> 0.5	> 3	> 8	> 3
AOX			25	
UV 254 absorbance [$cm^{-1} \cdot (10)^3$]	30 - 70	30 - 70	10	
Electrical conductivity [µS/cm]	3000	3000	1400	
TSS	10 - 20	10 - 20		10 - 20
Active chlorine (only if chlorination)	0.2 – 1.0	0.05		0.05
Total Kjeldahl N	15 – 25	10 - 20		10
Ammonium-N	2 - 20	1.5	0.2**	1.5
Parameters of high analytical frequency (monthly)				
Sodium absorption ratio [$mmol/L^{0.5}$]	5	5		
Na	150	150 - 200		200
Nitrate			25	
Chloride	250	250 - 400	100	400
Sulphate	500	500	100	
Total P	2 - 5	0.2 - 1		0.2
Parameters of medium analytical frequency (monthly – once a year)				
As	0.1 - 0.02	0.1 - 0.02	0.005	
B (total)	0.4	0.4	0.2**	
Cd	0.005	0.005	0.003	
Cr (total)	0.1 - 0.01	0.1 - 0.01	0.025	
Cr III	0.1	0.1		
Cr VI	0.005	0.005		
Hg	0.001 – 0.002	0.001 – 0.002	0.0005	
Pb	0.1	0.1	0.005	
F (total)	1.5 – 2.0	1.5 – 2.0		
Surfactant (total)	0.5 - 1	0.5 - 1		
Mineral oil	0.05	0.05		
Parameters of low analytical frequency (once per year – once per 5 years)				
Al	1 - 5	1 – 5		
Ba	10	10		
Be	0.1	0.1		
Co	0.05	0.05		
Cu	0.2 – 1.0	0.2 – 1.0		
Fe	2	2		
Li	2.5	2.5		
Mn	0.2	0.2		
Mo	0.01	0.01		
Ni	0.2	0.2	0.01	
Se	0.01 – 0.02	0.01 – 0.02		
Sn	3	3		
Th	0.001	0.001		
V	0.1	0.1		
Zn	0.5 – 2.0	0.5 – 2.0		

	1	2	3	4
Parameter / chemical category	**Private, urban and irrigation**	**Environmental and aquaculture**	**Indirect aquifer recharge****	**Industrial cooling**
Parameters of low analytical frequency (once per year – once per 5 years)				
CN (total)	0.1 - 0.05	0.1 - 0.05		
Pesticides (total)	0.05	0.05		
Pesticides and their metabolites, per subst. (country specific)			0.0001	
Pentachlorophenol	0.003	0.003		
Synthetic complex-forming subst., per subst. (e.g. EDTA)	0.0001	0.0001	0.0001	
Chloride solvent (total, if AOX > limit)	0.04	0.04		
Tetrachloroethylene, Trichloro ethylene	0.01	0.01		
Disinfection (by)products (only if chlorination)				
NDMA	0.0001*		0.0001*	
Trihalomethane	0.03	0.03		
Aldehyde (total)	0.5	0.5		
Aromatic organic solvent (total)	0.01	0.01		
Benzene	0.001	0.001		
Benzene(a)pyrene	0.00001	0.00001		
Phenol (total)	0.1	0.1		
Endocrine active substances (E-Screen)	0.0001*	0.0001*	0.0001*	
Pharmaceuticals (per subst., e.g. Carbamazepine, X-ray contrast media, Sulfamethoxazole)	0.0001*	0.0001*	0.0001*	

* proposed value

** When performing indirect aquifer recharge there has to be an option for the operator not to desalinate, if the recovered water is not used for potable uses, or even if used as potable water. This can be done at higher dilution of the aquifer. This is also true for the other salinity parameters like boron and the ammonium concentration. In the case of water for indirect recharge the ammonium concentration should be permited to be up to 20 ppm, depending on the aquifer conditions.

5. RISK ASSESSMENT

5.1. General principles

Risk evaluation in relation with wastewater reclamation and reuse became at the end of the last century one of the more interesting tools to establish relationships between reclaimed water quality and the health hazards associated to the practice of reuse.

In this sense, there are several steps to develop in any facility, initially devoted to establish the quality of reclaimed water. The application of the risk related tools starts with risk assessment, is followed by risk calculation and ends with the management and communication.

The basis of it all is the availability of reliable data on the analytical parameters which exert a noticeable impact on the whole risk. Those data are to be elaborated in a way that allows ranking the parameters and giving a figure, a magnitude of the risk associated to a unique chemical or microorganism. Apart from it, and making the analysis more difficult, there are antagonisms and agonisms (synergies) which can exert a final real effect on the target: the human being, the animals or the whole ecosystem.

The studies to be undertaken before the calculations for a hazardous substance or organism are varied and derive from several sciences, plus the studies on the degradation of the component in the environment. The solubility, decay rate, capability to be degraded by biological or physico-chemical means, and the calculation of several constants appear to be paramount.

The final results are complicated formulae, which can give an estimation of the difficulties to reach an objective determination of the risk associated to reuse. The figures are always approximate, because the ecosystems, and especially the biocenosis in the ecosystem, never act in the same way. The main cause is the variability inherent to any system in nature.

The chemical and microbial limits for different reclaimed water uses need a reliable risk assessment , especially for the not assessed compounds like the organic trace pollutants and for several microorganisms. The risk assessment should follow the European Technical Guidance Document (TGD) on Risk Assessment, based on the Commission Directive 93/67/EEC, Commission Regulation (EC) 1488/94 and Directive 98/8/EC. Part I Chapter 2 of the TGD describes the human risk assessment and in Part II Chapter 3 the environmental risk assessment is specified. The human and environmental risk assessment proceeds always in the following sequence:

a. hazard identification

b. dose (concentration) – response (effect) assessment

c. exposure assessment and

d. risk characterization.

5.2. Human health risks by reclaimed water reuse

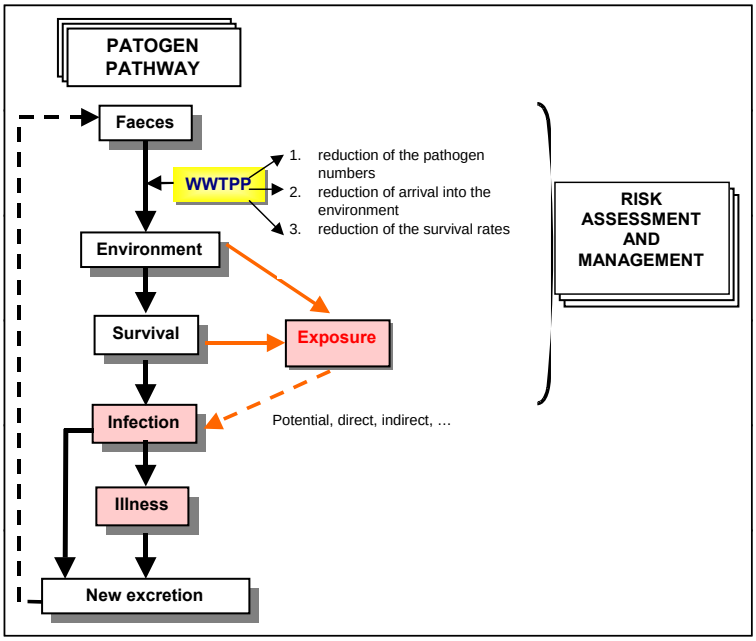
Risks associated with microorganisms have been addressed with the greatest urgency and are now dealt with in a thorough manner by guidelines used in many countries. Conversely, guidelines devoted to chemical contaminants are typically limited to bulk parameters like chemical oxygen demand (COD), biochemical oxygen demand (BOD), pH and total suspended solids (TSS). In many situations these simple parameters provide suitable surrogate indications of the likely presence of chemical species of

concern. However, for more highly treated reuse waters, they are of limited sensitivity. Furthermore, for some applications, an accurate assurance of specific chemical concentrations will be important.
For the description of microbiological hazard it seems necessary to describe the pathway of pathogens through the treatment, reclamation and reuse schemes and establish how this pathway is modified (Figure 1). Faeces, and consequently pathogens, can reach the environment through wastewater, either untreated, treated, or reclaimed. For this reason, one of the objectives of wastewater treatment plants is to reduce the microbiological content by:

1. reducing the number of pathogens,
2. reducing the number of pathogens reaching the environment,
3. reducing the survival of pathogens in the environment.

It is to remember that if there is an exposition to pathogens, an infection or illness can possibly appear. Nevertheless, the cycle does not finish here, because there is a feed-back of pathogens. In other words, people who are infected, ill or are a healthy carrier, re-excrete the pathogens through faeces.

Figure 1 Pathogen pathway in reclamation and reuse systems



5.2.1. Hazard identification

The first step of qualitative human exposure assessment will be to identify likely and potential routes of human exposition to contaminants. This will determine the significance of contaminants concentrations in environmental media such as soil, air or water.
The human health hazards by reclaimed water reuse depend on the contaminant pathways and the reclamation processes. The most relevant pathways concerning human hazards are specified in Table 11 for the case of irrigation. In individual cases, it will be necessary to make initial judgements regarding the significance of each identified exposure route.

Table 11 Possible exposure pathways of contaminants affecting human health via reclaimed water reuse for irrigation

No.	Pathway	Scenario
1	Reclaimed water irrigation → soil → plant uptake → food production → human	• Ingestion of food plants cultivated on land irrigated with reclaimed water
2	Reclaimed water irrigation → soil → plant uptake → animal uptake → human	• Ingestion of meat/animal products from animal pasture on land irrigated with reclaimed water
3	Reclaimed water irrigation → soil → vadose zone → groundwater → human	• Ingestion of drinking water produced from groundwater polluted by reclaimed water
4	Reclaimed water irrigation → atmosphere → human	• Incidental exposure to aerosols generated by spray irrigation
5	Reclaimed water irrigation → human	• Residential cross connection: proximity to wastewater irrigated zones (children) • Direct contact with irrigation water
6	Reclaimed water irrigation → soil → human	• Ingestion of irrigated soil (children)

Microbial risks

The risk of infection from waterborne pathogens is still not well known and consequently the list of identified waterborne human pathogens continues to expand. The steps in microbial hazard identification are as indicated in Table 12.

Table 12 Description of the steps usually found in microbiological hazard identification [modified from 18]

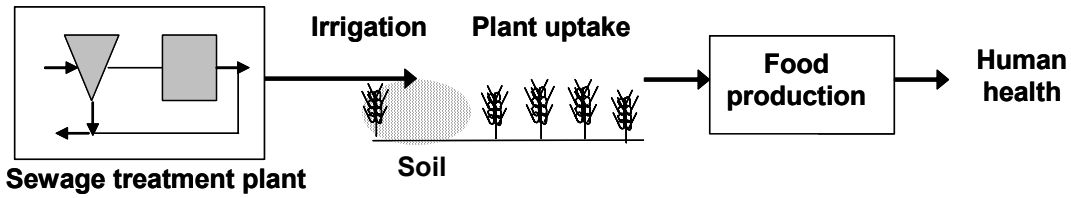
Step		Comments/Observations
1.	Identification of the microorganisms causing illness	Use Koch's postulates, which demonstrate that the agent is found and is the cause of specific types of disease; and when transmitted causes a similar disease in the person newly exposed.
2.	Diagnostic tools development	For the identification of the symptoms, the infection, and the microorganism in host specimens (sputum, stools, blood, …)
3.	Understanding the disease process	From exposure (e.g. respiratory) to infection (colonisation of the human body) to development of pathology, disease and death.
4.	Transmission routes	Identification of possible routes.
5.	Assessment of microorganism factors	Virulence factors and components of the microorganism and its life cycle that aid in understanding transmission and the disease process.
6.	Use of diagnostic tools	Evaluation of the incidence and prevalence of disease in populations (endemic risks) and investigation of outbreaks (epidemic risks).
7.	Development of models	Study the disease process and approaches for treatment (usually, animal models).
8.	Host evaluation	Evaluation of the role of the host immune system in combating the infection and the possible development of vaccines for prevention.
9.	Epidemiology	Epidemiological studies associated with various exposures.

Chemical risks

The consumption of crops irrigated with wastewater, the major anticipated pathway of human exposure, is illustrated in Figure 2. This risk is not limited to farmers who consume their own agricultural products, but it also concerns the population that buys irrigated products.

In case of dual reticulation systems in urban reuse applications, cross-connection at residential use must be strictly avoided by the reclaimed water supply and distribution management (installation of separated pipelines, fool proof identification of the pipelines, etc.).

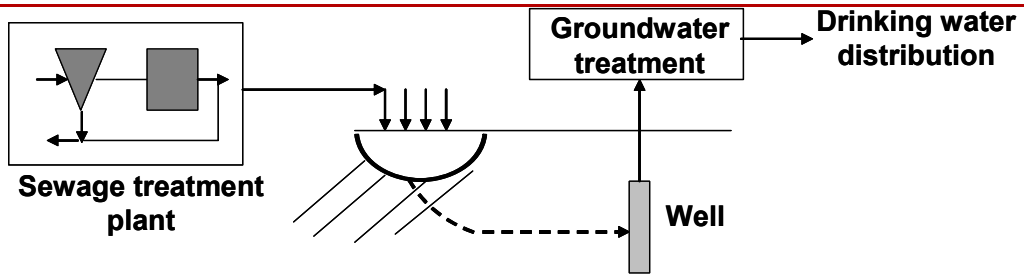
Figure 2 Primary exposure pathway of humans to chemicals via agricultural irrigation



Direct contact with water during the irrigation process (pathways 4 and 5) concerns farmers who cultivate without taking the necessary precautions to protect themselves from the risk of contamination. Contamination can take place through simple direct contact with wastewater containing parasites of human or animal origin; these are able to cross the cutaneous barrier and penetrate in the organism.

Groundwater recharge with reclaimed water may affect humans' health if the recharged water is used for drinking water production and if the pollutants are persistent or not removed sufficiently by the soil/aquifer barrier (Figure 3). Contaminants in the subsurface environment are subject to processes such as biodegradation by microorganisms, adsorption and subsequent biodegradation, filtration, ion exchange, volatilization, dilution, chemical oxidation and reduction, chemical precipitation and complex formation, and photochemical reactions (in spreading basins) [9].

Figure 3 Primary exposure pathways of chemicals to humans via groundwater recharge



5.2.2. Dose-response assessment

Dose-response assessment is the second step in the risk analysis when working with reclaimed water reuse. The different types of risks are to be discussed, like in the previous steps.

Microbial risks

The objective of dose-response assessment is to develop a relationship between the level of microbial exposure and the likelihood of occurrence of an adverse consequence [18]. Rowe and Abdel-Magid [49] indicate more simply that is the analysis of what degree of harm results from differing "doses" of the threat (exposure duration and intensity).

Current pathogen dose-response data is limited, but this limited information is often used to extrapolate risk estimates of the risk of infection due to exposure to reclaimed water into regions for which no data exists. Such estimates may demonstrate that the risk of infection is lower than endemic levels of infection and may provide a rationale means for establishing public policy [18].

Pathogen infectivity can vary due to a range of factors. These factors may be due to the pathogen's own characteristics, individual host response, and environmental factors. In any case, a contact is necessary.

Table 13 Summary of health risk associated with use of wastewater in irrigation [50] – a potential disease transmission.

Group Exposed	**Health Threats**		
	Nematode infection	**Bacteria / Viruses**	**Protozoa**
Consumers	Significant risks of *Ascaris* infection for both adults and children with untreated wastewater; no excess risk when wastewater treated to < nematode egg/liter except where conditions favour survival of eggs	Cholera, typhoid and shigellosis outbreaks reported from use of untreated wastewater; ser-positive responses for *Helicobacter pylori* (untreated); increase in non-specific diarrhoea when water quality exceeds 104 FC/100 mL	Evidence of parasitic protozoa found on wastewater irrigated vegetable surfaces but not direct evidence of disease transmission
Farm workers and their families	Significant risks of *Ascaris* infection for both adults and children with contact with untreated wastewater; risk remain, especially for children in contact with wastewater treated to < 1 nematode egg/liter; increased risk of hookworm infection to workers	Increased risk of diarrhoeal disease in young children with wastewater contact if water quality exceeds 10^4 FC/100mL; Elevated risk of *Salmonella* infection in children exposed to untreated wastewater; elevated seroresponse to Norovirus in adults exposed to partially treated wastewater	Risk of *Giardia intestinalis* infection was insignificant for contact with both untreated and treated wastewater, Increased risk of Amoebiasis observed with contact with untreated wastewater
Nearby communities	*Ascaris* transmission not studied for sprinkler irrigation but some as above for flood or furrow irrigation with heavy contact.	Sprinkler irrigation with poor quality water 10^{6-8} TC/100 mL, and high aerosol exposure associated with increased rates of infection; use of partially treated water 10 4-5 FC/100 mL or less in sprinkler irrigation is not associated with increased infection rates	No data for transmission of protozoan infections during sprinkler irrigation with wastewater.

Source: Blumenthal and Peasey, 2002; Blumenthal, 2000; Armon 2002; adapted from [50].

Upon entering the host and for a short period of time, the pathogen must adapt to a new environment (incubation). During this period the pathogen may be undergoing biochemical transformations that will allow the organism to utilise the host's resources, which can result in infection or colonisation of the host. While the pathogen is adapting to the new environment, the host's immune system may be attempting to fend off the infection by producing antibodies. If the pathogen is successful in colonisation and reproduction, the host may manifest symptoms of the disease. Unsuccessful attempts to colonise and reproduce may mean the host may not show signs of illness, but may only show evidence of infection through biochemical tests designed to find the antibodies produced to fend off the pathogen [18].

Because of limited analytical or diagnostic tools, microbial risk assessments attempt to cover a range of situations often extrapolating into regions of low pathogen concentration by using models. Such an approach assumes that small pathogen concentration may produce an adverse response. Ideally, a dose-response model should be able to differentiate between infection and illness, because although may be used interchangeably by some, the terms have very distinct meanings. The current models used for microbial risk assessment only predict the risk of infection, not the risk of contracting the disease [18].

Except viral infections, no other pathogenic agent can confer a solid immunity that can protect human being against reinfection.
Low quality treated wastewater may contain bacteria and other organisms which are harmful to agricultural workers and to those who handle, cook, or eat the plants. Wastewater may even contain bacteria and other organisms which, when eaten by animals, may in turn infect the people who eat the contaminated meat (see Table 13).
There are three kinds of useful models: log-normal, exponential, and beta-Poisson.

Chemical risks

Chemicals can cause acute and chronic toxicity by oral disposal or inhalation. Toxicity of chemicals to an organism is normally defined in terms of dose-response relationships. Chronic toxicity includes teratogenic, mutagenic and carcinogenic effects. When the target organisms are humans, dose-response relationships may be derived from data obtained in epidemiological investigations, extrapolations from animal studies, or toxicity assays on mammalian or bacterial cells. Epidemiological data can provide the most realistic cause-effect relationships, but are only available for a very limited number of chemicals. The long latency period of disorders caused by environmental toxicants, such as cancer, reduces the quality of the data by hindering the determination of the effects. Dose-response relationships, once established, may then be used to derive an “acceptable daily intake” (ADI) or reference doses (RfD) for each specific chemical, usually with the application of safety factors to account for any uncertainty. Finally, established ADI values can be used to determine environmental predicted no effect concentrations (PNEC).
Although ADI data are available for heavy metals, only a few human ADI or RfD values exist for organic (trace) compounds until now, because their toxicity is often unknown.

5.2.3. Exposure assessment

While the hazard identification portion of the risk assessment can establish causality, it does not provide quantitative information on exposures. Often models were used to estimate the exposure. Depending on the origin of the wastewater, the reclamation process and the specific reuse the exposure assessment has to be adapted to the specific case. In this report three characteristic case studies were analyzed.

Microbiological risks

The objective of the exposure assessment is to determine the amount, or number, of organisms that corresponds to a single exposure (dose), or the total amount or number of organisms that constitute a set of exposures [15].

The exposure of consumers (a specific case) to contaminants due to wastewater irrigation depends on several factors:

1. The quality of the applied wastewater (reclaimed water)
2. The irrigation method
3. The elapsed time between irrigation, harvest and subsequent product consumption
4. Consumer's habits.

The factors can be enlarged or modified, including initial pathogen concentration in wastewater and pathogen removal and survival in reclamation treatments (finally define 1); release to the environment (corresponds to 2); survival in the environment (factor 3), route of human exposure to target organ, exposure frequency, infection/infectivity and disease or outcome; in some way related to consumer's habits (factor 4).

There is a variety of exposure routes. Each route of exposure can be characterised by a contact rate and an exposure frequency. The contact rate is the amount of that particular material that is ingested, inhaled, or results in other direct exposure for each time the activity that results in exposure occurs. The exposure frequency is the rate at which the activity resulting in the exposure occurs. Both the contact rate and the exposure frequency may be characterised by point estimates and also by distributions reflecting uncertainty and variability [15]. Table 14 summarises point estimates for a number of exposures that may carry microbial agents.

For microbiological risk in reclamation and reuse, it is important to characterise some routes of exposure in greater detail than in chemical risk assessment, and there are some routes of exposure that are virtually unique to microbial risk (i.e. transfers between skin and hands).

There are several steps or barriers that decrease the likelihood of pathogen survival, but exposure assessments are only as good as the understanding of the disease etiology. The less is known about the etiology of disease, the weaker the model and the less certain is the outcome. If each of the exposure steps could be identified, then the estimates of pathogen survival would accurately estimate the overall threat posed by the pathogen [18].

Table 14 Point estimates for consumption/contact factors [15]

Pathway	Contact rate	Exposure frequency
Ingestion of drinking water	1.4 L/day	365 days/year
Ingestion of surface water while swimming	50 mL/h 2.6 h/swim	7 swims/year
Soil ingestion by children	200 mg/day (< 6 years in age) 100 mg/day (> 6 years in age)	Depends on the context
Inhalation	20 m^3/day (adult) 15 m^3/day (child)	365 days/year
Inhalation during showering	0.07 m^3/shower	365 showers/year
Ingestion of fish	0.113 kg/meal	48 meals/year

Chemical risks

An essential component of any chemical risk assessment process must be a means of determining or estimating levels of exposure. This requires a qualitative analysis of routes of exposure of a chemical to the target organism, as well as quantitative information such as the concentration of target chemicals in relevant media. For preventive risk assessment, both aspects inevitably require some degree of prediction. In this example the environmental concentrations in the soil matrix are of greatest significance to this case. Accordingly, PEC and PNEC will be determined for the soil matrix.

5.2.4. Risk calculation

Characterization of risk is the final step in risk assessment. Risk characterization integrates the results of dose-response and exposure assessment into a risk statement that includes one or more quantitative estimates of risk [15]. The goal of risk characterization is to provide an understanding of the type and magnitude of the adverse effect that a particular pathogen could exert under particular circumstances [18]. More simply, is the description of the nature and often the magnitude of human risk, including attendant uncertainty [49].

The elements of risk characterization at a minimum should include [18]:

- Generation of a quantitative estimate of risk
- Description of uncertainty
- Presentation of the risk estimate
- Communication of the results of the risk analysis to the risk manager

Uncertainty can be defined as a lack of precise knowledge as to what the truth, whether qualitative or quantitative, really is. There is uncertainty in the estimated risk number because of uncertainties in the data and models used at each step in the risk assessment process [18].

In a NRC book (1996) two types of uncertainty in risk assessment are stated: parameter uncertainty and model uncertainty [51].

1. Parameter uncertainty: estimated values of parameters have uncertainties stemming from the following sources:

- measurement errors
- random errors in analytic devices
- systematic biases using generic or surrogate data instead of analysing the desired parameter directly
- random sampling error
- non-representativeness

2. Model uncertainty. These uncertainties arise because of unknowns in the scientific theory that require the use of reasonable assumptions:

- errors in model structures
- errors induced by oversimplified representations of reality
- usage of surrogate variable(s) for ones that cannot be measured
- failing to account for correlations that cause seemingly unrelated events
- the extent of aggregation used in the model

There are two mathematical procedures which can be used to propagate uncertainty in exposure assessment: one is a procedure using the single "best guess" value and its standard deviation as a measure of uncertainty, the other is a way of estimating probability density function by Monte Carlo simulation. The former is commonly referred to as a deterministic analysis, whereas the latter is known as a stochastic analysis. The Monte Carlo simulation method builds up successive scenarios using input values that are randomly selected from probability distributions, commonly utilising the pertinent computer software.

In order to ensure adequate performance to protect public health, it is important that overall process performance be robust enough to compensate for minor excursions in unit process performance.

Microbiological risks. Quantitative risk assessment

There is growing interest in using risk assessment as a means of developing water quality standards and other environmental regulations. Unfortunately, risk assessment related to human pathogens is not as advanced or accepted as are the risk assessment methodologies for chemicals. Quantitative risk assessment appeared promising, but those methods had not been sufficiently refined to a level where there was reasonable consensus on the validity and acceptance of the models.

Risk assessment for pathogenic organisms involves the evaluation of the potential for an adverse health effect to occur as a result of human exposure to reclaimed water. Models have been developed for a relatively small number of microorganisms.

The quantitative risk assessment models attempt to estimate the risk of infection for an exposed individual. It should be noted that only a fraction (for some organisms, a relatively small percentage) of individuals that become infected will actually exhibit symptoms of disease.

Most investigators attempting to use quantitative risk assessment models for human pathogens have standardized on the use of an acceptable risk threshold of 1×10^{-4}. In some instances, this 1×10^{-4} threshold has been applied to annual risks, while some investigators have evaluated single events [52].

Once the etiologic agent has been identified and human exposure to the pathogen is known to cause infection and disease, the next step is to determine how exposure to different concentrations of the

pathogens might elicit a response in humans. Current pathogen dose-response data is limited. The current models used for microbial risk assessment only predict the risk of infection, not the risk of contracting the disease. Table 15 presents the dose-response models for different microorganisms from various studies.

Table 15 Probability of infection models and best fit dose-response parameters for various human feeding studies [53]

Organism	Best model[a]	Model parameters[b]
Rotavirus	Beta-Poisson	$\alpha = 0.26$ $\beta = 0.42$
Echovirus 12	Beta-Poisson	$\alpha = 0.374$ $\beta = 186.7$
Cryptosporidium	Exponential	$r = 0.00467$
Giardia	Exponential	$r = 0.0198$
Salmonella	Exponential	$r = 0.00752$
E. coli	Beta-Poisson	$\alpha = 0.1705$ $\beta = 1.61 \times 10^6$
Shigella	Beta-Poisson	$\alpha = 0.248$ $\beta = 3.45$

a. Models

$Pi = Pi = 1 - (1 + N/\beta)^{-\alpha}$ (Beta-Poison)

$Pi = 1 - e^{-rN}$ (Exponential)

b. Model parameters

Pi = probability of infection (ability of the organism to establish and reproduce in the intestine)

N = exposure, expressed as numbers of microorganisms ingested (CFU of bacteria, cysts, or oocysts of *Giardia* or *Cryptosporidium*; or PFU (plaque forming units) of viruses)

α, β, r = constants for specific organisms that define the dose-response model

For nonpotable reuse activities, some investigators have focused on single exposures, while others have looked at annual risks. Dose is based on the type of reuse activity. Suggested single event consumption values are shown in Table 17 for a range of nonpotable reuse activity [52].

Table 16 Doses used for nonpotable reuse activities [52]

Reuse activity	Dose (mL)
Recreational use, swimming, full body contact	100
Residential irrigation (worst-case, single-event ingestion)	100
Consumption of edible crops (assumes direct contact application methods)	10
Residential irrigation (routine exposure)	1
Irrigation of public access areas, golf courses, parks, etc.	1
Exposure to aerosols	0.1

Risk assessment of St. Petersburg's water reclamation facility

The risk models used are based on dose-response models and are shown in Table 16. Their analysis was based on ingestion of 100 mL of reclaimed water.

Table 15 presents models for a highly infective Rotavirus and a moderately infective Echovirus. Risks associated with ingestion of the moderately infective virus are orders of magnitude less than the risks associated with ingestion of the same numbers of a highly infective virus. Of course, there are also viruses that can be considered as having low infectivities. When analyzing a water sample for enterovirus, it is not attempted to identify individual viruses present (due to factors and limitations such as cell cultures used, analytical equipment, laboratory capabilities, and cost). As a result, in estimating risks using the model, assumption must be made that all viruses present are highly infective Rotavirus. If it is to assume that the virus present were distributed in equal proportions between highly infective virus, moderately infective virus, and virus having low infectivity, the overall risk (based on expected value calculations) would be approximately one-third of the risk predicted for a highly infective virus.

The results of the risk calculations for St. Petersburg's reclaimed water in Table 17 consider only a single ingestion of 100 mL of reclaimed water. Simple probability calculations can be used to estimate annual risks resulting from more prolonged exposure to reclaimed water [52].

Table 17 Estimated risks form the St. Petersburg's study [52]

Organism	Concentration #/100 mL[a]	Exposure #/100 mL	Units	Estimated risk[b]	Notes
Rotavirus	0.01	1.0×10^{-5}	PFU[h]	6.2×10^{-6}	c; d
Echovirus	0.01	1.0×10^{-5}	PFU[h]	2.0×10^{-8}	d; e
Rotavirus	0.13	1.3×10^{-4}	PFU[h]	8.0×10^{-5}	c; f
Echovirus	0.13	1.3×10^{-4}	PFU[h]	2.7×10^{-7}	e; f
Cryptosporidium	0.75	7.5×10^{-4}	Oocysts	3.5×10^{-6}	d; g
Cryptosporidium	5.35	5.35×10^{-3}	Oocysts	2.5×10^{-5}	f; g
Giardia	0.49	4.9×10^{-4}	Cysts	9.8×10^{-6}	d; g
Giardia	3.3	3.3×10^{-3}	Cysts	6.6×10^{-5}	f; g

a. Observed concentrations in reclaimed water at a St. Petersburg water reclamation facility.
b. Estimated risk of infection to an exposed individual.
c. Assumes that all viruses are highly infectious Rotavirus.
d. Average concentration observed in the reclaimed water.
e. Assumes that all viruses are moderately infectious Echovirus 12.
f. Maximum concentration observed in reclaimed water.
g. Assumes that all cysts and oocysts are viable.
h. Plaque forming units.

Table 18 presents the results of such calculations for four scenarios. In the first scenario, an individual is exposed to aerosols. The exposure (thorough ingestion) was estimated at 0.1 mL per day for 365 days. In the second scenario, a golfer or park visitor is exposed to 1 mL on each of 60 days during a year. In the third scenario, a resident is exposed to 1 mL of reclaimed water on each of 150 days during a year. The fourth scenario involves the conservative assumption of exposure of a residential property owner to 1 mL of reclaimed water on each of the 365 days in a year. In all cases, the average concentrations observed in the St. Petersburg study were used [52].

Table 19 presents concentrations of the various pathogens corresponding to a risk of 1×10^{-4}. This is based on a single event and data are presented for several doses. The doses used are: 100 mL for recreation (swimming), 100 mL for residential irrigation using hose bibs, 1 mL for irrigation of other public access areas, and 0.1 mL for exposure to aerosols.

Table 18 Annual risks [52]

Organism	Exposure scenario			
	Case 1 **0.1 mL/day** **365 days/year**	**Case 2** **1 mL/day** **60 days/year**	**Case 3** **1 mL/day** **150 days/year**	**Case 4** **1 mL/day** **365 days/year**
Rotavirus[a]	2.3×10^{-6}	3.7×10^{-6}	9.3×10^{-6}	2.3×10^{-5}
Echovirus[b]	7.3×10^{-9}	1.2×10^{-8}	3.0×10^{-8}	7.3×10^{-8}
Cryptosporidium	1.3×10^{-6}	2.1×10^{-6}	5.3×10^{-6}	1.3×10^{-5}
Giardia	3.5×10^{-6}	5.8×10^{-6}	1.5×10^{-5}	3.5×10^{-5}

a. Assumes all virus are highly infective Rotavirus
b. Assumes all virus are moderately infective Echovirus

Table 19 Pathogen concentrations corresponding to 1 x 10^{-4} risk [52]

Organism	Units	Concentration needed for 1 x 10^{-4} risk (#/100 mL)			
		0.1 mL	**1 mL**	**10 mL**	**100 mL**
Rotavirus[a]	PFU	165	16.5	1.65	0.165
Echovirus[b]	PFU	50,000	5,000	500	50
Cryptosporidium	Oocysts	22,000	2,200	220	22
Giardia	Cysts	5,000	500	50	5

a. Assumes all virus are highly infective Rotavirus
b. Assumes all virus are moderately infective Echovirus

The risk models can be used to calculate the volume of reclaimed water that would have to be consumed in order to result in a risk of 1 x 10^{-4}. Table 20 presents the volume of reclaimed water that would have to be ingested to result in a 1 x 10^{-4} risk of infection. The results show that for all organisms, over 1.0 L of reclaimed water would have to be ingested.

Table 20 Volume needed to be ingested to have a 1 x 10^{-4} risk [52]

Organism	Volume (L)
Rotavirus (highly infective)	1.65
Echovirus (moderately infective)	500
Cryptosporidium	2.9
Giardia	1.0

Several of these organisms are reported to be "highly infectious". Some individuals maintain that exposure to even a single organism may be sufficient to cause infection. Using models described in Table 15 to ingestion of a single microbe yields probabilities of infection of 27 percent for a Rotavirus, 0.2 percent for a Echovirus, 0.5 percent for a *Cryptosporidium* oocyst, and 2 percent for a *Giardia* cyst. As noted previously, infection does not necessarily result in a symptomatic disease.

Microbiological risks. Disability adjusted life years

Disability adjusted life years (DALYs) have been launched by the World Bank and backed by the World Health Organisation as a measure of the global burden of disease [54, 55].

The Disability Adjusted Life Year (DALY) is the only quantitative indicator of burden of disease that reflects the total amount of healthy life lost, to all causes, whether from premature mortality or from some degree of disability during a period of time [56]. These disabilities can be physical or mental. The intended use of the DALY is to assist (i) in setting health service priorities; (ii) in identifying disadvantaged groups and targeting of health interventions; and (iii) in providing a comparable measure of output for intervention, program and sector evaluation and planning.

The number of DALYs estimated at any moment reflect the amount of health care already being provided to the population, as well as the effects of all other actions which protect or damage health. Where treatment is possible -whether preventive, curative or palliative- the effectiveness of the intervention is the reduction in disease burden which the treatment produces. Effectiveness is measured in the same units (DALYs) as disease burden, and so can be compared across interventions which deal with different problems and produce different outcomes. In other words, the DALY can be used to measure the gains in health attributable to different actions and add them up.

The proponents of the DALY use this measure for two purposes: (i) to measure the burden of disease, and (ii) to increase the allocative efficiency of the sector by identifying health interventions that, for a given budget, will purchase the largest improvement in health, as measured by the burden of disease indicator (DALY). DALYs can be used in three interrelated areas:

i) for epidemiological surveillance of the total disease burden (number of DALYs)

ii) to measure cost-effectiveness of interventions (cost per avoided DALY)

iii) to decide what should be included in a country's "core services" (the package of essential health care services). Within a fixed budget, it has been suggested that only the most cost-effective interventions should be included (cost per avoided DALY).

Definition

The disability adjusted life year or DALY is a health gap measure that extends the concept of potential years of life lost due to premature death (PYLL) to include equivalent years of 'healthy' life lost by virtue of being in states of poor health or disability. The DALY combines in one measure the time lived with disability and the time lost due to premature mortality. One DALY can be thought of as one lost year of 'healthy' life and the burden of disease as a measurement of the gap between current health status and an ideal situation where everyone lives into old age free of disease and disability [3].

DALY is a health outcome measure with two main components:

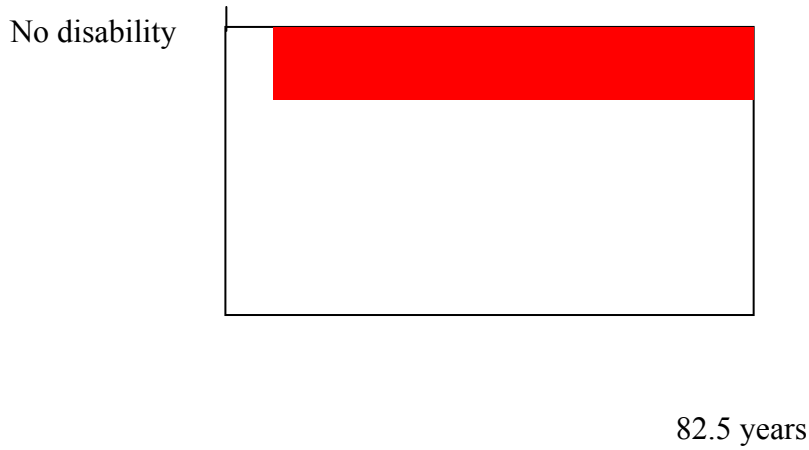
- Quality of life reduced due to a disability
- Lifetime lost due to premature mortality.

The Figures 4 to 6 show how the burden of disease is measured for a "standard" individual. Burden is measured along two dimensions: time lived with disability and time lost due to premature mortality [57].

The x-axis shows life expectancy for the "normal life. The "standardised" maximum life span is 82.5 years for females and 80 years for males. The y-axis shows degree of disability. The "normal" life is quantified as the total area in the box, a combination of the number of years lived and the quality of life, or degree of disability. From this ideal state of the world it is possible to calculate the burden of disease caused by premature death or disability.

If for example a girl aged 5 happens to become a victim of a mine explosion causing a below-knee amputation, and she does not die but is rehabilitated to a health state with some loss of physical functioning, her DALY loss could be depicted as the red area in the Figure 4.

Figure 4 DALYs due to living with disability (red area measures DALYs. Red + White is a "normal" life) [58]



Premature death from a myocardial infarction, say at age 50, would produce the DALY loss as depicted by the red area in the Figure 9 This patient's loss is 32.5 years. No adjustment is made for disability because the patient dies.

Figure 5 DALYs due to early death (red area measures DALYs. Red + White is a "normal" life) [58]

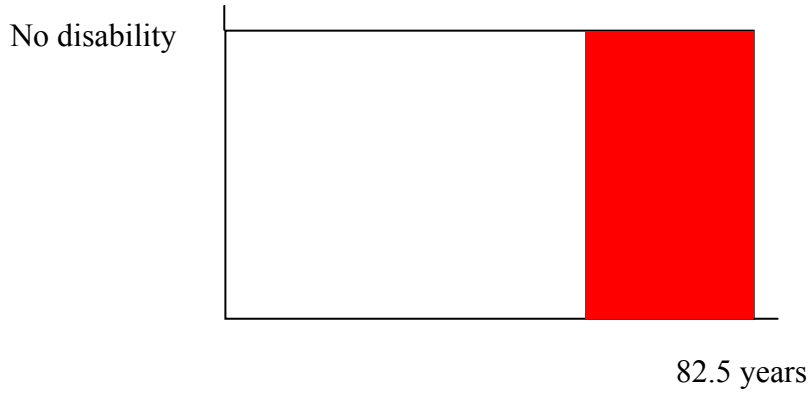
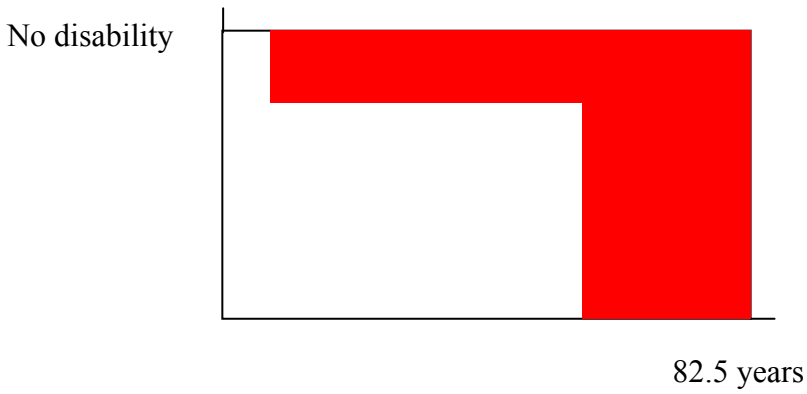


Figure 6 shows a woman who lives with a disability, for instance deafness from the age of 5 and dies prematurely at the age of 50.

Figure 6 DALYs due to disability and premature death combined (red area measures DALYs. Red + White is a "normal" life) [58]



Values incorporated in the DALY indicator

The five key social preferences or values that are incorporated into the indicator of burden of disease "DALY" are the following:

a. Duration of time lost due to a death at each age

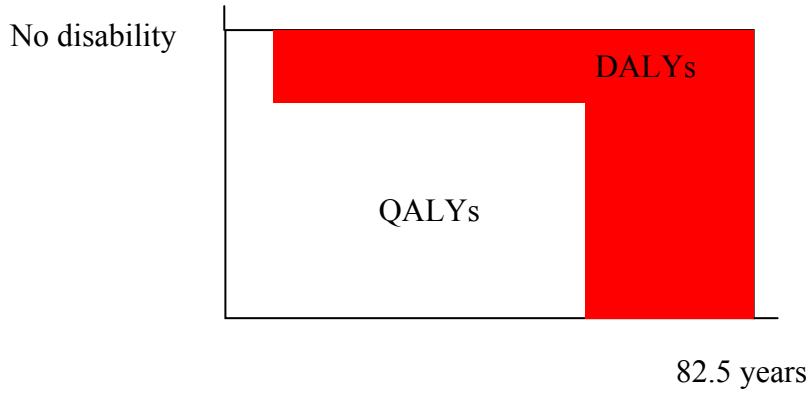
Duration of time lost due to a death at each age, which is used to measure years of life lost due to premature mortality (or the number of years of life gained by averting death). This measurement requires defining the potential limit of life; in the case of DALYs, standard years of life lost are used. The standard has been chosen to match the highest national life expectancy observed, which is that of Japanese women (82 years). For a specific standard, the expectations are based on model life-table West Level 26, which has a life expectancy at birth for females of 82.5. The potential life expectancy at birth for males has been set at 80. This is based on the highest current life expectancy, and thus avoids giving any population a lower expectancy than they actually experience. This also means, however, that the life expectancy assumed in DALYs is much higher than in, for example, sub-Saharan Africa. This will be seen to overestimate the burden of disease given current life expectancy. Thus the choice of life tables is fundamental to the results obtained; and the table chosen raises questions about comparisons between, for example, Japan and sub-Saharan Africa [59, 60].

b. Disability weights

Disability weights or degrees of incapacity or suffering associated with different non-fatal conditions aim to allow for years of life lost and years lived with a disability to be measured on the same scale. Six disability classes measuring the extent of loss of physical functioning associated with a certain condition were defined. Subsequently, a group of independent experts established a weight, ranging from 0 (perfect health) to 1 (death), for each of the six disability classes [59, 60].

Disability weights perform a similar function to that of quality adjusted life years (QALYs - which measure the years of life gained from the different interventions, weighted according to functional well-being) weights, though for DALYs perfect health is valued at zero and death at one. Premature mortality is measured as years of life lost (YLLs) and these years are implicity weighted at one. Figure 7 presents the relation between QALYs and DALYs [58].

Figure 7 Relation between QALYs and DALYs



DALYs represent a loss and should be minimised, QALYs represent a gain and should be maximised. In the DALY approach, the scale from zero, which indicates perfect health (no disability), to one, which indicates death. In the QALY approach, the scale goes the opposite way: A quality weighting (sometimes called "utility") of 1 indicates perfect health, whereas 0 indicates no quality of life, and is synonymous to death [59].

c. Age weights

Age-weights allows for bias in valuing life years at different ages. The age weights used in the World Bank report rise from birth until age 25 and decline slowly thereafter. The justification for this is to give, for example, more weight to the ages that are materially productive, or to prioritise in favour of the young over the elderly. The age weighting is 4% to give a "greater value to a year of young or middle-aged adult life as compared to a year of life lived by young children or the elderly". The correction constant (C), is required to adjust the total so that it remains the same with or without age weighting [59, 60].

According to the World Health Organization (1994), the formula to calculate those weights is:

$$Age\text{-}Weighting\ Function = Cxe^{-\beta^3}$$

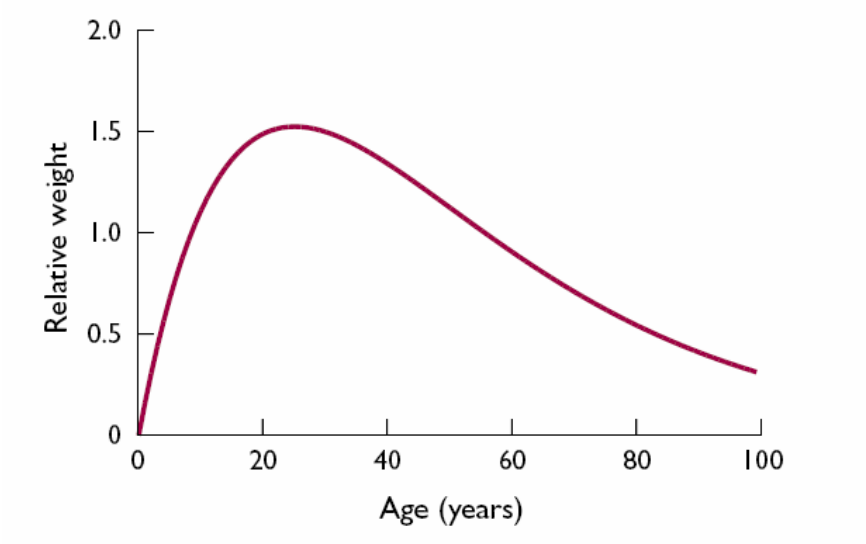
Where:

- C = Constant equal to 0.16243.
- ß = Constant equal to 0.04.
- x = Age. e = Constant equal to 2.71

Figure 8 presents the relative value assigned to each year of life in the calculation of disease burden. The relative value of a life year is below one for children under 10, and for persons more than about 55 years

of age. This implies that in the calculations, a life year lost for children is given less weight than a life year lost for adults below. The adjustments made introduce (explicitly) a bias both against children and the elderly. In a defence of age-weights, Murray (1996) argues that there is a widespread preference for age weighting in most cultures, and, that on average these preferences can be expressed as in the function given [58].

Figure 8 The relative value of a year of life lived at different ages, as incorporated into DALYs (World Bank data) [58]



d. Time preference

Time preference, which is the value of health gains today compared to the value attached to health gains in the future (in standard economic theory, the latter is assumed to be lower than the former). It is standard practice in economic appraisal of projects to use the discount rate to discount benefits in the future. The process of discounting future benefits converts them into net present-value terms; these benefits can then be compared with project costs (also discounted if they are spread over more than one year) to determine cost-effectiveness. The use of a discount rate gives greater weight to current life years than to those in the future. The rationale for this is the belief that individuals are, in the main, myopic and risk averse; and that there is likely to be diminishing marginal utility of future consumption of health. A "low, positive" discount rate (3%) was chosen to capture uncertainty that increases with the time. The formula to discount for time preference is [59, 60]:

$$Discounting\ Function = e^{-r(x-a)}$$

Where:

- r = Discount rate, fixed at 0.03
- x = Age.
- e= Constant equal to 2.71
- a = Onset year.

e. Health is simply added across individuals

That is, two people each losing 10 years of disability-free life are treated as the same loss as one person losing 20 years. One could also weight duration non-linearly, so as to give priority to fewer people suffering for long intervals over more people suffering for shorter intervals.

In summary, the disability-adjusted life year is an indicator of the time lived with a disability and the time lost due to premature mortality. The duration of time lost due to premature mortality is calculated using standard expected years of life lost with model life-tables. The reduction in physical capacity due to morbidity is measured using disability weights. The value of time lived at different ages has been calculated using an exponential function which reflects the dependence of the young and the elderly on the adults. Streams of time have been discounted at 3 percent. Accordingly, the number of DALYs lost due to disability at age "x" can be calculated using the following formula [59]:

$$DALYs(x) = (D)(Cxe^{-\beta x})(e^{-r(x-a)})$$

If the person lives up to the maximum of his life expectancy with disability, we need to add up the total number of DALYs lost from the onset of disability (a) to the age of death (a+L). The following formula can be used [7]:

$$DALY = \_[\frac{Dx0.16243x2.71^{(0.04xa)}}{(0.04+0.03)^2}]x[(2.71)^{(0.04+0.03)xL}x(1+(0.04+0.03)x(L+a))-(1+(0.03+0.04)xa]$$

Where:

L = Years of life left at age "a"

D = Disability weight (ranging from 1 death to 0 for perfect health).

Procedure to calculate DALYs

The following examples illustrate how the formula to calculate DALYs is applied. This section is based on the examples presented in the manual "Selecting an Essential Packages of Health Services Using Cost-Effectiveness Analysis" (Data for Decision Making, 1993: pages 16 - 20). Taking into account that each health problem results in four possible outcomes (death, disability before death, permanent disability, or full recovery), we will calculate the number of DALYs lost for each one of these four scenarios [58, 59, 60].

The example corresponds to the female child mentioned previously who contracts poliomyelitis at age five. As a result she can die; she can live for a period of 5 years and then die; she can be permanently disabled; or she can recover after a period of disability. Let's assume that the horizon of life (total) is 82.95 years, when she got sick at 5 she still had 77.95 years of life left. We will assume that the disability weight in her case is 0.5 [58, 59, 60].

The burden of disease can be estimated using either the incidence or the prevalence of the disease. Incidence is a measure of the rate at which new patients contract a disease. It records the number of new cases in a given time period. For example, if the annual incidence of a disease is given as 0.5% this indicates that in a population of one million 5 000 new cases were recorded that year. Prevalence, on the other hand, is a measure of the number of patients with the disease at any time. A prevalence rate of 6% will indicate that in a population of one million, 60 000 people suffer with the disease. Point prevalence

indicates the prevalence rate at one point in time and life time prevalence gives the number of patients who have ever suffered with the disease.

The burden of disease for a given population expressed in DALYs is [59]:

DALYs = YLLs + YLDs

The years of life lost (YLL) basically correspond to the number of deaths multiplied by the standard life expectancy at the age at which death occurs. The basic formula for YLL (without yet including other social preferences discussed below), is the following for a given cause, age and sex:

YLL = N x L

Where:

- N = number of deaths
- L = standard life expectancy at age of death in years

Because YLL measure the incident stream of lost years of life due to deaths, an incidence perspective is also taken for the calculation of YLD. To estimate YLD for a particular cause in a particular time period, the number of incident cases in that period is multiplied by the average duration of the disease and a weight factor that reflects the severity of the disease on a scale from 0 (perfect health) to 1 (dead). The basic formula for YLD is the following (again, without applying social preferences):

YLD = I x DW x L

Where:

- I = number of incident cases
- DW = disability weight
- L = average duration of the case until remission or death (years)

The number of years with a disability may be measured by:

- Incidence x duration (which is consistent with estimating YLLs)

  or

- Prevalence x one year

<u>Case 1. DALYs lost due to immediate death [59]</u>

We have the following information:

C = 0.16243.

D = 1 (This is because the person dies, in case of disability it is 0.5).

r = 0.03 (Discount rate of 3 percent).

ß = 0.04 (Value fixed by experts, see World Health Organization, 1994).

a = 5 (Year of death).

L = 77.95 (Remaining years of life. It is equal to 82.95 years minus 5 years).

e = 2.71.

Replacing in the DALY formula the above values we have:

$$DALY = \_[\frac{1x0.16243x2.71^{-(0.04x5)}}{(0.04+0.03)^2}]x[(2.71)^{(0.04+0.03)x77.95}x(1+(0.04+0.03)x(77.95+5))-(1+(0.03+0.04)x5]$$

The number of DALYs lost to premature mortality is equal to 35.85.

Case 2. DALYs lost due to death following disability [59]

In this case we have to calculate the number of DALYs lost due to disability and the number of DALYs lost due to premature mortality. To calculate the number of DALYs lost due to disability we have the following information:

C = 0.16243.

D = 0.5

r = 0.03 (Discount rate of 3 percent).

ß = 0.04

a = 5 (Year when the disability starts).

L = 5 (Years with the disability).

e = 2.71.

$$DALY = \_[\frac{0.5x0.16243x2.71^{-(0.04x5)}}{(0.04+0.03)^2}]x[(2.71)^{(0.04+0.03)x5}x(1+(0.04+0.03)x(5+5))-(1+(0.03+0.04)x5]$$

Then the number DALYs lost due to disability is 2.0.

To account for the number of DALYs lost due to premature death (72.95 yrs.), we have the following information:

C = 0.16243.

D = 1.

r = 0.03

ß = 0.04

a = 10 (Year when the person dies).

L = 72.95 (Potential years of life left at time of death).

e = 2.71.

$$DALY = \_[\frac{1x0.16243x2.71^{(0.04x10)}}{(0.04+0.03)^2}]x[(2.71)^{(0.04+0.03)x72.95}\ x(1+(0.04+0.03)x(72.95+10))-(1+(0.03+0.04)x10]$$

The number of DALYs lost due to premature death are 36.85 years. Here there is an important point to take into account. Those 36.85 years are the DALYs calculated at the age of 10; to add them up with the DALYs lost due to disability calculated at age of onset (5 years), we have to convert the 36.85 DALYs calculated at age 10 to their value at the age of onset of the disease, that is at age 5. This can be done using the following formula:

$$DALY\ at\ \ age\ of\ x\ =\ DALY(10)\ e^{-rs}$$

The variables in the formula have been defined previously, except for "s" which is the number of years we have to discount (age of 10 minus age of 5). Applying the formula we have:

$$DALY\ at\ the\ age\ of\ 5 =\ 36.85\ x(2.71)^{(-0.03\ x(10-5))}\ =\ 31.7$$

That is, at the time of the onset of the disease (5), the number of DALYs lost due to premature mortality at age 10 equals the number of DALYs lost at age 10 (36.85) times 0.86, which is 31.7 years.

In summary, the total number of DALYs lost due to a period of disability followed by death equals the number of DALYs lost to disability (ii) plus the number of DALYs lost to premature death (31.7), that is 33.7 DALYs.

Case 3. DALYs lost due to permanent disability

We have the following information:

C = 0.16243.

D = 0.5

r = 0.03 (Discount rate of 3 percent).

ß = 0.04

a = 5 (Year of onset).

L = 77.95 (Remaining years of life. It is equal to 82.95 years minus 5 years).

e = 2.71.

Replacing in the formula the above values we have:

$$DALY = \_[\frac{0.5x0.16243x2.71^{(0.04x5)}}{(0.04+0.03)^2}]x[(2.71)^{(0.04+0.03)x77.95}x(1+(0.04+0.03)x(77.95+5))-(1+(0.03+0.04)x5]$$

The total number of DALYs lost due to permanent disability equals to 17.92.

DALYs lost due to disability followed by complete recovery [59]

We have the following data:

C = 0.16243.

D = 0.5

r = 0.03

ß = 0.04

a = 5.

L = 5 (number of years with disability).

e = 2.71.

Replacing the above values in the DALY formula we have:

$$DALY = \_[\frac{0.5x0.16243x2.71^{(0.04x5)}}{(0.04+0.03)^2}]x[(2.71)^{(0.04+0.03)x5}x(1+(0.04+0.03)x(5+5))-(1+(0.03+0.04)x5]$$

The number of DALYs lost to disability equal to 2.0 years.

To calculate the total number of DALYs lost due to poliomyelitis in a community we have to add the number of DALYs lost by each individual. Let's imagine that in a particular community there is a total of 20 female children who contract poliomyelitis, all of them at age 5, and that 4 of them die immediately, 4 die at age 10 after a 5-year period of disability, 4 of them are permanently disabled, and 4 recover completely after a disability period of 5 years. In this case the total number of DALYs lost in that community due to poliomyelitis equals to:

$$Total\ DALYs\ lost = 5x(35.85) + 5x(33.7) + 5x17.92 + 5x2 = 447.4$$

Data needed to estimate the burden disease

a. Cause of death patterns by age and gender

Cause of death patterns by age and gender are needed to calculate the years of life lost to premature death. Worldwide, only about 30-35 percent of all deaths are captured by vital registration. For the remainder, cause of death structure as a function of the level of mortality can provide broad cause-of-death groups. Such methods are generally unreliable for more specific causes [58]

b. Time lived with a disability measurement

To measure the time lived with a disability in a manner that can be meaningfully compared with the time lost due to premature mortality, there is a need for the following data: (i) age and gender specific

information on the incidence of disease, (ii) the proportion of disease incidence leading to a disabling outcome, (iii) the average age of disability onset, the duration of disability, and (iv) the distribution of disability across the six classes of disability severity. In most cases this data is unavailable and researchers are forced to rely on estimates, many of which are uncertain. There are two important sources of error: (i) some disabilities might have been omitted, which would give a downward bias to our estimates of the years lived with disability, and (ii) the computations do not take into account comorbidity (an individual experiencing multiple illnesses) and biases the results upwards. The fact that individuals can have more than one disability of the same or different classes at the same time cannot be ignored. Presumably, several class 1 disabilities may combine to raise someone's total disability severity to a higher class. However, the effect of three distinct class 1 disability will not be to triple the disability severity weight for the individual. In addition, the magnitude of the overestimation due to comorbidity will be greater if the probabilities of getting different disabilities are dependent on each other (for example, a diabetic has an increased risk of blindness, angina pectoris, amputation, neuropathy and renal failure) [58, 59].

Chemical risks

Currently, there is very little analytical information available concerning the occurrence and fate of chemical pollutants during reuse applications so risk assessment must often be performed based on models. Many approaches to risk assessment of environmental chemicals have been proposed, but fundamental to all of them is the general concept of a risk quotient relating predicted environmental concentrations to predicted no-effect concentrations:

$$Risk\ Quotient = \frac{Predicted\ Environmental\ Concentration\left(PEC\right)}{Predicted\ No\ Effect\ Concentration\ \left(PNEC\right)}$$

The calculation of risk quotients is an essential step for the quantitative evaluation of risks from chemical exposure. In order to demonstrate no risk to health or the environment, the PEC should be less than the PNEC. In other words, the risk quotient should be less than 1.

5.3. Human risk assessment of microorganisms from reclaimed water

5.3.1. General considerations

Pathogens are generating an increasing public concern as the use of reclaimed water has become more prevalent. However, numeric pathogen criteria for reclaimed water should be established before regulation and it should be ensured that criteria are based on sound science, as have been demonstrated through viable risk assessment methods [1]. The unknown and uncertainties continue to raise doubts about the efficacy of using reclaimed water when human exposure to pathogens is high [50]. For pathogen determination, indicator organisms are usually used, but no individual indicator capable for establishing all the biological-derived risk exists.

There are also several suggestions that large-scale irrigation of crops with municipal and industrial wastewater not sufficiently treated could cause damage to crops and injuries to humans because of poor

control of toxic and hazardous contaminants in waste discharges [61]. Health regulations governing wastewater reclamation and reuse impose strict limitations, based on a near "zero risk" alternative approach [62] or on an "acceptable risk level" approach [17].

In this respect, it has been a long time controversy on the strictness of the microbiological criteria fixing the quality of reclaimed water. Although the numerical values of the indicated parameters are important, the uncertainties associated to the non-determined microbiological parameters (namely viruses, protozoa…) need to be assessed and taken into account for further work on water reclamation and reuse safety. Comprehensive indicators or model organisms are strongly needed for better defining the risk associated to reclaimed water pathogens content. In any case, it appears that current sampling procedures are capable to define only the quality of the water in a specific moment, corresponding to the time of sampling. Regarding sampling procedures and the subsequent analysis, there is the possibility to assess the quality reflecting a wider time span. Nevertheless, using classical analytical procedures, especially microbiological ones, it is impossible to establish if there is a point problem, i.e. a really low level quality (or a point microbiological contamination) of water during few minutes that can compromise aquatic life for a long time or cause problems for consumers [63].

The types of enteric pathogens that can be found in contaminated water include viruses, bacteria, protozoa and helminths (Table 21). The risk of water-borne infection from any of these pathogens can be reliant on a range of factors including pathogen numbers and dispersion in water, the infective dose required and the susceptibility of an exposed population, the chance of faecal contamination of the water and amount of treatment undertaken before potential exposure to the water [15]. A range of other factors also exist such as the potential for asymptomatic carriers of pathogens, person-to-person spread of disease within a community and the socio-economic status of the community(s) at risk [15].

5.3.2. Pathogen fate during irrigation applications of reclaimed water

Increasing efficiencies in crop management and the continuing rise in crop yields has increased demands on water resources for irrigation purposes. Effluents are reused for irrigation purposes in many countries around the world on all of the populated continents [62]. A number of these countries have developed guidelines that give quality criteria and advice on how effluents should be reused for irrigation purposes. Examples of these guidelines are summarised in the USEPA guidelines for water reuse manual [62].

Table 21 Infectious dose of common pathogens detected in reclaimed water

Pathogens	Predominant Illness	Infectious Dose ($ID_{50}^{‡}$) [1]	Amount shed by infected individual (particles/ gram faeces)[¶]
Enterovirus			
Poliovirus		1-10^{4} *	10^{3} - 10^{6} *
Coxsackievirus		10^{3}	10^{2} - 10^{5} *
Echovirus			
Enterovirus			
Hepatitis A virus	Hepatitis		
Hepatitis E virus	Hepatitis		
Astrovirus	Gastroenteritis		
Norwalk virus	Gastroenteritis		10^{5}-10^{6} [2]
Rotavirus	Gastroenteritis	6 [4]	Up to 10^{10} [3]
Adenovirus			

Vibrio cholerae	Cholera	9 – 3x10^{8} [§]	
Salmonella spp.			
S. typhi	Typhoid	8x10^{5}	
Campylobacter jejuni	gastroenteritis	9x10^{2}	
Shigella dysenteriae	Dysentery	7x10^{2}	
Yersinia enterocolitica			
Giardia lamblia	Gastroenteritis (Giardiasis)	35	10^{6} [4]
Cryptosporidium parvum	Gastroenteritis (Cryptoriadiasis)	173	10^{6} - 10^{7} [4]
Entamoeba histolytica			
Entamoeba coli		204	
Ascaris lumbricoides	Round Worm		
Necator americanus	Hook Worm		
Trichuris trichiura	Whip Worm		

[‡] ID_{50} = dose at which 50% of an exposed population exhibit a response.
[§] ID_{50} for *Vibrio cholerae* varied depending on the strain being suspended in pH-buffered solution (9) or water (10^{8}).
[*] ID_{50} for Poliovirus varied depending on strain tested.
[¶] Values given are estimated possible maximum [64]
[1] Teunis *et al*. (1996) [65] [2] Metcalf *et al.* (1995) [66]
[3] Gerba *et al.* (1996) [67] [4] Ward *et al.* (1986) [68]

In many regions of the world, irrigation demand has placed severe strains on existing resources with resulting environmental impacts. An example is in Perth, Australia where the major drinking water aquifer is being depleted by a combination of extractions for public drinking water source, horticultural irrigation and the positioning of pine plantations of large area of the mound [69]. Saudi Arabia is another example of a country with demonstrated impacts on natural water resources due to increasing demands on groundwater by the agricultural sector [70]. Large decreases in groundwater levels (up to 200 m in some places) have been observed due to overextraction. In a number of countries, for example Australia, this has been compounded by prolonged periods of drought or seasons of low rainfall. In addition, predicted climate impacts from global warming also point to further stresses on water resources, thus reducing the amount of water available for both irrigation and the environment.

There are a range of mechanisms that can be used to reduce the pressure on fresh water resources for irrigation use. One possible mechanism is the reclamation of wastewaters and drainage water that can be used in the place of other fresh water sources for irrigation. Types of wastewaters used for reclamation include treated and untreated sewage effluent [71, 50, 5], storm water runoff [72, 50], domestic greywater [73], and industrial wastewater [50, 74]. For the purposes of this paper all of these water types are considered effluents which have the capacity to be reused. These different water types, however, can vary in quality and in the contaminants that could be potentially present. The quality and contaminants present will also impact on the level of treatment required, which in turn impacts on the economic viability of reusing the various wastewaters.

Risks from pathogens in recycled water used for irrigation

There have been a number of risk factors identified for using reclaimed water for purposes such as agricultural irrigation. The major health risk factor is microbial pathogens although the risk can vary in severity depending on the potential for human, animal or environmental contact. As with any wastewater the risk of water-borne infection from microbial pathogens is reliant on a range of factors including pathogen numbers and dispersion in water, the infective dose required and the susceptibility of an

exposed population, the chance of faecal contamination of the water and amount of treatment undertaken before potential exposure to the water [15].

Viruses

Viruses are all obligate intercellular parasites that require the infection of host cells of a suitable host and then force the host cell to produce multiple copies of the virus [75]. Most enteric viruses have a narrow host range meaning that most viruses of interest in reclaimed water only infect humans [15]. This means that only human faecal contamination of water need to be considered a concern for viral infection of humans. Conversely, water borne human viruses are rarely a problem for other animals.

Bacteria

Bacteria are the most common of the microbial pathogens found in reclaimed waters [76]. There is a wide range of bacterial pathogens and opportunistic pathogens which can be detected in wastewater. Many of the bacterial pathogens are enteric in origin; however, bacterial pathogens which cause non-enteric illnesses (e.g., *Legionella* spp., *Mycobacterium* spp., and *Leptospira*) have also been detected in wastewaters [77, 78, 79]. In reality, these introduced pathogens are prevented from doing so by environmental pressures [80]. Like other enteric pathogens, a common mode of transmission is via contaminated water and food and by direct person to person contact [15]. A number of these bacterial pathogens can also infect, or be carried by, wild and domestic animals.

Protozoa

As previously mentioned, outside of an infected host protozoan parasites persist as dormant stages known as cysts or oocysts. Infection from protozoan pathogens can occur after consumption of food or water contaminated with the (oo)cysts or through person to person contact [81]; thus unprocessed and raw food crops could be a potential infection risk from these pathogens. Like with the bacterial pathogens, various domestic and wild animals can be a source of these protozoa and be infected by them.

Helminths

The presence of microbial pathogens in reclaimed water, particularly when sourced from sewage effluent, is arguably the major concern for health regulators, farmers and the general public. One of the major sources of helminth infections around the world is the use of raw or partially-treated sewage effluent and sludge for the irrigation of food crops [17].

Observed health concerns from reclaimed water irrigation

A major observed risk from pathogens in reclaimed water used for irrigation purposes is that from helminths in developing nations where sewage effluents are used with minimal or no treatment prior to reuse. In Mexico, farmer workers and their children that work in fields irrigated with untreated sewage effluent have been found to have a greater prevalence of round worm infection than the general population [82]. The authors found that infection rates, particularly for adults, decreased with treatment of the sewage effluent with infection rates decreasing at a rate that could be linked to the level of treatment. Peasey *et al.* (2000) also found that the consumption of raw vegetables (such as carrots, cauliflower, lettuce and cucumber) irrigated with partially treated sewage effluent did not confer any greater prevalence of infection from any age group to the general population [82].

Once the reclaimed water is treated, the risks have been observed to fall dramatically. An epidemiological study relating to agricultural irrigation using reclaimed water that met the WHO guidelines of $\leq 10^3$ faecal coliform/100mL in Asian, South American and European countries had determined that the risk from eating raw vegetables (10^{-5} to 10^{-8}) is significantly less than the USEPA acceptable risk for viral infection from consuming drinking water (10^{-4}) [83]. In comparison, Petterson *et al.* (2001) examined the potential health risk from enteric viruses due to the consumption of salad vegetables (as example lettuce which was used in this study) that had been irrigated with reclaimed water [84]. They found through the use of a simulation exercise that there was a potential risk for human health.

Sheikh *et al.* (1987) examined the influence of treatment levels on the quality of tertiary treated sewage effluent used for the irrigation of salad crops in the Monterey County area in California [85]. The treatment levels tested were filtered secondary effluent and coagulated, flocculated, settled, and filtered secondary effluent (also known as Title-22 water). These two reclaimed waters were compared to local groundwater which was used as a control. The study determined that there were no significant differences in coliform levels that could be linked to any of the three water types. In addition, no pathogenic bacteria (*Salmonella* or *Shigella*), enteric viruses or helminth parasites could be detected in any of the samples. In addition to the monitoring program, a seeding experiment was undertaken where the vaccine strain of poliovirus was irrigated onto plants in the field. Virus decay on the plants was observed to be 2 log in less than 8 days.

Public concern

A major issue relating to all water reuse schemes is public opinion. Communities tend to be favourable in general to reusing water, in fact at times demanding that it is undertaken (e.g, the Western Australia State Water Strategy [69]). Most people, however, tend to become less favourable towards reclaimed water as it physically comes closer to them. In other words, they are very supportive of the irrigation of public open spaces in some ill defined region, but baulk at the use of reclaimed water in the household or when the chance of personal physical contact increases [86, 87].

The amount of public unease about water reuse also depends on the type of reclaimed water and treatment levels. For example, people have much less concern about using untreated captured stormwater than they have about highly treated sewage effluent [86]. While the actual physical risk from reclaimed effluent can be similar or less than that of untreated stormwater, the public perception (the "Yuck-factor") can lead to a belief within the community of a greater risk from the effluent [88, 89].

Public and commercial concern does exist regarding pathogens through the use of reclaimed water and biosolids on cereal crops [90]. It should be expected, however, that if there is a reduction of risk for the consumption of raw vegetables irrigated with partially treated effluent, then it can be surmised that grain crops irrigated with reclaimed waters would have even less risk from microbial pathogens. Even more specifically, grains are commonly processed further before they are consumed by humans which decreases the human health risk even further. Little is known, however, about the risk to domestic grazing animals that may be fed this grain unprocessed as a stock feed source. It is acknowledged that more scientific research is needed to confirm this lack of confirmatory information and funding is being provided to undertake this research [90].

Treatment Processes

One mechanism that can be used to minimise the risk from microbial pathogens is through the treatment and disinfection of the recycled water before it is used for irrigation purposes. As detailed in Section 2.1 of this paper, there are various types of treatment processes that can be used based on the treatment efficiency required and the desired end use of this recycled water. The best quality of recycled water is obtained through the use of dual membrane tertiary treatment. This is a very expensive process, unlikely to be economically viable except perhaps for very high value crops. Due to the importance of keeping treatment costs to a minimum, the use of the water is often treated to a "fit-for-purpose" level which may mean a significantly lower level of treatment than the dual membrane tertiary treatment "gold standard". As observed in Section 2.2 of this paper as treatment levels decrease the potential risk from microbial pathogens increases [15].

Treatment levels required to produce fit-for-purpose recycled water varies depending on a range of factors such as the potential for human contact with the irrigated water, but in particular on the end use of the crop irrigated with the recycled water. For example, salad crops and other human foods that are eaten raw would require the use of recycled water that has been treated to a much higher level than water that is used to treat a fodder crop that is used for animal feed. The need for a variety of treatment levels to suit different crops has lead to a number of nations producing guidelines (eg, US EPA and Australian NH&MR) which state the level of treatment and disinfection that is needed for different crop types [75].

As treatment levels are decreased the potential for microbial pathogens to be present in recycled water increases with pathogens even being detected in water that has been treated using non-dual membrane tertiary treatment methods [91]. As mentioned above, this may not be of a concern where the crop is not used for human consumption or where the recycled water does not come in contact with the produce (eg, drip irrigation of fruit trees), however, there are situations where the risks from microbial pathogens being present on an irrigated crop needs to be managed.

Environmental Pressures

Where there is a concern that there may be a chance of human or animal infection through contact or consumption of the crop, then environmental processes are sometimes used as a barrier to assist in the removal of pathogens from the crop. These environmental pressures utilised most commonly include non-irrigation and drying times while, other processes can also have an influence on the persistence of pathogens in a crop. The environmental processes that influence pathogen survival in and on crops irrigated with recycled water can include desiccation and moisture levels, temperature, sunlight and adsorption to plant and soil materials.

The use of recycled water for the irrigation of crops has benefits in using a resource that would otherwise be discarded and wasted. Using recycled water also reduces the pressures on the environment by reducing the use of environmental waters. There are factors that need to be considered, including the presence of pathogens and chemical contaminants as well as salinity and impacts on soil structure. These can all be controlled through treatment and effective farm management practices. Ongoing research and development will also improve and increase the use of recycled water for irrigation purposes as well as increasing public confidence.

5.3.3. Human risk assessment of microbiological components from reclaimed water

Hazard identification

While the risk of infection from waterborne pathogens is not well known, the list of known waterborne human pathogens continues to expand. Table 22 presents the most common wastewater associated disease-causing microbiological agents

Table 22 Most common wastewater associated disease-causing microbiological agents [modified from 15 and 16]

	Agent	Disease
Bacteria	*Salmonella typhimurium*	Salmonellosis
	Salmonella typhosa	Typhoid fever
	Salmonella paratyphi	Paratyphoid fever
	Shigella spp	Bacillary dysentery
	Vibrio cholera	Cholera
	Vibrio parahaemolyticus	Gastroenteritis from seafood
	Mycobacterium tuberculosis	Tuberculosis
	Campilobacter jejuni	Diarrhoea
	Pathogenic *Escherichia coli*	Diarrhoea
	Enterobacter aerogenes	Nosocomial infections
	Klebsiella pneumoniae	Pneumonia
	Proteus mirabilis	Urinary tract infections
	Serratia marcescens	Opportunistic inflections
	Haemophilus influenzae	Meningitis, other pediatric diseases
	Coxiella brunetti	Q fever
	Chlamydia psittaci	Psittacosis
	Mycoplasma pneumoniae	Primary, atypical pneumonia
	Staphylococcus aureus	Food intoxication, skin infection
	Streptococcus pyogenes	Pharyngitis, skin infections
	Streptococcus faecalis	Opportunistic infection
	Bacillus anthracis	Anthrax
Bacteria	**Agent**	**Disease**
	Clostridium botulinum	Botulism, food intoxications
	Clostridium perfringens	Gas gangrene, food intoxications
	Clostridium difficile	Gastroenteritis, colitis
	Listeria monocytogenes	Meningitis
	Corynebacterium diphtheria	Diphtheria
	Actinomycetes israelli	Actinomycosis
	Mycobacterium tuberculosis	Tuberculosis
	Mycobacterium avium	Pulmonary disease, disseminated disease in immunocompromised
	Pseudomonas aeruginosa	Wound, burn, urinary tract infections
	Brucella abortus	Brucellosis
	Brucella melitensis	Brucellosis
	Brucella suis	Brucellosis
	Bordetella pertussis	Pertussis (whooping cough)
	Francisella tularensis	Tularemia
	Yersinia enterocolitica	Diarrhoea and septicemia
	Legionella pneumophila	Legionellosis
	Leptospira interrogans	Leptospirosis
	Leptospira icterohaemorrhagiae	Leptospirosis
Viruses	Poliovirus	Poliomyelitis
	Picornaviruses (animal viruses)	Paralysis, common cold, mycocarditis
	Togaviruses (animal viruses)	Encephalitis, yellow fever
	Paramyxoviruses, rhabdoviruses (animal viruses)	Measles, mumps, rabies
	Orthomyxoviruses, arenaviruses (animal viruses)	Influenza, haemorrhagic fevers

	Hepatitis A virus	Infectious hepatitis
	Hepatitis E virus	Hepatitis
	Rotavirus	Diarrhoea/gastroenteritis
	Retroviruses (animal viruses)	Leukemia, tumors, AIDs
	Adenoviruses	Respiratory disease
	Herpesviruses (animal viruses)	Oral and genital herpes, chickenpox, shingles, mononucleosis
	Poxviruses (animal viruses)	Smallpox, cowpox
	Papovaviruses (animal viruses)	Warts
	Parvoviruses (animal viruses)	Roseola in children, aggravates sickle cell anemia
	Norwalk agent	Gastroenteritis
Protozoa	*Entamoeba histolytica*	Ameabiasis
	Naegleria fowleri	Primary meningoencephalitis
	Acanthamoeba spp.	Meningoencephalitis, eyes damages, breath and skin damages
	Giardia duodenalis	Diarrhoea
	Cryptosporidium parvum	Diarrhoea
	Isospora spp.	Diarrhoea
	Balantidium coli	Diarrhoea, dysentery
	Cyclospora cayetanensis	Intestinal illness
	Toxoplasma gondii	Toxoplasmosis
	Enterocytozoon bieneusi	Diarrhoea
	Encephalitzoon cuniculi	Spread illness of lung and liver
	Encephalitzoon intestinalis	Spread illness of lung and liver
	Filum *microspora*	Microsporiadosis
Helminths	*Schistosoma haematobium (T)*	Esquistosomiasis
	Schistosoma mansoni (T)	Esquistosomiasis
	Schistosoma japonicum (T)	Esquistosomiasis
	Ascaris lumbricoides (N)	Ascariasis
	Ancyclostoma duodenale (N)	Anaemia, intestinal illnesses
	Necator americanus (N)	Anaemia, intestinal illnesses
	Clorchis spp. (T)	Clonorquiasis
	Taenia spp (N)	Teniasis
	Enterbius vermicularis (N)	Enterobiasis
	Hymenolepis nana (C)	Himenolepiasis
	Trichuris trichiura (N)	Tricuriasis
	Strongyloides stercolaris (N)	Diarrhoea, abdominal pain
	Toxocara canis (N)	Fever, intestinal pain
	Toxocara cati (N)	Fever, intestinal pain

There are different types of epidemiological studies; ecological studies are the easiest to conduct, but there are other types of studies: cross-sectional, case-control, and cohort studies from which to choose. The type of study undertaken depends on whether one is looking from the point of disease to a potential cause backwards (retrospective) or is starting at some point collecting data as time moves forward (prospective) [92].

Dose-response

Once etiologic agent has been identified and human exposure to the pathogen is known to cause infection and disease, the next step is to determine how exposure to different concentrations of the pathogens might elicit a response in humans. Current pathogen dose-response data is limited. Ideally, a dose-response model should be able to differentiate between infection and illness, because although the terms may be used interchangeably by some, the terms have very distinct meanings. The current models used for microbial risk assessment only predict the risk of infection, not the risk of contracting the disease [92]. See also before point 3.3.

a. Log-normal model

It is assumed that each individual in a population has a minimum infective dose and that this minimum infective dose varies between individuals. It is further assumed that the distribution of the minimum

infective dose, among the individuals in the population follows a log-normal distribution. The fraction of the population that is predicted to respond to a given dose is given by the following equations [92]:

$$P^* = \frac{1}{\sqrt{2\pi}} \int_{-\infty}^{z} \exp\left(-\frac{z^2}{2}\right) dz$$

$$Z = \frac{\ln(D-\mu)}{\sigma}$$

Where

D = the number of pathogens (dose)

μ = average logarithm

σ = log standard deviation

P* = probability that a single individual exposed to a single dose, N, will become infected

b. Exponential and beta-Poisson models

They are described as single-hit models. In the exponential model, the calculation to determine the probability of infection is the product of two independent events. First, the probability that a single dose will include *j* organisms and second, the probability that *k* organisms will survive to reach a site at which infection can be initiated. When the host-pathogen interaction is assumed to be constant, then one pathogen equals one infection. If the distribution of microorganisms in the environment follows a Poisson distribution and the probability of survival to the target organ can be derived from binomial theory, then the probability is given by [92]:

$$P^* = 1 - \exp(-rD)$$

Where:

D = number of pathogens (dose)

r = fraction of pathogens that survives to produce an infection

Unfortunately, this derivation assumes the probability of survival is constant, *r*. While this single hit, constant survival model fits the data from in vitro studies, it does not fit all dose-response data.

In reality, human response to a given dose probably follows some sort of distribution as no individual in a population can be singled out as being representative of the whole group. Each person in the population will react to disease differently, i.e., the state of the person's immune system, their immunological condition at the time of exposure, and their genetic makeup. It is more likely that a range or distribution of responses from the general population better accounts for reaction to pathogen exposure [92].

If *r*, the fraction of organisms that survives to produce an infection, is replaced by a probability distribution, such as a beta distribution, then the following beta-Poisson model results [92]:

$$P^* = 1 - \left(1 + \left(\frac{D}{\beta}\right)\right)^{-\alpha}$$

Where

D = number of organisms (dose)

α, β = parameters to fit the dose-response curve

P* can be interpreted in at least two different ways:
If a person is exposed to the identical dose of pathogens *m* times, then he or she may be infected $m \cdot P^*$ times.
If *N* members of a human population are exposed to the identical dose of pathogens, then $N \cdot P^*$ of persons may be infected.

Exposure assessment

The overall probability of infection from exposure to Reclaimed water is a function of the frequency of occurrence in wastewater, survival through the treatment process, survival in Reclaimed water, human exposure, and dose-response. Expressed mathematically, the overall probability of infection Pi, depends on a series of independent events represented by the equation [92]:
$Pi = Pw \cdot Pt \cdot Pe \cdot Pd$
Where
Pi = probability of pathogen presence in the source water
Pt = probability of pathogen surviving the treatment process train
Pe = probability of pathogen surviving after exposure to the environment
Pd = probability of human infection from a given dose (calculated form dose-response models)

Risk characterization

The risk infection represent a quantitative estimate of the risk of infection. These are singular values (deterministic models) with no estimate of the uncertainty associated with each of the estimate. Risk managers should understand that these elements are capable of producing values that represent an absolute risk only when the models can predict with absolute certainty the fate of the pathogen in each step of the risk assessment, otherwise the risk assessment only provide a relative measure of risk [92].

5.4. Human risk assessment of organic trace contaminants from reclaimed water

Emerging chemicals of concern, such as the endocrine disrupting chemicals (EDCs) and pharmaceutically active compounds (PhACs), are not specifically regulated by any current wastewater reuse guidelines of which we are aware. This situation is now on the very verge of change as scientists and regulators are grappling with how best to address the issues presented by a wide range of individual chemical contaminants.

5.4.1. General considerations

As example for highly biological and human active compounds, Webb *et al.* (2003) calculated the human exposure to pharmaceuticals via drinking water [93]. The daily drinking water intake and the lifetime (i.e. 70 years) intake of 2 L/day of water were derived from (maximum) reported values of pharmaceuticals in drinking water in Germany. Only few pharmaceuticals occur in drinking water at levels above their limit of detection. Over 90 % of the compounds assessed had lifetime intake values less than the daily therapeutical dose. The concentration of these compounds that were not detected in the drinking water

was estimated to be the limits of detection. The lifetime intake levels of four compounds (Clenbuterol, Salbutamol, Terbutalin and Ethinylestradiol) were higher than the therapeutical dose, but none of these were detected in the drinking water above the LOD of 5 – 10 ng/L. In general, the estimated indirect exposures are low and well below the dose that would cause a pharmacological effect. But this calculation does not take mixture effects into account. Biological effect studies have proven that even a mixture of biologically active compounds below their LOEC cause a higher effect than the single compounds separately [94, 95].

Table 23 Human estimated daily intake of pharmaceuticals of STP effluents compared to the daily therapeutic doses

Organic compound	maximum effluent concentration [µg/L]	daily intake via reclaimed water [mg/day]	Daily therapeutic dose [mg/day]	% of daily therapeutic dose
Ethinylestradiol	**4.E-03**	**8.E-03**	**0.01**	**80**
Mefenamic	1.1	2.2	1500	0.1
Paracetamol	35	70.0	1000	7
Salicylic acid	5.E-04	1.E-03	3000	3.E-05
Ibuprofen	3.8	7.6	1200	0.6
Naproxen	0.1	0.2	375	0.1
Diclofenac	**2.5**	**5.0**	**25**	**20**
Trimethoprim	0.83	1.7	200	0.8
Sulfamethoxazole	0.62	1.2	800	0.2
Clarithromycin	0.21	0.4	500	0.1
Roxithromycin	0.54	1.1	150	0.7
Erythromycin	0.18	0.4	1000	4.E-02
Carbamazepine	2.1	4.2	400	1.1
Clofibric acid	0.48	1.0	500	0.2
Gemfibrozil	0.2	0.4	1200	3.E-02
Fenofibric acid	0.13	0.3	100	0.3
Atenolol	0.36	0.7	50	1.4
Sotalol	1.32	2.6	80	3.3
Propranolol	0.18	0.4	80	0.5
Metoprolol	1.7	3.4	100	3.4
Iopamidol	1.1	2.2	20000	1.E-02
Iopromide	5.2	10.4	20000	0.1
Diatrizoate	5.7	11.4	20000	0.1
Iomeprol	2.3	4.6		
Caffeine	0.22	0.4	100	0.4

According to Webb *et al.* (2003) we calculated the human daily input of organic trace pollutants of STP effluent, as a general source for wastewater reclamation. As a worst case calculation we proposed that the effluent will undergo the reclamation process without any further biological degradation of organic compounds and supposed to be reused for drinking water production. Furthermore, only the maximum detected values of the compounds from the literature (see Aquarec WP2 Deliverable 1 Report) were used. For the compounds that were not detected in the STP effluent the limit of detection concentration was estimated. Assuming 2 L of daily reclaimed drinking water intake, we estimated the daily human input of a wide range of organic micropollutants like pharmaceuticals, endocrine disrupters, pesticides, organic solvents as well as inorganic species like heavy metals. We compared it with the daily therapeutic dose for the pharmaceuticals, derived from **www.rxlist.com** and [93], with the ADI value (RfD value,

respectively) for the other compounds [96]. In addition, we calculated the percentage of the therapeutic dose or ADI that would be reached by the daily intake of 2 L.
It seems to be not hazardous, concerning the content of organic micropollutants in STP effluents, if the effluent will be reused for drinking water production (see Table 23). Ethinylestradiol and diclofenac seem to be the most risky candidates relating to the percentages of the daily therapeutic dose intake by the hypothetical reclaimed effluent water. Nevertheless, in this calculation no biodegradation is included. Too little is known of the chronical and mixture effects of all organic micropollutants. More effort is required to get further human and biological effect data.

5.4.2. Reclaimed water irrigation – theoretical risk model

Since one of the most important reuse applications concerning wastewater reclamation and human risk is the irrigation of agricultural land we have focused on such scenario in this study. A highly simplified illustrative analysis was undertaken for a human risk assessment of three model chemicals (chloroform, 1,1,2-trichloroethane and pyrene) in the irrigation wastewater.

Hazard identification

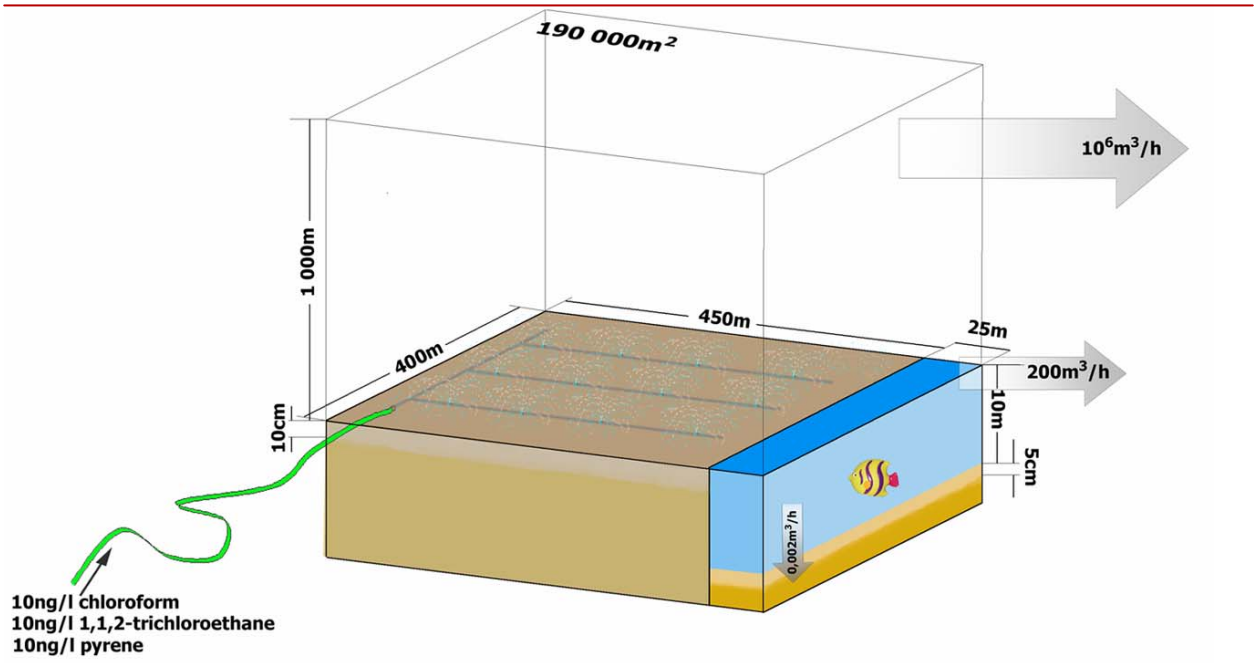
Chlorinated solvents like trichloroethane are widely used in industrial processes and therefore they can be found in the aqueous environment in the lower ng/L range [35]. Chloroform is also generated during water disinfection by the reaction of chlorine with humic compounds. Further production of chloroform can be initiated by reactions of strong oxidising agents and organic matter in the presence of chloride ions. Such conditions may arise from industrial effluents including those from pulp and paper manufacturing or the pharmaceutical industry [44]. The observed average chloroform concentration of US drinking water is approximately 13 µg/L [44]. Polycyclic aromatic hydrocarbons (PAH) such as pyrene are ubiquitous in the environment, produced mainly by incomplete combustion of organic matter such as wood, oil or coal. They are often one of the predominant contaminants in sediments and soils at concentrations in the upper µg/kg range [97]. We selected these well-known compounds for evaluating the risk models because (1) their toxic effects are known, (2) our estimations can be compared with observed concentrations in the environment and (3) they include a range of the important physical-chemical properties required by the models. The two chlorinated compounds are similarly lipophilic (Log K_{ow}), but chloroform has a greater water solubility and is more volatile than 1,1,2-trichloroethane. In contrast, pyrene is many orders of magnitude less volatile and largely insoluble in water.

Exposure Assessment: Determine Predicted environmental concentrations by a fugacity model

One approach to establishing PEC is the use of fugacity modelling techniques. In the absence of comprehensive quantitative analytical data, fugacity calculations can provide useful estimations of overall fate and behaviour of individual chemical components. Although such techniques are limited by the availability of high-quality site-specific input data, they represent a valuable tool for the broad evaluation of the distribution and removal of chemicals during water treatment, storage and reuse applications.
Environmental fugacity-based models have largely been developed over the last 25 years [98, 99]. These models rely on defined environmental compartments such as air, water, soil, etc. By equating the fugacity or “escaping tendency” of a chemical in each of the compartments, an environmental distribution is

calculated. Variable removal processes such as advection or degradation provide determinations of final concentrations among the defined compartments.

Figure 9 Hypothetical field irrigation scenario



Here we provide an illustration of how fugacity modelling may be used to predict the fate of organic chemicals in water reuse applications. Various "Levels" of fugacity models have been described, ranging in complexity from Level 1 to Level 4. It is considered that a Level 3 model is most suitable for the current application. The Level 3 model provides predictions of steady-state, non-equilibrium concentrations and distributions of environmental contaminants.

The hypothetical irrigated property consisted of a rectangular field of dimensions 450 m x 400 m (Figure 9). A 25 m wide waterway ran along one of the two 400 m boundaries, thus the overall area of the property including the irrigated soil and the waterway was 190,000 m^2. The average depth of the waterway was 10 m. The water compartment was composed of water, suspended sediment (2% organic carbon) and aquatic biota (5% lipid). The atmospheric depth was modelled as 1000 m and was composed of air and aerosols. The soil compartment was composed of air, water and solids comprising 2% organic carbon. Soil was presumed to be well mixed to a depth of 10 cm. The sediment on the floor of the waterway comprised water and solids (4% organic carbon) and was mixed to a depth of 5 cm. The organic fractions and densities of all components were selected as recommended by Mackay et al. [99]. Advective flow of air out of the modelled region was set at 10^6 m^3/h. Advective removal by the waterway was 200 m^3/h. Burial by sediment was also treated as an advective loss and was modelled at 0.002 m^3/h. Full details of the environmental parameters are provided in Table 24.

Table 24 Compartment dimensions and properties for field irrigation

Compartment		Depth (m)	Area (m^2)	Volume (m^3)	Organic fraction	Density (kg/m^3)	Advective flow (m^3/h)
Air	Total compartment	1000	2×10^5	2×10^8	-	1.2	10^6
	Air			2×10^8	-	1.2	10^6
	Aerosol			2×10^{-3}	-	2000	10^6
Water	Total compartment	10	10^4	10^5	-	1000	200
	Water			10^5	-	1000	200
	Suspended sediment			10	0.02	1500	200
	Aquatic biota			0.2	0.05 lipid	1000	200
Soil	Total compartment	0.1	18×10^4	18×10^3	-	1500	-
	Air			3.6×10^3	-	1.2	-
	Water			5.4×10^3	-	1000	-
	Solid			9×10^3	0.02	2400	-
Sediment	Total compartment	0.05	10^4	500	-	1280	0.002
	Water			400	-	1000	0.002
	Solid			100	0.04	2400	0.002

The field was assumed to be irrigated at an average of 700 mm per year over the entire surface area. The three contaminants were each presumed to be present in the treated effluent at a concentration of 10 ng/L. Therefore the total application of each chemical to the field was 1.26 g per year. The physical properties of the contaminants are provided in Table 25. These data are all extracted from the experimental database packaged with the EPI Suite software package, except for the BIOWIN and AOPWIN values which are predicted values from the same software [100].

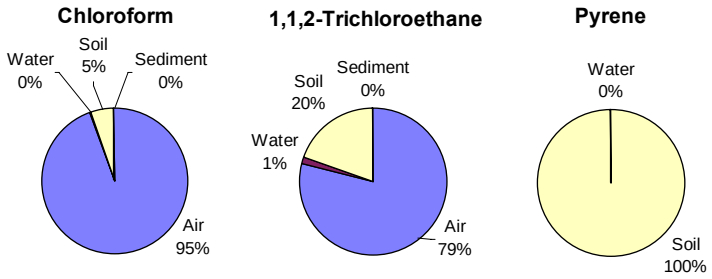
Environmental biodegradation rates typically represent the greatest obstacle to reliable modelling. This is due to a general lack of information regarding degradation of specific compounds in various environmental media, as well as the extreme variation that can result from seemingly subtle environmental variations. Consequently, there are no highly reliable methods of predicting environmental biodegradation. A "best available" method has previously been described for extrapolating predicted "BIOWIN4" data from the EPI Suite for use in fugacity modelling [101]. We have used an analogous relationship to determine approximate biodegradation half lives in water: Half-life in water (h) = $10^{(6 - \text{BIOWIN4 value})}$. Following the typical variations of degradation rates among phases described by Mackay et al [99], average half-lives in soil and sediment were approximated by multiplying the half-life in water by factors of 2 and 10 respectively. Half-lives in air were estimated by AOPWIN, which is also packaged as a component of the EPI Suite. Data derived by these processes should be considered only as reasonable estimates of the order-of-magnitudes. Level 3 fugacity calculations were undertaken as previously described [99]. Key summary data are provided graphically for chloroform, 1,1,2-trichloroethane and pyrene.

Table 25 Properties of modelled contaminants from EPI Suite [100]

Chemical Name:	chloroform	1,1,2-trichloroethane	pyrene
Molecular Weight ($g.mol^{-1}$):	119.38	133.41	202.26
Aqueous Solubility ($g.m^{-3}$):	7950	4590	0.135
Log K_{OW} :	1.97	1.89	4.88
melting point (^oC):	-63.6	-36.6	151.2
vapour pressure (Pa):	2.6×10^4	3.1×10^3	6.0×10^{-4}
BIOWIN4	3.3737	3.3534	2.8537
AOPWIN half-life (h)	1206	561	3

Figure 10 shows the relative distributions of each of the contaminants among the modelled phases. Chloroform is a highly volatile chemical and was thus predicted to partition significantly (95%) to the air phase. The vast majority of the remainder of this contaminant is partitioned to the soil phase. 1,1,2-trichloroethane is less volatile than chloroform and only 79% of the steady-state mass was predicted in the air phase. 20% was in soil and 1% in water. The most extreme results were predicted for pyrene which is non-volatile and highly hydrophobic. Pyrene was predicted to be partitioned exclusively to soil.

Figure 10 Relative distribution of three contaminants among modelled phases



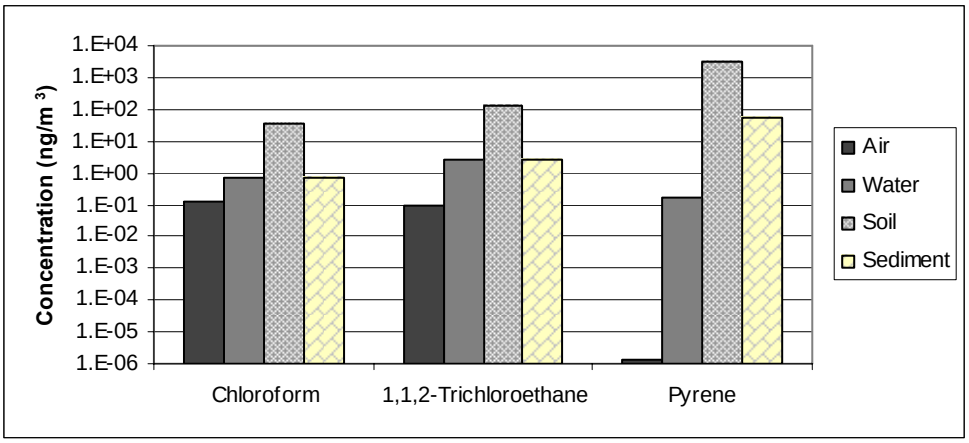
As shown in Figure 10, the predicted relative amount of 1,1,2-trichloroethane in soil is about one fifth of the relative amount of pyrene in soil. However, the predicted *absolute* steady-state mass of 1,1,2-trichloroethane in soil is only about 4% the predicted absolute steady-state mass of pyrene in soil. This is because pyrene is not removed from the overall system as efficiently as 1,1,2-trichloroethane or chloroform.

There are seven possible removal mechanisms for each of the chemicals: degradation in each of the four phases and advection from water, air or sediment. Pyrene was predicted to be the least biodegradable of the contaminants, but it was also the least susceptible to advection since it is partitioned exclusively to soil, from which no advection occurs. Furthermore, although pyrene was predicted to have a relatively short degradation half life in air, this was not significant since pyrene was not appreciably partitioned to air. It was a combination of these effects that lead to the relative accumulation of pyrene.

Figure 15 is presented in Log scale to clearly display the extreme variations in steady-state concentrations of the contaminants among the principal phases of air, water, soil and sediment. Given the considerable partitioning of chloroform to air (Figure 10), it may at first be surprising to note the relatively low concentration of this compound in this phase. The explanation is that the volume of the air phase (2×10^8 m^3) is numerous orders of magnitude greater than the volume of any of the other phases. Hence, air can accommodate substantially more of a compound without building up high concentrations. The opposite

extreme of this effect was observed for sediment. Although none of the contaminants were predicted to partition significantly to sediment (Figure 10), all three were predicted in sediment at moderate concentrations. Again, the explanation is that even when sediment is heavily concentrated with contaminant, the small volume of sediment requires only a relatively minor fraction of the overall contaminant to achieve this. As shown in Figure 11, chloroform was predicted in the irrigated soil at around 4 x 10^1 ng/m^3, 1,1,2-trichloroethane at 1 x 10^2 ng/m^3 and pyrene at 3 x 10^3 ng/m^3.

Figure 11 Steady-state concentrations of contaminants among phases



Sensitivity analysis of the exposure modelling

Examination of the parameters that contribute most to determining the predicted concentrations of the contaminants was undertaken by sensitivity analysis of the fugacity model to its parameters.

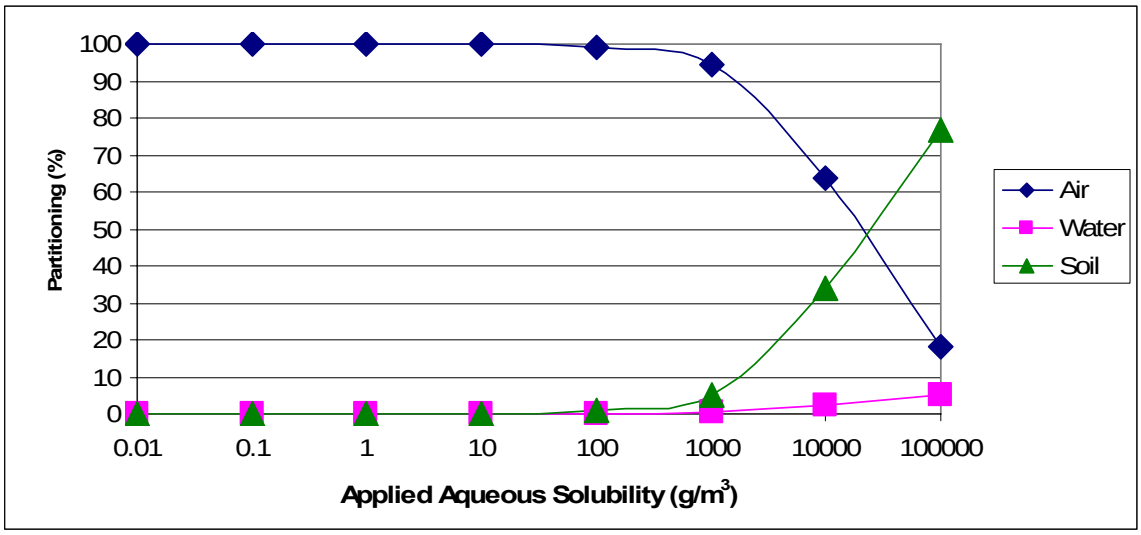
The effects of changes in assumed recycled water concentrations are directly proportional to the magnitude of the predicted environmental concentrations. Accordingly, where accurate absolute (rather than relative) environmental predictions are required, similar accuracy in initial recycled water concentrations is necessary.

The relative values of aqueous solubility, Log Kow and vapour pressures have a complex interaction and relationship to each other. For example, partitioning between atmospheric and aqueous phases is governed by a determined Henry's law constant which is calculated according to both the aqueous solubility and vapour pressure. Therefore, compounds with different aqueous solubilities will have variable sensitivities to variations in data pertaining to vapour pressures.

As a result of these considerations, care should be taken when generalising according to single compound examples. In this case an illustrative sensitivity analysis is provided for 1,1,2-trichloroethane since its general behaviour is intermediate among the example compounds provided. However, in real environmental applications, a sensitivity analysis should be undertaken for a range of compounds.

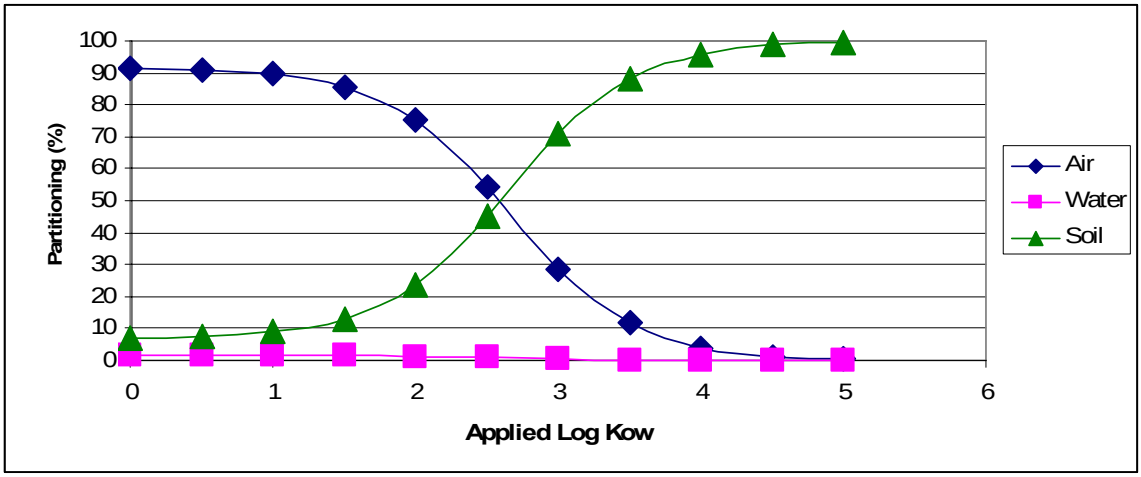
Sensitivity of predicted 1,1,2-trichloroethane distribution to applied aqueous solubility value is displayed in Figure 16. The modelled application assumed an aqueous solubility of 4590 g.m^{-3}. According to the trends given in Figure 12 an order of magnitude error in this figure could result in an under-prediction of the relative proportion in air (predominantly by partitioning to the soil phase) by up to about 50% of the total quantity. Analogous compounds with predicted aqueous solubilities less than 1000 g.m^{-3} would be substantially less affected by solubility errors in this current system.

Figure 12 Sensitivity of predicted 1,1,2-trichloroethane distribution to applied aqueous solubility



Sensitivity of predicted 1,1,2-trichloroethane distribution to applied Log Kow value is displayed in Figure 13. This indicates that in the modelled system, there is considerable susceptibility to error for analogous compounds with predicted Log Kow values in the range 2-3. Outside of that range, the relative partitioning between the most significant phases is not strongly effected by small variations in applied Log K_{ow}.

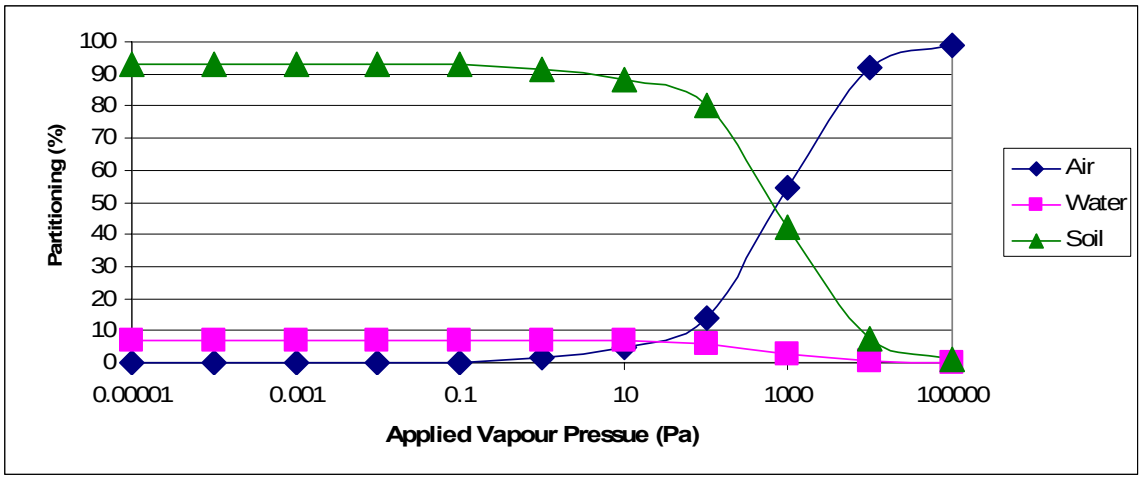
Figure 13 Sensitivity of predicted 1,1,2-trichloroethane distribution to applied Log Kow



Sensitivity of predicted 1,1,2-trichloroethane distribution to applied vapour pressure value is displayed in Figure 14. This indicates that analogous non-volatile compounds (for example those with vapour pressure < 10 Pa) are almost exclusively partitioned to soil and only very major errors in vapour pressure will have any significant impact. Similarly, those with vapour pressure > 10 kPa will be almost exclusively partitioned to the air phase and minor errors in the applied value will not affect this prediction. However, compounds with vapour pressures of between 0.1- 10 kPa are susceptible to miss-prediction in relative

partitioning between soil and air. Accordingly, concentration values calculated for such compounds should be interpreted as order-of-magnitude estimations only.

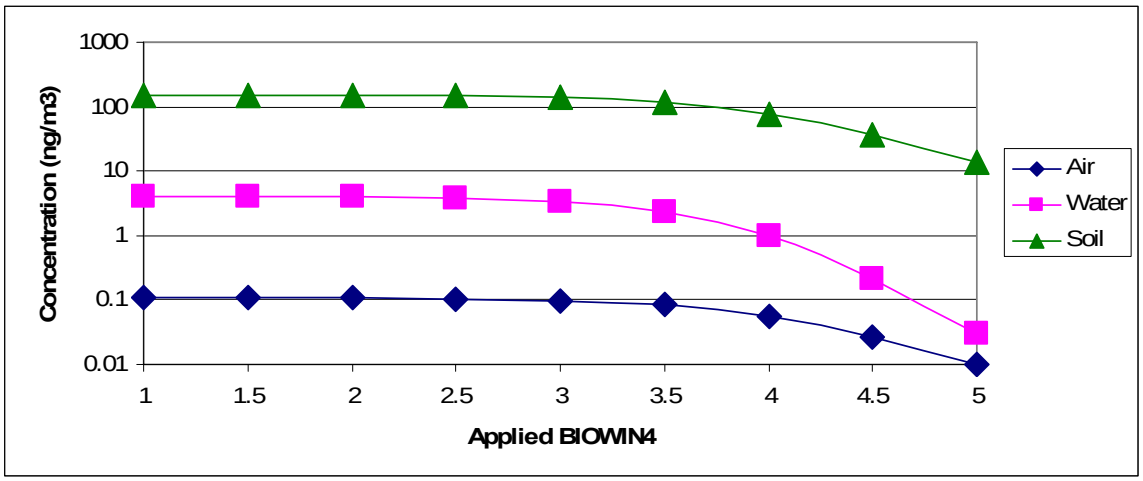
Figure 14 Sensitivity of predicted 1,1,2-trichloroethane distribution to applied vapour pressure



The applied parameters of BIOWIN4 and AOPWIN half life affect the overall removal of the compounds from the system. While this does have an effect on the relative distributions, the major impact can be more precisely observed by monitoring the predicted absolute concentrations. Accordingly, the sensitivity analysis of these two parameters are presented in terms of absolute concentrations ($ng.m^{-3}$), rather than relative distributions (%). It is important to recall that high relative partitioning of a chemical to a particular phase does not necessarily imply a corresponding high absolute concentration in that phase. Refer to the earlier discussion of steady-state concentrations for a detailed explanation of this apparent contradiction.

As mentioned previously, biodegradation rates are notoriously difficult to estimate and often constitute major sources of error in environmental modelling exercises. Sensitivity of predicted 1,1,2-trichloroethane concentrations to applied BIOWIN4 are provided in Figure 15. From this figure it is clear that an applied BIOWIN4 value less than about 3.5 will not significantly impact predicted concentrations. However, inaccuracies in BIOWIN4 values greater than 3.5 have the potential to affect predicted concentrations in air and soil by an order-of-magnitude and possibly two orders of magnitude in water.

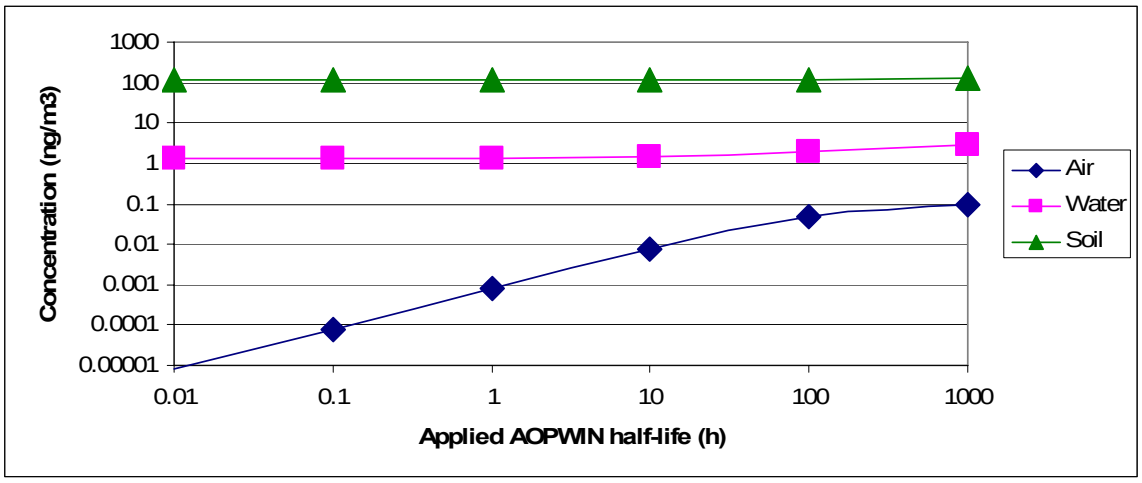
Figure 15 Sensitivity of predicted 1,1,2-trichloroethane concentrations to applied BIOWIN4



Sensitivity of predicted 1,1,2-trichloroethane concentrations to applied AOPWIN half lives are provided in Figure 16. As is apparent, predicted values of AOPWIN half lives have negligible effect on the predicted concentrations in the soil and water phases. On the other hand, a very significant relationship can be observed between predicted half-life and concentration in the air phase. In this case, an order of magnitude change in atmospheric half-life results in roughly an order-of magnitude change in predicted concentration in the air phase. The influence of AOPWIN half lives is lessoned after a value of around 100 hours.

It must be emphasised that the fugacity modelling approaches presented in this document are not intended to provide exact predictions of chemical concentrations. Rather, their value is in identifying expected trends in chemical behaviour and distribution. This will be of significant value when assessing potential impacts of environmental exposures of chemicals as a result of water recycling applications. The above sensitivity analysis has confirmed the earlier-stated precaution that such models are generally useful for order-of-magnitude estimations and should never be presumed to provide more accurate predictions that this.

Figure 16 Sensitivity of predicted 1,1,2-trichloroethane concentrations to applied AOPWIN



Dose-response-assessment: Predicted no effect concentrations

A simple theoretical model has been developed for the estimation of the maximum permissible soil pollutant concentration based on toxicological data [23, 102]. It is useful to determine the maximum soil pollutant concentration instead of a wastewater concentration because the wastewater composition is highly fluctuating and accumulation of contaminants takes place in the soil phase. The values obtained from this model may be applied as the PNEC of a specific chemical. The model is used to calculate a maximum allowed soil pollutant concentrations Cs (mg/kg) from a chemical's ADI in milligrams per kilogram of body weight (BW) per day [23, 102]:

$$C_s(mg/kg) = \frac{ADI(mg/kg\ BW\ day) * 60(kg\ BW) * B}{\Sigma(I_i * f_i * K_{spi} * FD_i)}$$

The additional parameters of this equation include an index (i) representing the food groups grain/cereal, vegetable, root/tuber, and fruit; a daily consumption rate (I_i) of i-th food group; the fraction (f_i) of i-th food group affected by land application; a plant/soil partition factor (K_{spi}) for i-th food group; and a fresh weight to dry weight conversion factor (FD_i) for i-th food group. Depending on the background exposure of each pollutant, the portion of the ADI assigned to pollutant exposure due to land application of wastes may vary. To account for this, the model incorporates a coefficient of background (B) exposures from consumption of food plants that do not exceed 50% of the ADI.

In this analysis we have applied the values for the constant parameters as those previously recommended by Cheng *et al*. [23, 102]. We have also applied the method suggested by them for the estimation of plant/soil partition factors: Log Ksp = 1.588 – 0.578 x Log Kow [23, 102].

As a worst-case scenario, the hypothetical exposure in this study was represented by the food chain exposure of an adult (60 kg) whose entire dietary intakes of grain/cereal, vegetable, root/tuber, and fruit are derived from food plants grown in reclaimed water-irrigated soils. For each of the modelled chemicals, the ADI was selected as the published reference dose (RfD) from the US-EPA Integrated Risk Information System (IRIS) [96]. The soil concentrations calculated from this model are given in Table 26 as PNEC.

Table 26 PNEC established by modelling from reported reference doses (RfD)

Organic compound	RfD (mg/kg·d) [96]	PNEC (mg/kg)
Chloroform	0.01	2×10^{-1}
Pyrene	0.03	3×10^{1}
1,1,2-Trichloroethane	0.004	9×10^{-2}

The calculated PNEC values are significant above average soil concentrations described in the literature [3]. It is important to emphasise that the PNEC values calculated here are based purely on human toxicological implication and do not consider potential environmental impacts. Furthermore, there is a lack of supporting subchronic, chronic and reproductive studies data, which reduces the confidence in the resulting RfD value used in this simple model. A small sensitivity analysis showed that apart from the RfD value and the bodyweight (see below) the further parameters such as log K_{ow} and food composition do not influence significantly the resulting soil pollutant concentration.

Risk characterisation

The exposure assessment indicates that during reclaimed water irrigation the selected organic model contaminants would accumulate in varying degrees in the soil of the irrigated property. Accordingly, the uptake of the chemicals to food grown in the soil will be an important exposure route for human risk assessment.

Calculation of risk quotients can be undertaken, for each of the three chemicals in the hypothetical study, from the determined values of PEC and PNEC. By adjusting the soil PEC values from Section 4 and the PNEC from Section 5 to the same units, a risk quotient can be calculated for each of the three contaminants. The risk quotients then are chloroform (10^{-7}), pyrene (10^{-7}) and 1,1,2-trichloroethane (10^{-6}). Presuming a higher effluent concentration of 10 µg/L instead of 10 ng/L the risk quotients are accordingly to the sensitivity analysis a thousand times higher (chloroform, 10^{-4}; pyrene, 10^{-4}; 1,1,2-trichloroethane, 10^{-3}). When small children instead of adults are considered, one can assume in the worst case a roughly tenfold lower ADI due to a higher sensitivity of children towards toxic compounds and an approximately tenfold lower body weight (6 kg would be a six month old baby). The risk quotients would be therefore 100 times higher than for adults. Alltogether this could lead to a reduction of the risk quotient by 10^{5}.

In addition to the three model compounds with different physical-chemical properties we modelled the PEC and PNEC values of a broad range of organic micropollutants of concern. Our results for the predicted no effect concentrations as maximum permissible pollutant concentration of soil receiving sewage sludge or untreated municipal wastewater are shown in Table 27.

In such a scenario, the risk quotients for all chemicals would be acceptably low for human health effects via the uptake from food. While the risk quotients would be acceptable, the high degree of uncertainty in the modelling processes would indicate the strong desirability for analytical confirmation including a monitoring program throughout numerous stages of the agricultural irrigation scheme. A further high degree of uncertainty emanates from availability of comprehensive data for appropriate ADI or RfD levels. Data of subchronic, chronic and reproductive effects were not sufficiently available. For many compounds ADI and RfD values are not available at all.

Table 27 Predicted environmental concentrations (PEC) and predicted no effect concentrations (PNEC) as maximum permissible pollutant concentrations of soil receiving sewage sludge and/or untreated municipal wastewater based on human toxicological implications [23]

Organic compound	Log K_{OW}	PEC [mg/kg]	PNEC [mg/kg]	PEC/PNEC
Chloroform	1.97 [103]	2.4E-08	0.24	9.8E-08
1,1,2-Trichloro-ethane	1.89 [103]	8.2E-08	0.08	1.0E-06
Pyrene	4.88 [103]	2.2E-06	34.67	6.2E-08
Atrazine	2.81*	1.2E-06	2.60*	4.5E-07
Simazine	2.18 [103]	9.1E-07	0.21	4.3E-06
Diuron	2.68 [103]	1.1E-06	0.12*	8.9E-08
Bisphenol A	3.32 [103]	6.2E-07	7.25	8.6E-08
Methyl-tert-butylether	1.43*	3.0E-08	35.11*	8.5E-10
Phenol	1.46 [103]	1.1E-07	3.65	2.9E-08
Benzene	2.13 [103]	1.5E-08	0.13	1.1E-07

* estimated Log K_{OW} values by EPIWIN

In cases where unacceptably high risk quotients are predicted, modifications to the intended water recycling strategy will be necessary. Such modifications could include a more effective treatment of the water prior to irrigation; a decrease in the amount of reclaimed water to be used on the property; or a change in the choice of crops to be grown.

This study has presented an illustrative analysis of a highly simplified hypothetical wastewater reuse application. However, it has successfully demonstrated the general concepts of an approach to chemical risk assessment that, with refinement, should be highly applicable to a very wide variety of circumstances. While the procedure does rely on a certain amount of real input data for a particular system, it will be possible for judiciously selected "typical" values to be used for many parameters in real risk assessment cases. This is because the final values predicted by the models will not always be highly sensitive to all of the modelled parameters.

Finally, risk assessment of chemicals regarding wastewater reuse will, in many cases, need to be undertaken with respect to environmental organisms, as well as humans. Consequently, a wide variety of PNEC techniques will be required. In practice, the number of analyses can be minimised by undertaking risk assessments for "worst case scenarios". This may be achieved by identifying the most highly susceptible organisms which are affected in the reuse application and calculating PNEC values solely on the basis of those organisms.

5.5. Ecological Risk Assessment

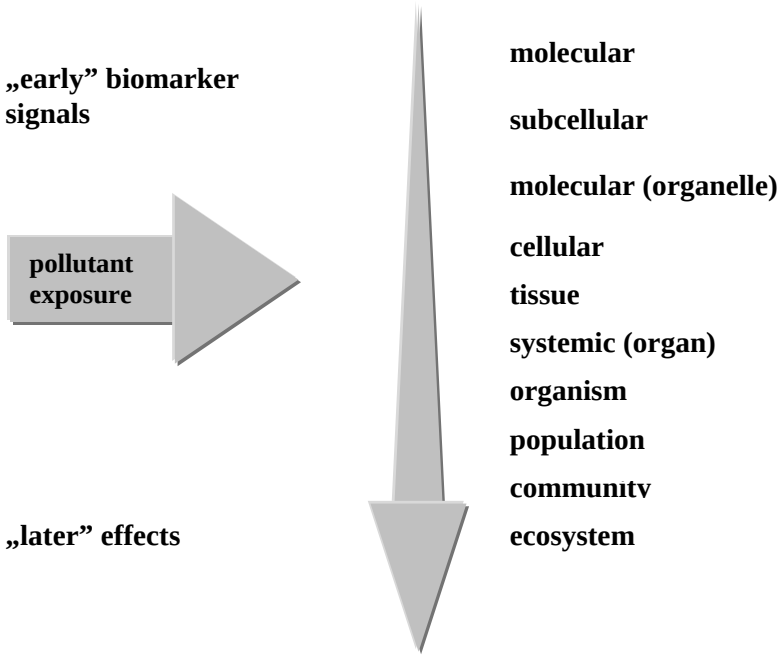
5.5.1. Introduction

Ecological/environmental risk assessment is defined as the complex evaluation process by which the potential or actual adverse effects of pollutants and other anthropogenic activities on ecosystems and their components are estimated - with a known degree of certainty using scientific methodologies [104].

The ability of various pollutants (and their derivatives) to mutually affect their toxic actions complicates the risk assessment based solely on environmental levels.

Deleterious effects on populations are often difficult to detect in feral organisms since many of these effects tend to manifest only after longer periods of time. When the effect finally becomes clear, the destructive process may have gone beyond the point where it can be reversed by remedial actions or risk reduction. The sequential order of responses to pollutant stress within a biological system is visualized in Figure 17.

Figure 17 Schematic representation of the sequential order of responses to pollutant stress within a biologic system [modified from 104]



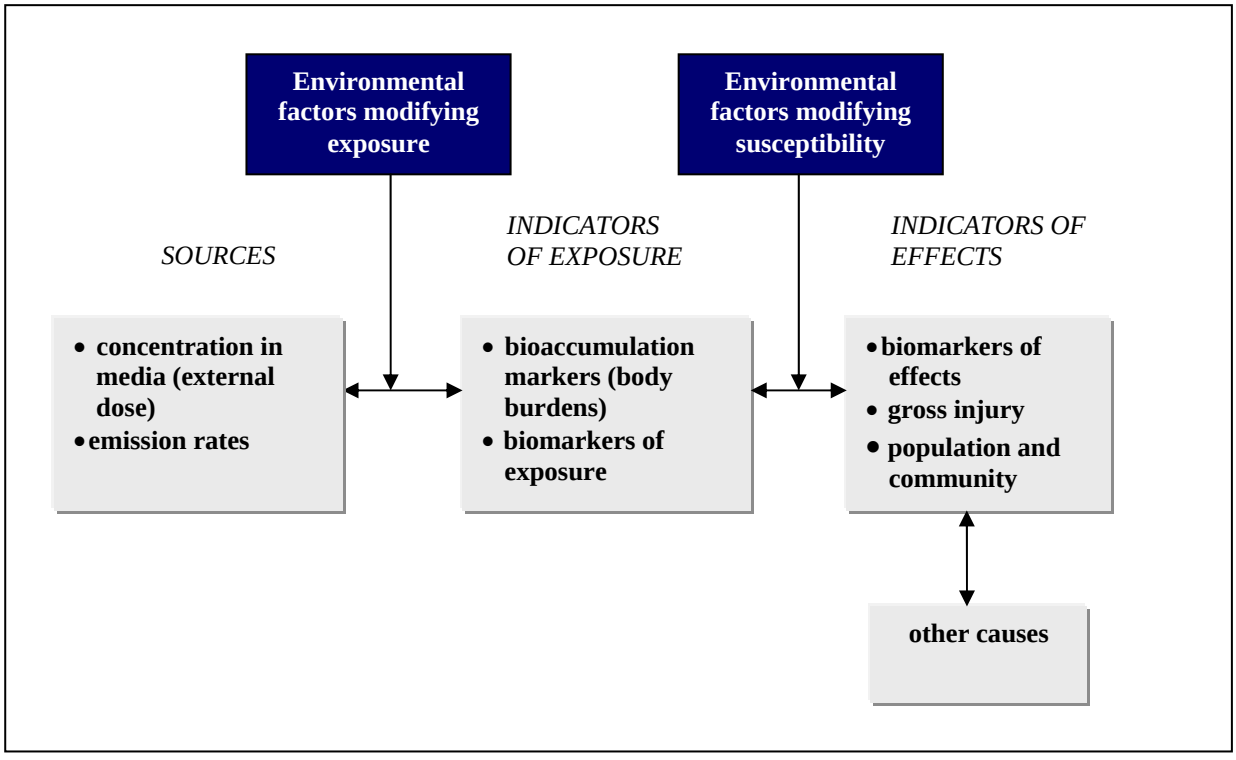
Risk management and risk analysis are closely related but different processes: in risk analysis the risk of a certain situation is determined, whereas risk management examines solutions to the problem. Managing risks involves making decisions based on the information collected in the previous steps of risk assessment along with a consideration of social and cultural values, economic realities and political factors. Ecological risk assessments are developed within a risk management context to evaluate human-induced changes that are considered undesirable. It is very important to define adversity because a stressor may cause adverse effects on one ecosystem component but be neutral or even beneficial to other components. Changes often considered undesirable are those that alter important structural or functional characteristics or components of ecosystems. An evaluation of adversity may include a consideration of the type, intensity, and scale of the effect as well as the potential for recovery. The acceptability of adverse effects is determined by risk managers. Although intended to evaluate adverse effects, the ecological risk assessment process can be adapted to predict beneficial changes or risk from natural events.

Ecological risk assessment (ERA) has been employed primarily to deal with chemicals. In the last decade much research has been devoted to ERA, mainly by international bodies like the World Health Organization (WHO), the Organization for Economic Co-operation and Development (OECD) and the European Centre for Ecotoxicology and Toxicology of Chemicals (ECETOC).

The objective of risk based environmental regulation is to balance the degree of permitted risk against the cost of risk reduction and against competing risks. Ecological risk assessment has several advantageous properties in environmental decision-making [106]. It provides a quantitative basis for comparing and prioritizing risks as well as a systematic means of improving the understanding of risks. In addition, it estimates clear consistent endpoints.

Ecological Risk Assessment (ERA) and risk assessment in general, is primarily concerned with predictive assessments that estimate the nature, probability and magnitude of effects of proposed actions [106]. However, emphasis has been shifting to retrospective assessments, i.e. assessments of human actions that were initiated in the past and may have ongoing consequences in the future, such as waste sites, acid rain and existing pesticides [106]. Retrospective assessment falls into three categories with respect to the direction of inference (Figure 18).

Figure 18 The relationship among the components of the risk characteristics stage of retrospective assessment based on the process of ecological epidemiology, including their respective environmental methods [modified from 105]



Source-driven assessments begin with an existing pollution source, such as a spill or an effluent, and attempt to determine the nature of the effects. Exposure-driven assessments are prompted by evidence of exposure without prior evidence of a source or effects. Effects-driven assessments begin with an observed effect such as a declining animal population or a fishless lake, and attempt to determine a cause. In all cases, the logical link between sources and effects is exposure [106]. The retrospective assessment of hazards and risks of existing chemicals may be established by actual measurements of concentrations and effects in the field, using bioaccumulation and biomarkers.

The last four stages of risk assessment, distinguished by Van der Oost (2003) [105] are usually categorized as part of risk management. Anyway, monitoring process is worth exposing as an essential element of ecological risk assessment and the most relevant with respect to biomarkers.

Any risk assessment must have defined endpoints. An assessment endpoint is a formal expression of the environmental values to be protected [106]. Defining an assessment endpoint involves two steps: (1) identifying the valued attributes of the environment that are considered to be at risk, and (2) defining these attributes in operational terms.

Suter [106] proposed five criteria that any endpoint should undertake:

1) societal relevance (understood and valued by public and decision-makers);

2) biological relevance (important to a higher level of the biological hierarchy);

3) unambiguous operational definition;

4) accessibility to prediction and measurement;

5) susceptibility to the hazardous agent(s).

5.5.2. Environmental risk monitoring

Monitoring is a repetitive observation for defined purposes of one or more chemical or biological elements according to a prearranged schedule over time and space, using comparable and standardized methods (according to the definition of the United Nations Environmental Program, UNEP).

This last step in the risk management process, which is most relevant with respect to e.g. biomarkers, may serve a number of purposes: the control function to verify the effectiveness of risk reduction or to ensure that previously formulated standards are being met, the signal or alarm function to detect sudden adverse changes in the environment, the trend function to enable the prediction of future developments and the instrument function for the recognition and clarification of underlying processes. It is important that environmental monitoring programs should only be undertaken if the objectives clearly state what the data are going to be used for [105].

There are five main types of environmental risk monitoring methods [105] which are performed to assess risks of contaminants for organisms and to classify the environmental quality of ecosystems:

1. Chemical monitoring (CM): exposure assessment by measuring levels of a selected set of well-known contaminants in abiotic environmental compartments;

2. Bioaccumulation monitoring (BAM): exposure assessment by measuring contaminant levels in biota or determining the critical dose at a critical site (bioaccumulation);
3. Biological effect monitoring (BEM): exposure and effect assessment by determining the early adverse alterations that are partly or fully reversible (biomarkers);
4. Health monitoring (HM): effect assessment by examining the occurrence of irreversible diseases or tissue damage in organisms;
5. Ecosystem monitoring (EM): assessment of the integrity of an ecosystem by making an inventory of, for instance, species composition, density and diversity.

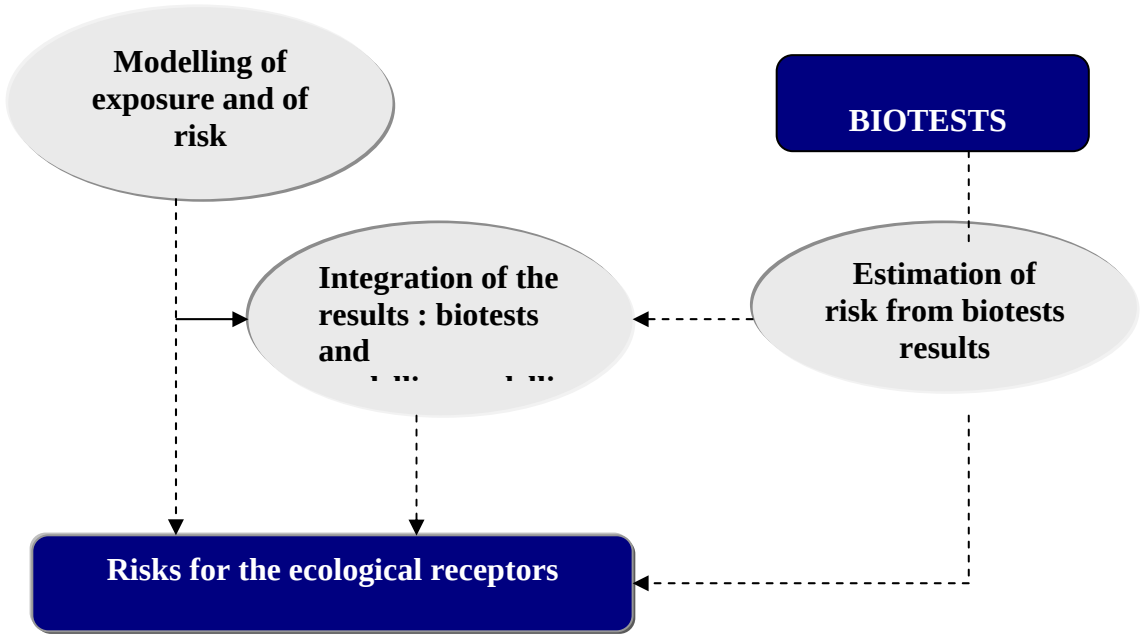
A regular, systematic use of living organisms to evaluate changes in environmental or water quality, as in BAM, BEM, HM and EM, is called biological monitoring (BM) or biomonitoring. The term 'toxicity monitoring' is used to describe measurements on the direct biomolecular and physiological responses of individual organisms towards toxicants in an experimental setup, including bioassays and biological early-warning systems, e.g. in BEM and HM. The integrated monitoring program is a study consisting of coordinated monitoring activities comprising both chemical and biological measurements in a variety of environmental media or compartments.

5.5.3. Monitoring tools in the environmental risk assessment

Biological tests

Biological tests are the perfect monitoring tools for ecological assessment. By using toxicity tests it is possible to estimate the toxic impacts of different, often fluctuating, contaminations for human health and determinate risk assessment in all environmental media.

Figure 19 Mathematical modelling and biotests to assess the risk related to the contamination of a particular site [modified from 107]



Usefu

lness of biotests in the estimation of ecological risk assessment is shown on Figure 19.

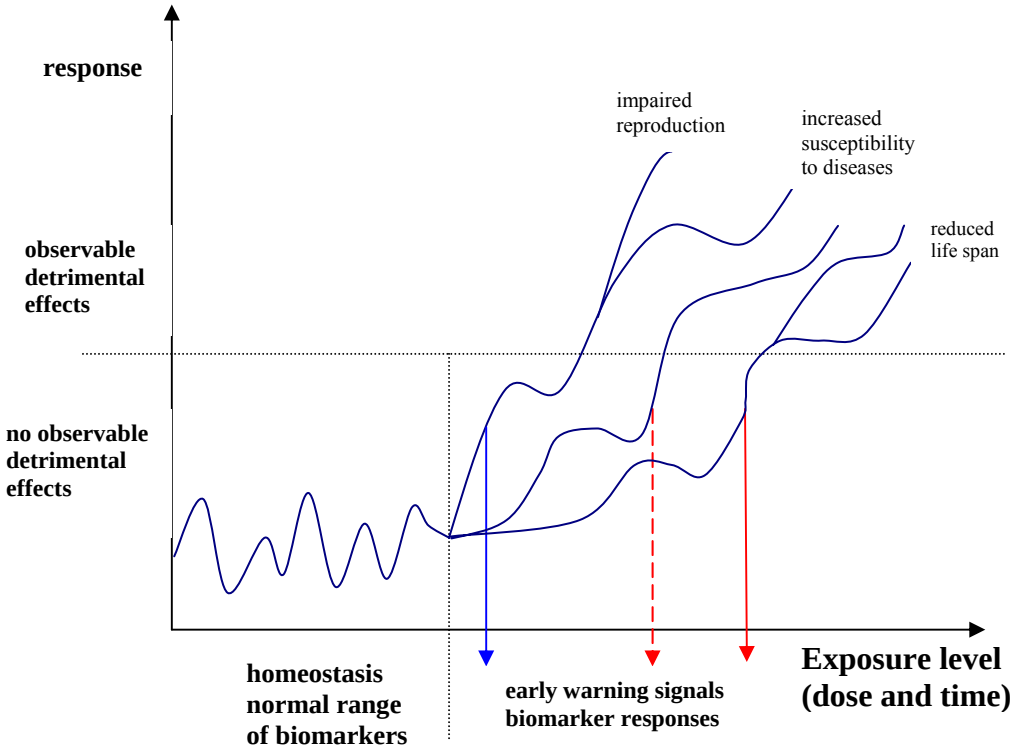
⇒ Biotests are used as a monitoring tool, since the primary goal of bioremediation is to reduce toxic effects;

⇒ Biotests can help guide the application of bioremediation agents, in order to reduce the probability of detrimental treatment effect.

Microorganisms applied in water toxicity testing [microbial tests are simple, rapid, sensitive and inexpensive] are also defined as biomarkers. As a result a biomarker is considered as a change in a biological response (ranging from molecular through cellular and physiological responses to behavioral changes) which can be related to exposure or toxic effects of environmental chemicals.

Figure 20 shows the pollutant-responsive biomarker signals deviation from the normal range in an unstressed situation.

Figure 20 The principal scheme of responses in organisms to the detrimental effects of pollutant exposure [105]



Most of the microbiological tests [108] are based on measurements of:

- Growth inhibition

  (monitored by optical density measurements; ISO 10721, 1995 and ISO 15522, 1999 or by cell counting or fluometry in algal tests: ISO 8692, 1986 and ISO 10253, 1998);

- Respiration

  (based on carbon dioxide production or oxygen consumption during the test: ISO 8192, 1986);

- Mineralization

  (material containing organic nitrogen is used in tests measuring nitrification: ISO 9509,1989 and OECD TG 216, 2000);

- Metabolism enzyme activity and viability of bacterial cells

  (measured directly by luminescence measurements: ISO 11348, 1998).

Types of mostly applied biotests (examples) :

- AMES tests
- ERO
- Vitelogenin
- Yeast Estrogen Screen (YES)

Surface and groundwater

Microbial tests used to assess the toxicity for surface and groundwater are compiled in Table 28.

Table 28 Microbial test used to assess the toxicity for surface and groundwater [108]

Test	Organism(s)	Principle	Standardization
Escherichia coli luminescence inhibition test	GMO *E.coli*	Inhibition of luminescence in GMO *E.coli*	-
Bacillus subtilis luminescence inhibition test	GMO *B.subtilis*	Inhibition of eukariotic luminescence in GMO *B.subtilis*	-
ATP-TOX test	*Pseudomonas fluorescens, Salmonella typhimurium E.coli*	Inhibition of growth and luminescence	-
Rapid automated bacterial impedance test	Mixed population	Inhibition of production of ionizisng compounds by bacteria	-
Spirillum flagella test	*Spirillum volutans*	Inhibition of flagellated bacteria movement	-
Activated sludge respiration inhibition test	Activated sludge	Oxygen consumption inhibition in activated sludge	ISO 9509
Activated sludge nitrification inhibition	Activated sludge	Inhibition of nitrate production from ammonium salts by activated sludge	-
Dehydrogenase activity inhibition test (EPA)	*E.coli*	Inhibition of dehydrogenase activity	-
Galactosidase activity inhibition tests : MetPAD, MetPLATE	*E.coli*	Inhibition of galactosidase activity	-
Galactosidase activity inhibition tests (Toxi- Chromo)	*E.coli*	Inhibition of production of galactosidase	-
Galactosidase induction inhibition	*Bacillus licheniformis*	Inhibition of production	-

Test	Organism(s)	Principle	Standardization
test *Pseudomonas* fluorescens growth rate inhibition test	*Pseudomonas* fluorescens	of galactosidase Cell multiplication inhibition (rate)	

Effluent quality

The risk associated with pathogens is not fully managed with effluent treatment. Activated sludge secondary biological treatment and disinfection are ineffective treatments for the total elimination of these parasites; nevertheless, helminth eggs and protozoa cysts generally reach the effluent in big numbers.

Test battery for effluents consist of species belonging to different taxonomic groups and different trophic levels :

- 72h growth inhibition test with *Selenastrum capricornutum*
- 24h growth inhibition test with *Thamnoscephalus platyurus*
- 48h immobilisation test with *Daphnia magna Stratuss*
- 24h inhibition test with *Tetrahymena thermophila*
- 7 days growth inhibition with *Lemna minor* - test was performed according to Swedish Guidelines [109].

Taking into account the whole spectrum of results the investigated test species according to their sensitivity, the results showed that *Thamnoscephalus platyurus* is the most sensitive species. According to their sensitivity, the species can be ranged as follows [110]: *Thamnoscephalus platyurus* > *Lemna minor* > *Selenastrum capricornutum* > *Daphnia magna* > *Tetrahymena thermophila.*

5.5.4. Bioassays methods

A major goal of ecotoxicological studies is the development of cause-effect linkages between biological systems and the complex pollutant mixtures to which they are exposed.

The integrated environmental risk assessment [105], is consisting of four different biomonitoring levels using biomarkers, bioassays, bioindicators and ecological indicators:

(I) Sub-organismal (biomarkers): At the level of biochemical and physiological processes, deviations from the normal situation (‘health’) can be measured using biochemical techniques.

(II) Organisms (bioassays): Survival, growth and reproduction of individuals are chosen as endpoints of the classic laboratory ecotoxicity tests.

(III) Populations (bioindicators): At this level, effects are manifested as changes in the genetic structure, the age structure or the abundance of a population.

(IV) Ecosystems (ecological indicators): At this level, changes in species composition, abundance and diversity may be indicative of the effects of pollution on communities.

Biomarkers

A biomarker is defined as: "a change induced by a contaminant in the biochemical or cellular components of a process, structure or function, that can be measured in a biological system" [111]. This change provides qualitative and semiquantitative information on :

- the nature of chemical insult
- relation between biological effects
- levels of environmental contamination

The most compelling reason for using biomarkers is that they can give information on the biological effects of pollutants rather than a mere quantification of their environmental levels. Biomarkers may provide insight into the potential mechanisms of contaminant effects. By screening multiple biomarker responses, important information will be obtained about organism toxicant exposure and stress. A pollutant stress situation normally triggers a cascade of biological responses, each of which may, in theory, serve as a biomarker [112]. Above a certain threshold (in pollutant dose or exposure time) the pollutant-responsive biomarker signals deviate from the normal range in an unstressed situation, finally leading to the manifestation of a multiple effect situation at higher hierarchical levels of biological organization.

Ecotoxicological biomarkers: A biochemical, cellular, physiological, or behavioral variation that can be measured in tissue or body fluid samples or at the level of whole organisms that provides evidence of exposure to and/or effects of one or more chemical pollutants (and/or radiation):

- biomarkers of exposure - which signal exposure of an organism, a population, or a community to chemical pollutants;
- biomarkers of effect - which signal that an organism, a population, or a community has been affected (usually adversely) by one or more pollutants;
- biomarkers of susceptibility - an indicator of an inherent or acquired ability of an organism to respond to the challenge of exposure to a specific xenobiotic substance.

Molecular biomarkers : the research tools which are an early sign of change in an organism's physiological state - such as adaptation, stress or injury - due to environmental factors or disease.

Biosensors

Biosensors are devices that detect, record, and transmit information regarding a physiological change, the presence of various durable signals or presence of various chemical or biological materials in the environment. More technically, a biosensor is a probe that integrates a biological component, such as a whole bacterium or biological product (e.g., an enzyme or antibody) with an electronic component to yield a measurable signal. Biosensors, which come in a large variety of sizes and shapes, are used to monitor changes in environmental conditions. They can detect and measure concentrations of specific bacteria or hazardous chemicals. They can measure acidity (pH). Biosensors can use bacteria and detect them, too.Biosensors consists of two components :

Biosensor = bioreceptor + transducer

The bioreceptor is a biomolecule that recognizes the target analyte whereas the transducer converts the recognition event into a measurable signal.

Biosensors technology provides :

- Identification and diagnosis of contamination in a much shorter time scale than existing technologies.
- Information which is environmentally relevant.
- Laboratory simulation of reclamation strategies before site application to give certainty in results and estimation of time scale to completed clean-up.

Biosensors technology is :

- Rapid: diagnosis risk assessment and solution
- Relevant: approach is environmentally relevant
- Reliable: solutions are laboratory proven before site application
- Relasing: optimizing land value

Bioassays/Biotests - criteria of selection

Selection of bioassays of environmental pollution to evaluate risk assessment depends upon a suitable model whose data could be extrapolated to determinate the risk to humans [the toxicant can produce metabolites in certain organisms, which may or not be toxic to humans]. |

According to Jamil [2001] there are two types of criteria: (1) obligatory = conditions essential to the realization of a correct test; (2) desirable = indicators that can improve the precision of the test. To verify the applicability of bioassays according to the field conditions and objectives there are Standard Operating Procedure (SOP) for :

a) Obligatory criteria :
   - methods published by organizations such as the ISO, ASTM, OCDE, EPA.
   - the mention of toxic product of the reference biological identification.
   - criteria of acceptability of good health with test organisms.

b) Optional tests :
   - identification of one assay organisms (age, sex, stage)
   - identification of measurable end point
   - selection organisms
   - number of organisms
   - frequency of observation
   - culture and handling of organisms colonies
   - environmental conditions
   - definition and manipulation of media
   - statistical analysis.

Bioindicators

Bioindicators: organisms and organisms associations which respond to pollutants on an association of organisms (see also chapter 5.5.8).

The most important reasons for using bioindicators are:

- the direct determination of biological effects
- the determination of synergic effects of multiple pollutants on an organism
- the early recognition of pollutant damage to plants as well as toxic dangers to humans and animals
- relatively low cost compared to technical measuring methods.

Bioindicators are commonly grouped into accumulation indicators and response indicators**:**

- accumulation indicators store pollutants without any evident changes in their metabolisms.
- response indicators react with cell changes or visible symptoms of damage.

5.5.5. Bioaccumulation in aquatic organisms

Exposure assessment has to provide information on steady-state concentrations of potentially toxic xenobiotics in a selected environmental compartment methods for assessing exposure to a chemical fall into two categories :

⇒ Measurement of levels of chemical agents and their metabolites and/or derivatives in cells, tissues, body fluids or excreta.

⇒ Measurement of biological responses such as cytogenetic and reversible physiological changes in the exposed individuals**.**

In evaluating exposure, a distinction is made between the external dose, defined as the amount of chemical agent in environmental contact with the organisms as determinated by chemical monitoring (CM), and internal dose which is the total amount of a chemical agent adsorbed by the organism over a period of time as determined by bioaccumulation monitoring (BAM) [105].

Bioaccumulation markers and biomarkers of exposure will reflect the distribution of the chemicals or its metabolites, respectively, throughout the organism .

Bioaccumulation

Persistent hydrophobic chemicals may accumulate in aquatic organisms through different mechanisms:

- via the direct uptake from water by gills or skin (bioconcentration),
- via uptake of suspended particles (ingestion).
- via the consumption of contaminated food (biomagnification).

Even without detectable acute or chronic effects in standard ecotoxicity tests, bioaccumulation should be regarded as a hazard criterion in itself, since some effects may only be recognized in a later phase of life, are multi-generation effects, or manifest only in higher members of a food-web, e.g. impact of PCBs on the hatching success of fish eggs.

Bioaccumulation of chemicals in biota may be a prerequisite for adverse effects on ecosystems. Contaminant levels in biota are determined primarily by the uptake and elimination kinetics, which are

typical for both chemicals and organisms. A model of the processes governing bioaccumulation (uptake and clearance) in aquatic organisms is presented in Figure 21. According to this model, the concentration of a chemical in biota (CB) over time (t) can be expressed by:

$$d_B/dt = [k_w C_w + k_F C_F] - k_B C_B + [k_w C_w + k_F C_F] - [k_{EXC} + k_{MET}] C_B$$

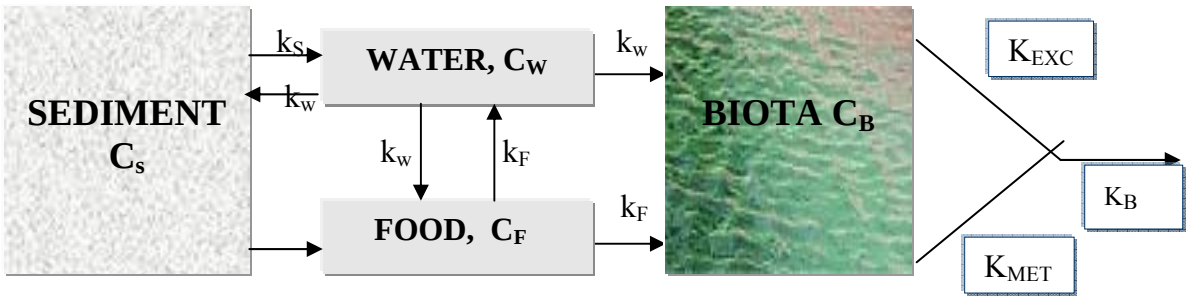
C, refers to a concentration; k to a rate constant; and the subscripts W, F, B, EXC and MET to water, food, biota, excretion and metabolism, respectively.

Uptake of organic pollutants in fish may be direct via exchange with the water phase ($k_w C_w$) or indirect via the consumption of contaminated food ($k_F C_F$) .

Figure 21 Bioaccumulation model for aquatic organisms [modified from 105]

BSAF (biota-sediment)

K_{OC} (sediment-water) **BCF** (biota-sediment)



BMF (biota- food)

K_{OC} ; sorption coefficient; **BCF** : biomagnification factor. C refers to a concentration and *k* to a rate constant. The substripts **S, W, F, B, EXC** and **MET** refer to sediment, water, food, biota, excretion and metabolism, respectively. The digestible sediment fraction is considered to be part of food [105].

Bioconcentration

The bioconcentration factor (BCF) of a chemical is the ratio of its concentrations in the organism and in water during steady state or equilibrium. For the partitioning of chemicals between water and the lipid phases of organisms, the steady state BCF is defined as:

$$BCF = k_w / k_B = C_B / Cw$$

Uptake of chemicals in organisms from water probably follows a passive diffusion mechanism analogous to that of oxygen uptake. The rate of uptake of hydrophobic chemicals in fish increase with a higher lipid content of the biological membranes [113].

The time required reaching a steady state between the water and fish concentrations can be determined by caging uncontaminated fish in polluted areas and measuring pollutant tissue levels after different exposure times.

The fate of chemicals is largely determined by sorption to suspended particulates and sediments [114]. Sorption depends on the characteristics of both the sediments and the chemicals involved. If sorption of hydrophobic chemicals is considered as a partitioning between water and the organic fraction of sediment, then the equilibrium sorption coefficient (K_{OC}) can be expressed as:

$$K_{OC} = k_w / k_s = C_s / Cw$$

If the processes of bioconcentration and sorption on sediments (i.e. the upper part of the model in Fig. 4) have both reached equilibrium, then equilibrium (2) and (3) can be combined to define the biota sediment accumulation factor (BSAF):

$$BSAF = C_B / C_S = BCF / K_{OC}$$

Biomagnification

Biomagnification is the ratio between the uptake of the chemicals from food and their clearance. During steady state, the biomagnification factor (BMF) can be defined as:

$$BMF = k_F / k_B = F_F (E_F / k_B)$$

$F_{F,}$ refers to the amount of food transported through the intestines per gram of fish per day and E_F to the efficiency of uptake of the chemical from food. Since bioaccumulation of persistent and extremely hydrophobic compounds cannot always be explained satisfactorily by simple partitioning processes between sediment, water and fish [105] it is likely that the uptake via contaminated food (biomagnification) contributes significantly to the bioaccumulation of these contaminants in fish.

If a primary source of chemical input to an aquatic ecosystem is slow release from polluted sediments, then it is plausible that uptake by benthic organisms followed by predation by larger organisms such as fish may be a significant source of bioaccumulation.

The mechanism of biomagnification and food chain accumulation of organic chemicals can be explained with a fugacity-based hypothesis, which has been validated by experimental findings. Fugacity is equivalent to chemical activity or chemical potential as it pertains to the tendency of a chemical to escape from a phase, such as water or food [115].

A difference in fugacity provides a driving force for net passive chemical transport from high to low fugacity phases. Food digestion in the gastrointestinal tract was found to increase the chemical fugacity in the food 4- 5-fold by altering the fugacity capacity, while an additional 2- 3- fold increase in the chemical concentration and fugacity is caused by a reduction of the food volume due to absorption from the gastrointestinal tract.

Biomagnification may be more important for e.g. in larger fish than for smaller fish, since relative gill ventilation volumes decrease with size while relative feeding rates are almost equal. When biomagnification is an important uptake route, individual and site-specific variations in bioaccumulation patterns may, at least partly, be due to differences in the fish diet.

This hypothesis may be investigated by determining the levels of d^{15} N in the contents of the gastrointestinal tract of fish, since this stable isotope is reported to be an integrative measure of trophic position, increasing with higher trophic levels [116].

Bioavailability

The bioavailability is defined as the fraction of the bulk amount of the chemical present in soil/sediment and (interstitial) water that can potentially be taken up during the organism's lifetime into the organism's tissues (excluding the digestive tract). When the concentration in fish is not related to the real bioavailable concentration in the water, this might result in underestimation of the bioconcentration potential.

Usually, organisms preferentially ingest the smaller sediment particles which results in an increased contaminant uptake. Normalization of the contaminant concentrations to OM [organic matter] or OC [organic carbon] only partially accounts for this increased uptake. OM is the main determinant of sorption of hydrophobic compounds to soils and sediments. The bioavailability of these chemicals generally decreases with increasing soil or sediment OM content.

Biotransformation

An organism has two major ways of eliminating a chemical :

- it is either excreted in its original form (the parent compound)
- it is biotransformed by the organism.

Biotransformation generally leads to the formation of a more hydrophilic compound which is more easily excreted than the parent compound. The vertebrate organ most commonly involved in the biotransformation of foreign compounds is the liver, because of its function, position and blood supply. Biotransformation may also alter the toxicity of a compound, which may be either beneficial or harmful to the organism.

In case of a detoxication reaction, the toxicity of the compound is reduced while the excretion is generally elevated. In case of bioactivation, however, the compound is transformed into a reactive metabolite, which is more toxic than the parent compound. The biotransformation process may thus be important in determining the activity of a compound, the duration of that activity and the half-life of the compound in the body [117].

Pollutant concentrations in tissues and differences in excretion of metabolites can be a function of tissues and conditions controlling the activity of biotransformation enzymes. These conditions include spawning, nutritional status, conditions and duration of exposure to organic pollutants and life cycle stage of the animal. Another important example involves the interactive effects of one chemical pollutant on another.

Simultaneous exposure of fish to PCBs and PAHs, for instance, significantly influences the extent of uptake and metabolism of each. Fish tissue levels of chemicals that are easily biotransformed (e.g. low chlorinated PCBs, PAHs, non-2,3,7,8-substituted PCDD/Fs) are most likely not suitable as bioaccumulation markers for exposure assessment, since their tissue levels do not reflect levels in the surrounding environment [105].

Bioaccumulation models

One of the most important steps in the risk assessment process is the determination of potential exposure. Exposure estimation involves combining predicted concentrations for target chemicals with certain assumptions about the environmental fate of these chemicals and the activity patterns of the receptors

[118]. Subsequently, the results of the exposure assessment are combined with toxicity information to provide a quantitative estimate of risk.

In the past decades there has been substantial progress in all aspects of biogeochemical research related to the issues of bioavailability and disposition of toxic organic chemicals: solubility, sorption, uptake, metabolism, retention, release and excretion. Predictive equations have been derived or have evolved empirically that relate molecular structural characteristics or properties to biogeochemical behavior. Some predictive models for the bioaccumulation of xenobiotic organic chemicals in fish are listed the Table 31.

5.5.6. ECORISK MINIMIZATION: EFFECTIVE ENVIRONMENTAL DECISION MAKING

Consequently, as far as the wastewater recycling and reuse generates a certain degree of risk for humans and the environment, such risk should be rigidly and properly assessed and lately managed. The ecological risk assessment process has several features that contribute to effective environmental decision making:

1. Through an iterative process, new information can be incorporated into risk assessments, which can be used to improve environmental decision making.

2. Risk assessments can be used to express changes in ecological effects as a function of changes in exposure to stressors. This capability may be particularly useful to the decision maker who must evaluate tradeoffs, examine different alternatives, or determine the extent to which stressors must be reduced to achieve a given outcome.

3. Risk assessments explicitly evaluate uncertainty. Uncertainty analysis describes the degree of confidence in the assessment and can help the risk manager focus research on those areas that will lead to the greatest reductions in uncertainty.

4. Risk assessments provide a basis for comparing, ranking and prioritizing risks.

5. The results can also be used in cost-benefit and cost-effectiveness analyses that offer additional interpretation of the effects of alternative management options.

6. Risk assessments consider management goals and objectives as well as scientific issues in developing assessment endpoints and conceptual models during problem formulation. Such initial planning activities help ensure that results will be useful to risk managers.

Table 29 Predictive models for bioaccumulation of xenobiotic organic chemicals in fish [105]

Bioaccumulation models for xenobiotic organic chemicals in fish
• Model for PCBs in the Lake Michigan food chain, using growth, respiration or metabolic rate, assimilation efficiency of food (biomagnification), and bioenergetics.The model was validated in the field with alewife and lake trout.
• Model used for the interrelationship between BCF and toxicity, in combination with first order, one-compartment assumptions, to estimate toxicant kinetic parameters. The proportional relationship could be used to convert kinetics data from a bioconcentration basis to a toxicity basis and vice versa.
• A partitioning model for toxic chemicals between water, organic matter (OM) of sediments, particulate matter and biota lipids. The BSAF was estimated to be approximately 2 for organic chemicals, independent of their log KOW value.
• A life-cycle biomagnification model for hydrophobic organic chemicals, using lipid content, growth, uptake and elimination kinetics, as well as reproduction and biotransformation. Most parameters were derived from long-term laboratory experiments with guppies.
• Models for predicting chemical residues in aquatic food webs, which have gained general scientific acceptance and are being used for both scientific and regulatory applications. Both models incorporate the cumulative results of research originating from the early 1980s on bioaccumulation processes in aquatic food webs. Both models contain rate equations for estimation of steady-state conditions but also treat some chemical distributions as equilibrium partitioning.
• A model to predict nonequilibrium concentrations of trace contaminants in biota by relating the main non-steady state parameter, the outflow rate and the size of the species. This intermediately complex model allowed estimations for fairly unknown substances and species.
• A kinetic model to simulate bioaccumulation of toxic chemicals under natural conditions, using sediment sorption/desorption, bioavailability, uptake and elimination kinetics. Biomagnification was not taken into account. The model was validated in the laboratory with chlordane in goldfish.
• A mathematical model that estimates chemical concentrations in phytoplankton, zooplankton, filter-feeding and detritivorous benthic invertebrates and fish. A validation experiment illustrated that 95% of observed concentrations in invertebrates and fish were within a factor 2 of model-predicted concentrations
• A bioenergetics based pollutant accumulation model, including age dependencies for diet composition and energy densities of prey and consumer. Sensitivity analysis indicated metabolic and growth-related parameters to be most critical. The model was fitted to the Lake Ontario water and fish PCB concentrations.

Ecological risk assessments help to anticipate the risks of planned actions, establish regulatory program and research priorities, identify existing problems and provide a scientific basis for regulatory actions. Most importantly, environmental risk assessment provides the basis to prioritize management and regulatory efforts because objective comparisons of a variety of situations can be made.

5.5.7. Ecological indicators – an application and operational methodology

Continuous monitoring of the environment quality is essential to assess the level of different types of toxic waste release. Traditional chemical monitoring methods do not permit the evaluation of the biological consequences of pollutants (e.g. Butterworth et al., 1995 [119]; Munawar et al., 1995 [120]; Markert et al., 2003 [121]). On the other hand, every organism provides a refined system by which long-changes in the quality of environment - with respect to bioavailability of pollutants, and their effect upon the health of organisms - may be determined.

Impact on aquatic ecosystems

The European water management, including wastewater reclamation recycling and reuse has to be fundamentally reorganized on the basis of the legislation in force. One major goal of the WFD is to achieve the good ecological and hydrochemical status of all surface water bodies of the EU. The term of ecological status correspond to the general philosophy of the WFD's integrated approach for evaluating the ecological integrity of water bodies (analysis of hydrochemistry; hydromorphology; and assessment of ecological status based on all 5 aquatic organisms groups: periphyton, phytoplankton, macrophytes, macroinvertebrates, fish).

Table 30 Standards for the ecological water quality assessment of freshwaters according to WFD requirements – algae communities

Bioindicator group	Key ecological metrics	EU analytical standards	Selected national methodological standards & references
ALGAE *Periphyton* *Plankton* WFD's standard monitoring frequency: 3 years Ecological risk assessment - frequency: seasonal sampling (summer-autumn)	**Richness – overall stressor** High species/genera/divisions richness is assumed to indicate high biotic integrity= many species are adapted to the conditions present in the habitat. Species richness is predicted to decrease with increasing pollution **Chlorophyll a concentration – eutrophication symptoms** oligotrophic-mesotrophic boundary: mean of benthic chl a = 2µg cm^{-2}, max. benthic chl a = 7µg cm^{-2}; the mesotrophic-eutrophic boundary: mean of benthic chl a = 6µg cm^{-2}, max of benthic chl a = 20µg cm^{-2} **% Cyanobacteria – eutrophication symptoms** **% Aberrant diatoms – hazard substances** - percentage of diatoms in a sample that have anomalies in striae patterns or frustule shape (e.g. long cells that are bent or cells with indentations); the metric has been positively correlated to heavy metal contamination in streams **% Motile Diatoms - siltation** - percentage of motile diatoms is a siltation index, expressed as the relative abundance of *Navicula* + *Nitzschia* + *Surirella*. These genera are able to crawl towards the surface if they are covered by silt; their abundance is thought to reflect the amount and frequency of siltation; **Tolerance optimum - organic pollution, pH tolerance** calculate a simple diagnostic metric as the sum of the percent relative abundances (range 0-100%) of species that have optimal environmental conditions in extreme environmental conditions, e.g.: % acidobiontic + % acidophilic + % alkalibiontic + % alkaliphilic;% halophilic; % mesosaprobic + % oligosaprobic + % saprophilic;% eutrophic. **Periphyton biomass** - ranges from 0.5 to 2% of total algal biomass; elevated under eutrophication; low biomass: toxic pollutant (also recent spate); **% Community similarity – overall stressor** - percent similarity can be used to compare control and test sites, or average community of a group of control or reference sites with a test site. **Pollution Tolerance Index** - three categories of diatoms according to their tolerance to increased pollution, with species assigned a value of 1 for most tolerant taxa to 3 for relatively sensitive species. **% Live diatoms – heavy sedimentation, siltation** - low percent live diatoms could be due to e.g. heavy sedimentation	**Hydrochemistry** EN 25667-1:1993 Water quality - Sampling - Part 1: Guidance on the design of sampling programmes EN 25667-2:1993 Water quality - Sampling - Part 2: Guidance on sampling techniques **Diatoms sampling**: EN 13946:2003 Water quality - Guidance standard for the routine sampling and pretreatment of benthic diatoms from rivers.	**UK** Trophic Diatom Index Kelly, 1998 [122] **France** L'indice biologique diatomées (IBD) [123]; AFNOR, 2000 [124] **CR** Saprobic Index Sladecek, 1973 [125] **Austria** Diatom Index - Lange-Bertalot, H. 1979 [126]; Trophic Index - Rott et al. 1997 [127]. **USA** Plafkin et al. 1989 [128]; Lowe, R. L. 1974 [129]

Algae as ecological indicators

Trophic classification of freshwater ecosystems from phytoplankton and periphyton (structural and functional metrics, Table 30) are now included in the EC-Water Framework Directive (2000), determining biologically based integrated water quality assessment. As primary producers – algae and

aquatic macrophytes are considered as early-warning system groups, that distinctly react on hydrochemical disturbances (e.g. eutrophication symptoms).
Translating the eutrophication processes into the components of ecological processes and functioning, high productivity is described by the primary production of algae (see the classification for large and mid-sized rivers – Tables 31 and 32) and the resulting nutrient richness of any type of stagnant waters (mainly lakes and lagoons).

Aquatic and terrestrial plants as ecological indicators

Depending on the scope of the environmental analysis, many options for indicative as well as for casual identification of environmental disturbance and changes were defined. The most common metrics, e.g. Ellenberg's indicator values (for each species the range is 1-10) give information on the qualitative relationships between the occurrence of plants and its natural environment, by highlighting major environmental components and determinants of plant habitat preferences and/or tolerance (the different species tolerance for salinity were listed in the last WP2 report). The indicator values for salinity and heavy metals are modified and improved (e.g. Germany: The Trophic Index of Macrophytes [130]; UK: The Mean Trophic Ranking [131]).

Table 31 Criteria for trophic classification of plankton-dominated large rivers [modified from 121]

Classes	Trophic classification	Primary productivity	Chlorophyl-a 90-percentilies [µ/L]	Chlorophyl-a average [µ/L]
I	oligotrophic	very low	3-8	<1-4
I-II	mesotrophic	low to moderate	8-30	3-8
II	eutrophic	moderate	20-100	7-30
II-III	eu- to polytrophic	moderate to high	70-150	25-50
III	polytrophic	high	120-250	50-100
III-IV	poly- to saprotrophic	very high	200-400	>100
IV	saprotrophic	over normative	> 400	> 400

Table 32 Parameters for the trophic characterization of mid-sized rivers and lakes [modified from 121]

Classes	Trophic level	Primary production [g C m^{-2} a^{-1}]	Chlorophyl-a [µ/L]	Total phosporous (TP) [µ/L]
1	ultra-oligotrophic	<10	<1	4..3-7.0
2	oligotrophic	10-25	1-3	7.0-11.6
3	oligo-mesotrophic	25-50	3-10	11.6-19.1
4	mesotrophic	50-100	10-20	19.1-31.5
5	meso-eutrophic	100-175	20-50	31.5-51.9
6	eutrophic	175-300	50-100	51.9-85.6
7	eu-polytrophic	300-500	100-200	85.6-141.2
8	polytrophic	500-800	200-800	141.2-232.8
9	hypertrophic	>800	>800	>232.8

Macroinvertebrates as ecological indicators

The role of macroinvertebrate assemblages in aquatic food web as primary consumers of producers (i.e. periphyton) and decomposers (i.e. heterotrophic bacteria and fungi) and as prey for secondary and tertiary consumers (i.e. fish) make this group of organisms important for the holistic assessment of streams: the community's total integrity of the system and multiple-stress indication (all stressor types and its intensity: chemical contaminations, organic pollution, acidification, morphological and biotic degradation). As a consequence, the biomonitoring methods based on this group (standards for field and laboratory procedures; metrics; indices, etc.; Table 33) were well-developed for all 22 European ecoregions.

Table 33 Standards for the ecological water quality assessment of

Bioindicator group	**Key ecological metrics**	**EU analytical standards**	**Selected national methodological standards & references**
MACRO INVERTE BRATES WFD's standard monitoring frequency: 3 years Ecological risk assessment - frequency: seasonally 3 times per year	In Europe 3 main group of bioassessment methods are developed: 1. Saprobic indices related to parameters: O_2, BOD_5, H_2S, BSB5, NH_4-N, T, pH - saprobic value of i-th indicator species - the quantitative abundance of i-th species 2. Diversity indices Algorithm - a function of abundance, species diversity and ecological tolerance Evenness - uniformity in the distribution of individuals among the species 3. Biotic indices (1) - methods that combined these two approaches –saprobity and diversity basic for all systems: Richness - number of present species Abundance - total number of organisms present in sample Density - total number of organisms present in 1 m^2 In USA the multimetrics system is involved, e.g.(2): Richness measures Taxa Richness No. EPT Ephemeroptera/Plecoptera/Trichoptera Taxa No. Ephemeroptera Taxa No. Plecoptera Taxa No. Trichoptera Taxa No. Diptera taxa No. Chironomidae Taxa Composition measures % EPT Taxa % Ephemeroptera Composition % Tribe Tanytarsini % other Diptera and non-insects % Oligochaeta Tolerance/intolerance measures No. of Intolerant Taxa % Tolerant Organisms % Dominant Taxon No. Intolerant Snail and Mussel species % Sediment Tolerant organisms Feeding measure % Filterers % Grazers and Scrapers % shreeders Habitat measures No. of Clinger Taxa % Clingers Life cycle measures % Multivoltine – having short life cycle (several generation per year) % Univoltine – relatively long-lived (life cycles of 1 or more years)	EN 27828:1994 Water quality - Methods of biological sampling - Guidance on handset sampling of aquatic benthic macro-invertebrates (ISO 7828:1985) EN 28265:1994 Water quality - Design and use of quantitative samplers for benthic macro-invertebrates on stony substrata in shallow freshwaters (ISO 8265:1988) EN ISO 9391:1995 Water quality - Sampling in deep waters for macro-invertebrates - Guidance on the use of colonization, qualitative and quantitative samplers (ISO 9391:1993) EN ISO 8689-1:2000 Water quality - Biological classification of rivers - Part 1: Guidance on the interpretation of biological quality data from surveys of benthic macroinvertebrates. EN ISO 8689-2:2000 Water quality - Biological classification of rivers - Part 2: Guidance on the presentation of biological quality data from surveys of benthic macroinvertebrates.	Saprobic system : Austria, CR Saprobien Water Quality Assessment (1995) [132] Germany Saprobienindex (1992) [133] Diversity Indices: (e.g. Shannon & Weaver, 1949 [134]; Margalef, 1951 [135]) Biotic indices: UK, IR, S RIVPACS River Invertebrate Prediction and Classification System (Armitage et al., 1983 [136]) BMWP/ASPT Average Score Per Taxon (Armitage et al., 1983 [136]) IR Quality Rating System (1983) Austria [137] Ecological Integrity (1997) [138] NL AMOEBA (1991) [139] F Indice Biologique Globale Normalise (IBGN, 1985 France) [123] USA Index of Biotic Integrity (IBI, Plafkin et al. 1989 [128] Rapid Bioassessment Protocol [140] (1999)

(1) The detailed description of the methodology and analytical procedures of most common biomonitoring methods in Europe is available in **www.aqem.de**; **www.eu-star.at**

(2) The new bioassessment system for the biogeographical gradient of Europe [22 ecoregions] and its standarisation across Europe – has been developing in currently on-going the EU project STAR (2002-2005; **www.eu-star.at** – and is partly based also on multi-metrics approach.

Fishes as ecological indicators

Fish are good bioindicators of "ecological integrity" assessment in the catchment-scale, because during their life cycle, the various ecological guilds integrate a wide range of river conditions, including the properties of bed sediment relevant for egg development and the longitudinal integrity for spawning migrations. As a result, they are good indicators of structural properties of river systems (habitat structure and connectivity). Hydrochemical conditions are one of the supplementary determinants of distribution in river systems, related to large scale impact – therefore the application for the assessment of wastewater reuse impact could be profitable by using this group of biota mainly to ecotoxicological studies and standard operative monitoring (see Table 34).

Fish active biomonitoring. Ecotoxicological laboratory experiments under controlled conditions yield valuable basic data on threshold concentrations, dose-specific responses ranging from the molecular to the behavioral level, detoxification processes, mechanism of adaptation, and modulation of toxicity caused by environmental factors. The use of fish as sensors has been recommended for actively monitoring the water quality of industrial effluents and drinking water systems in continuously running and automatically working as a early warning system, with the focus on short-term changes of toxicity.

Morphological anomalies. Most morphological anomalies found in fish concern bone malformations including skeletal anomalies, deformations of opercula, gills and scale-disorientations. Substances affecting neuromuscular functions (e.g. organochlorine, zinc) may also cause spinal deformations. Morphological anomalies are easily detectable indicators of disturbed development or genetic defects and are only one of many symptoms including physiological handicaps. Thus, with rising severity of deformation, larvae and fry will be eliminated rapidly from the population due to natural mortality, predation, and competition which considerably mask contamination effects.

Endocrine disrupture. Many natural and synthetic substances interfere with gonads (hypothalamus-pituitary-gonadal axis), leading to alterations in secondary sexual characteristics and the development of gonads affecting sexual behavior and fertility. These substances mimic or block the action of hormones or affect their synthesis, metabolism and transport. Domestic and industrial effluent may contain a variety of endocrine disruptors (e.g. natural and synthetic estrogens; natural phytosterols; alkyphenols: e.g. non-ionic surfactans, p-nonylphenols; and organochlorine components) however the high persistent mixture of hormone-like substances leading to additive or synergistic effects on the reproduction of fish which are difficult to predict. The vitellogenin induction in males, analyzed in the blood plasma using antibodies, is the most sensitive indicator of the presence of many kinds of EDCs. However, vitellogenin synthesis in vitro has been found to be depressed by polycyclic and halogenated aromatic hydrocarbons. Gonadosomatic indices (reduced testicular growth) and liver-somatic indices (increased relative liver weight) as well as sex ratios are other sensitive indicators of the presence of EDCs, but they are less specific as they may also depend on other environmental factors. The effect of toxicants on thyroid hormone receptors (e.g. hydroxylated PCBs) has not been thoroughly investigated [141, 142].

Table 34 Standards for the ecological water quality assessment of freshwaters according to WFD requirements – fish community

Bioindicator group	Key ecological metrics	EU analytical standards	Selected national methodological standards & references
FISH WFD's standard monitoring frequency: 3 years Ecological risk assessment - frequency: once per year	**Index of Biotic Integrity** score system 1-100 =>58-60 excellent; 48-52 good; 40-44 fair, 28-34 poor, 12-22 very poor *Composition* **Total Number of Species**: total number of resident native fish species; salmonid age classes. **Number and identity of darter species of intolerant species**: number and identity of sculpin species; benthic insectivore species; salmonid juveniles (individuals); number of sculpins (individuals); percentage of round-bodied suckers; sculpin and darter species. **Number and identity of Sunfish species:** number and identity of cyprinid species; water column species; salmonid species; headwater species; sunfish and trout species. **Number of Sucker Species:** adult trout species; minnow species; sucker and catfish species. **Number of Intolerant Species:** sensitive species; amphibian species; presence of brook trout. **% Green Sunfish**: % common carp; % white sucker; % tolerant species; % creek chub; % dace species; **% Omnivores:** % yearling salmonids; generalists; **% Insectivorous Cyprinids:**% insectivores; % specialized insectivores;% juvenile trout; % insectivorous species. **% Top Carnivores:** % catchable salmonids; % catchable trout; % pioneering species; density catchable wild trout. **Numbers of individuals**: density of individuals **Hybrids:** % introduced species; % simple lithophills; simple lithophills species; % native species; % native wild individual; % species with intolerant spawners. **Total Fish Biomass** ***Fish health (pathology, histopathology, deformation)*** **Diseased Individuals** –anomalies (deformations, eroded fins, lesions, tumors) **Tissue contaminants**	EN 14011:2003 Water quality - Sampling of fish with electricity	[143, 144, 145]

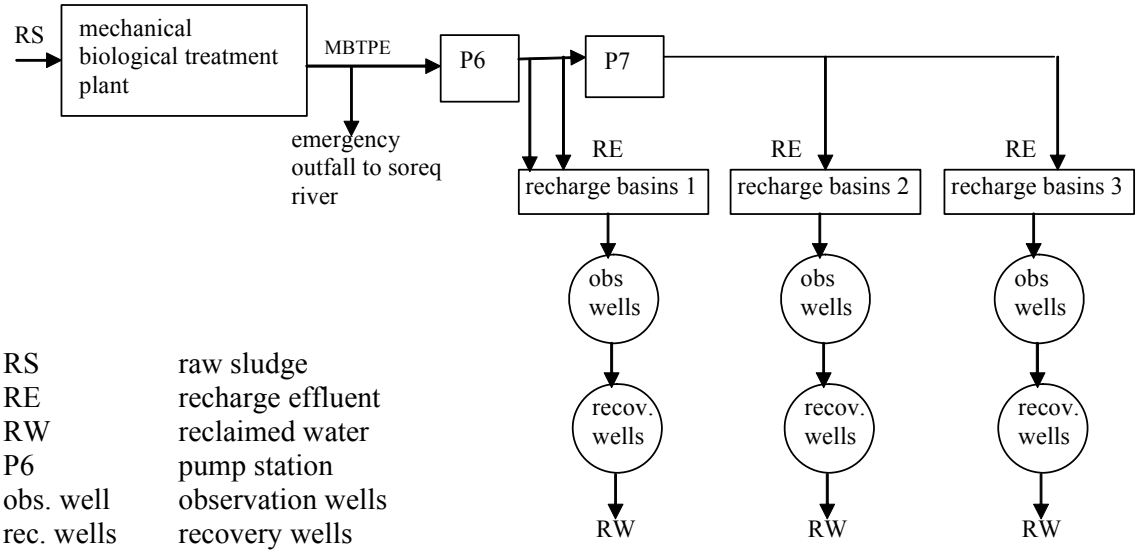
Implications for environmental policy and wastewater management

The application of these modern ecologically oriented assessment methods, based on the integrated assessment (hydrochemistry, hydromorphology and biota composition) are biomonitoring tools that match the scale of current EU policy and legislation and that could guide the use of considerable budgets (cost-effective) to enhance proper ecological functions and processes of surface and groundwater. Early watershed studies and management ignored in-stream contributions to watershed function or combined streams and catchments in a common watershed-stream ecosystem perspective. Nowadays, any river management programme that involves restoration, rehabilitation or preservation should include a decisive concern for ecosystem-functioning consequences. To avoid repeating the ecological river management deterioration of the past, it is necessary to define how the scale-dependent organization of ecosystems and functional reinforcement across scales combine to produce ecological resilience against human impact.

5.6. Case study 1: Dan Region Reclamation Centre, Israel

The Dan Region Reclamation Project (DRRC, Shafdan) is the largest wastewater treatment and reclamation project in Israel. Today it reclaims approximately 135 Mm³/yr of wastewater from the Tel Aviv Metropolitan area and several other neighbouring municipalities. The wastewater is treated in an activated sludge plant with single-stage simultaneous nitrification-denitrification. After secondary treatment, the effluent is then further treated through recharge of the aquifer and subsequent recovery using a special SAT system developed for the Dan Region Reclamation Project (Figure 22). The SAT system improves the effluent quality to an unrestricted irrigation and accidental drinking water quality standard (few hundred cc. accidentally drunk). The water is then pumped 100 km to the south of the country, where it is used to irrigate a variety of crops. Extensive water quality monitoring is performed to keep an efficient and safe wastewater reuse system. Separate monitoring systems and reliability criteria exist for all three parts of the Dan Region Project.

Figure 22 Scheme of the Dan Region Wastewater Treatment Plant and Recharge Basins, Israel



RS	raw sludge
RE	recharge effluent
RW	reclaimed water
P6	pump station
obs. well	observation wells
rec. wells	recovery wells
MBTPE	mechanical biological treatment plant effluent

The influent wastewater collected from the Dan Region consists of ≈ 90% domestic sewage and ≈ 10% industrial sewage. The treatment plant consists of pre-treatment (bar screens, grit removal and scum removal), four anaerobic selectors (for filament control and enhanced biological phosphorous removal), four 55,000 m³ aeration tanks each equipped with 36 horizontal Mammoth rotors and twelve 52 m diameter clarifiers. There are no primary clarifiers. The aeration tanks consist of alternating aerobic and anoxic zones, so that simultaneous nitrification-denitrification occurs together with carbonaceous BOD removal.

5.6.1. Soil Aquifer Treatment (SAT) system

Effluent from the Dan Region WWTP is conveyed to four recharge basins covering a total area of 80 ha. Hydraulic loading to the basins varies between 80 and 150 m/yr, depending on the infiltration capacity of the basins. The infiltration into the groundwater is carried out by alternate flooding and drying, a method

designed to maintain aerobic conditions in the soil aquifer treatment. The effluent percolates vertically through 15 to 30 m of the unsaturated zone, and spreads horizontally (radially) through the saturated zone, outward from the recharge basins to a series of recovery wells surrounding the recharge area. Passage through the soil aquifer filters the effluent and extends the biological treatment by means of the additional contact with oxygen present in the upper soil layers. Physical-chemical processes such as adsorption, ion-exchange and sedimentation also take place in the unsaturated zone. The long retention time in the saturated zone help to destroy harmful bacteria and viruses.

Approximately 100 recovery wells, located 300 to 1,500 m from the recharge basins, pump the recharged water from a depth of 100 to 200 m. In addition to the water quality improvement, the SAT system provides seasonal and multi-year water storage. Water recovered from the SAT system is of extremely high quality and can be used for unrestricted agricultural irrigation.

5.6.2. Effluent distribution system

The water produced from the SAT system is pumped through a single 87 km long pipeline to the southern part of the country, where it is used for unrestricted irrigation. Since this water is used for agricultural purposes, there is a need to regulate the water supply according to seasonal changes. Several local storage reservoirs were built for this purpose, with a total volume of about 15 Mm^3.

5.6.3. Efficiency of the Dan Region reclamation centre concerning chemical values

The efficiency of the Dan Region WWTP and Recharge Centre concerning the chemical values is well monitored. Before the tertiary Soil Aquifer Treatment (SAT) the secondary effluent should comply with the National parameters for Good SAT Treatment; i.e. the turbidity of the secondary effluent should not exceed 8 NTU and TSS should be below 15 mg/L. The annual average of the secondary effluent of the Dan Region WWTP comply with these limits (turbidity 3.1 NTU; TSS < 15 mg/L as it is shown in Table 35).

Table 35 Turbidity and TSS quality effluent in SAT

Parameter	Unit	2dry effluent, before SAT	Annual average
Turbidity	NTU	8	3.1
TSS	mg/L	< 15	7

Once a week the distribution system (pipelines) from the WWTP to the recharge area is chloraminated (10 mg/L) to keep the biofouling to a minimum. The biofouling is otherwise important and causes energy cost increase.

The water obtained from the SAT treatment should comply with the extremely high quality requirements to be used for unrestricted agricultural irrigation and accidental drinking water quality. The quality of water after the SAT system and at the end-user (after the reservoirs and intermediate chlorination) shows very high quality (Table 36).

Table 36 Reclaimed water quality after the SAT system and at the end user of the Dan Region Reclamation Centre

Parameter	Unit	after SAT	End user
BOD	mg/L	0.5	0.5
TSS	mg/L	0	2
COD	mg/L	10 – 20	
UV abs.		35	
Ammonia	mg/L	0.1	0.02
Nitrate	mg/L		32
Total N	mg/L	5 – 10	
Total P	mg/L	0.01 – 0.03	0.02
Detergents	mg/L	0.05 – 0.2	
Total Coliforms	1/100mL	0	
Faecal Coliforms	1/100mL	0	< 1.2
S. faecalis	1/100mL	0	
TB	1/mL	192	

In Table 37 the current recommendations used in the Dan region (unrestricted irrigation) and our proposed quality requirements of the different reuse categories (chapter 0) were compared to the limits achieved by the Dan Region Reclamation Centre. The requirements fit to the (1999) Israel guideline for "unrestricted irrigation", and furthermore fit to the most requirements postulated in our category "private, urban and irrigation". Only the dissolved solids, which should be absent, are measured to be 782 mg/L annual average, and the amount of chloride is at the upper limit (250 mg/L). It does not comply the requirements for the proposed category "indirect aquifer recharge": the annual average contains less dissolved oxygen (5.4 mg/L instead of > 8 mg/L), about 2.5 times too much chloride and nitrate concetration slightly above the upper limit.

5.6.4. Efficiency of the Dan Region reclamation centre concerning salinity

The salinity of the wastewater from the Dan Region near Tel-Aviv, Israel was, till lately, affected by the extensive use of washing powders containing phosphates, borax and sodium chloride besides the extensive use of sodium chloride in ion exchange and in meat processing. In recent years the Ministry of Environment is conducting intensive monitoring of the salinity due to industrial sources. Solutions like replacing sodium by potassium chloride in ion exchange regeneration process, separately pre-treating meat processing water and using low phosphate and borax free washing powders are applied. Already boron concentrations in the incoming wastewater decreased from 0.4–0.5 mg/L in 1999-2001 to 0.2–0.3 mg/L in 2002–2003 (Table 38).

Table 37 Comparison of requirements and efficiency of the Dan Region Reclamation Centre for chemical values [21]

Parameter / Category/ Compound	Unit	1	3	4	Recommentations used in DAN region			DAN region values (Annual average 1998)			
		Private, urban and irrigation	Environment and aquaculture	Indirect aquifer recharge	Unrestricted irrigation	Drinking water quality (1974)	Maximum desired	Biological treatment effluent	After SAT	Pipeline junctions (average of 7)	Reservoirs (average of 9)
pH		6.0 – 9.5	6.0-9.5	7 – 9	6.5 – 8.4	6.5 – 9.5	7.0 – 8.5	8		7.3	7.5
BOD	mg O_2/L	10-20	10-20					9	< 0.5		
COD	mg O_2/L	100	70-100					54	7		
Dissolved oxygen	mg/L	> 0.5	> 3	> 8						4.1	6.0
Dissolved solids	mg/L	absent	absent			1500	800	908		782	
Electrical conductivity	µS/cm	3000	3000	1400				1586		1326	
TSS (105°C)	mg/L	10	10		10			9	0	2.5	5.3
Na	mg/L	150	150-200				0.05			145	
Chloride	mg/L	250	250-400	100		0.6	250	284	283	252	
F (total)	mg/L	1.5 – 2.0	1.5 – 2.0		1	1.7	0.7–1.0	0.54		0.3	
Sulphate	mg/L	500	500	100		437.5	250	84		68	
Total Kjeldahl N	mg/L	15-20						9.8	5.4		
Ammonium-N	mg/L	2-20	1.5	0.2				7	< 0.05	0	
Nitrate-N	mg/L			25		90	45	0.3		28	
Total P	mg/L	2-5	0.2-1					2.9	0.04	0.1	
Surfactant (total)	mg/L	0.5	0.5			1	0.2	0.19		0.1	
Mineral oil	mg/L	0.05	0.05			0.3	0.01	0.2			
As	mg/L	0.1-0.02	0.1-0.02	0.005	0.1			< 0.001		0.001	
Ba	mg/L	10	10					0.267		0.165	
Cd	mg/L	0.005	0.005	0.003	0.01			< 0.0002		0.002	
Cr (total)	mg/L	0.1-0.01	0.1-0.01	0.025				0.004	< 0.003	0.003	
Co	mg/L	0.05	0.05		0.05	1.4	0.05	< 0.003		0.003	
Cu	mg/L	0.2-1.0	0.2-1.0		0.2			< 0.004		0.005	
Hg	mg/L	0.001-0.002	0.001-0.002	0.0005				< 0.0002		0.0001	
Mn	mg/L	0.2	0.2		0.2	0.5	0.05	0 033		0.016	
Ni	mg/L	0.2	0.2	0.01	0.2			0.012		0.005	
Pb	mg/L	0.1	0.1	0.005	5			0.003		0.003	
Phenols (total)	mg/L	0.1	0.1			0.002	0.001	0.004			

The boron concentration higher than 0.4 mg/L in reclaimed water can negatively influence the growth of sensitive crops.

From Table 38 it can be noticed that the improvement in the incoming boron concentration is not immediately reflected in the reclaimed water quality and there is a time delay.

From Table 38 it can be also noticed that the potassium concentration in the reclaimed water is still not stable.

Table 38 Infiltered effluents and reclaimed water quality after the SAT system, Dan Region Reclamation Centre

(Mean Value for all infiltration fields)

Parameters	Units	Infiltered effluents 2001	Reclaimed Water 2002	Infiltered Effluents 2002	Reclaimed Water 2003
Ammonia	mg/L	4.9	0.1	6.03	0.08
Potassium	mg/L	19	17.1	20	14
Sodium	mg/L	201	186	201	166
Calcium	mg/L	85	100	84	97
Magnesium	mg/L	32	26	33	25
Manganese	mg/L	22	84	33	65
Boron	mg/L	0.43	0.41	0.28	0.33
EC	µmhos/cm	1,696	1,542	1,728	1,255
TDS	mg/L	980	855	1030	1024
Chlorides	mg/L	306	235	312	196
Nitrite ($N\text{-}NO_2$)	mg/L	-	0.03	0.94	0.04
Nitrate ($N\text{-}NO_3$)	mg/L	0.4	26.6	0.3	25
Total Kjeldhal	mg/L	9.5	0.49	8.5	0.48
Total P	mg/L	1.7	0.12	2.3	0.03
Sulphate	mg/L	85	78.2	89	73
Alkalinity	mg/L	298	287	309	278
SAR		4.37	1.27	4.8	3.87

The chlorides and EC show a decrease in 2003, probably due to the opening of new infiltration fields (Yavne 4) and the dilution effect of background water.

Another aspect worth mentioning is the increase in manganese ion concentration in the reclaimed water. This is due to manganese dissolution (in certain areas of the aquifer) from the soil structure of the infiltration ponds into the effluent. Its movement is then into the deeper parts of the aquifer and accumulates in the reclaimed water. The manganese that can oxidize (precipitates MnO_2) in the distribution system with the assistance of bacteria found in biofilms can cause clogging of the end-users distribution systems. The manganese arrives in the Third Line water primarily from a small number of wells where the concentration of manganese is very high. Preventive methods like closing some of these wells are being applied to decrease the concentration of manganese in the reclaimed water.

5.6.5. Exposure assessment: breakthrough times of organic micropollutants

Rav-Acha et al. (1996) estimated the breakthrough times of four compounds in groundwater recharged with effluents in the DRRC with a simple model and with specific DRRC-soil and –aquifer characteristics of this area (Table 39). The saturated zone of DRRC aquifer is composed of sand and 10 % clay (mostly montmorilonite) with a negligible organic fraction (about 0.02 %). The relative residential time t_r of the micropollutants depends on the adsorption and desorption of the compounds in the aquifer and on soil

characteristics. In relation to the breakthrough time of a conservative tracer (pentafluorobenzoic acid, assumed to migrate similar to water) t_r can be estimated by the following equation:

$$t_r = K_p * \rho_b * (1 - \varepsilon)$$

where:

t_r = relative residence time of the micropollutant,

K_p = adsorption coefficient of the micropollutant,

ρ_b = average bulk density of aquifer material [g/mL], average value of DRRC = 1.86 and

ε = effective porosity of the aquifer, average value of DRRC = 0.46.

With the specific DRRC aquifer characteristics mentioned in [146] the residential time depends only on the K_{OW} value.

We calculated the hypothetical residential time during aquifer recharge in the Dan Region Reclamation Centre of organic micropollutants that were detected in the observation wells. The lower the t_r values the more likely this compound will occur in the groundwater. The measured analytical data from the observation well showed that the soil aquifer treatment nearly removes all contaminants, but only few organic trace pollutants were determined above the limit of quantification. The t_r values of these compounds were also rather low, so the theoretical breakthrough was likely, too.

Table 39 Calculated relative breakthrough times t_r of micropollutants and detected concentration in the observation well

Compound	Log K_{OW}	t_r	$C_{obs\text{-}well}$ [µg/L] [21, 147]
Alachlor	3.52 [103]	13.56	< 0.1
Atrazine	2.8175 [148]	4.61	0.1
Benzo(a)pyrene	5.97 [149]	967.84	< 0.1
Carbon Tetrachloride	2.83 [103]	4.69	< 0.2
Chlordane	5.8*	716.25	< 0.1
4,4´-DDT	6.91*	5119.66	< 0.1
Heptachlor	6.10 [150]	1218.46	< 0.1
Lindane	4.14 [103]	38.69	< 0.1
Simazine	2.18 [103]	2.17	0.41
Trifluralin	5.34 [103]	317.41	< 0.1
Methoxychlor	5.08 [103, 149]	200.55	< 0.1
Tetrachloro-ethylene	3.40 [103]	11.15	< 0.2
Toluene	2.73 [103]	4.09	< 0.2
1,1,1-Trichloro-ethane	2.49 [103]	3.02	< 0.2
Trichlorophenol	3.69 [103]	17.97	< 0.2
Benzene	2.13 [103]	2.07	0.1
Styrene	2.95 [103]	5.57	< 0.1

* calculated K_{OW} values; n.a. = data not available

5.6.6. Human lifetime input of organic and inorganic pollutants of dan region sat effluent

According to chapter 4.4.1. and in order to calculate reclamation plant effluent concentrations hypothetically used for drinking water reclamation, we calculated the human lifetime input of organic and inorganic wastewater pollutants of the Dan Region Reclamation Centre effluent, i.e. the observation wells (Table 40 and Table 41). The effluent of the DRRC is not considered to be reused for drinking water production but for irrigation and groundwater recharge purposes. Therefore this scenario is calculated as a

worst case scenario if the reclaimed water is used for accidental drinking water purposes. 2 Litres daily intake of reclaimed drinking water is compared to the acceptable daily intake (ADI or RfD, respectively) for a person of 60 kg of body weight (worst case). In addition, the percentage of the RfD value for the daily human intake was calculated.

Table 40 Estimated human daily input of organic pollutants of Dan Region Reclamation Centre effluent (observation well) compared with the RfD values

Organic compound	max detected effluent concentration (observation well) [µg/L]	Daily human intake effluent [mg/day]	RfD [mg/kg BW day] * 60 kg BW	% of RfD
Atrazine	0.1	0.0002	2.1	0.01
Simazine	0.41	0.00082	0.3	0.27
Benzene	0.1	0.0002	0.258	0.08

Table 41 Estimated human daily input of inorganic pollutants of Dan Region Reclamation Centre effluent (observation well) compared with the RfD values (Catch Water Project)

Inorganic compound	The maximum desired Level (Catch Water) [mg/L]	Daily human intake effluent [mg/day]	RfD [mg/kg day] * 60 kg BW	% of RfD
Manganese	0.05	0.0001	0.14	7.E-02
Fluoride	1	0.002	0.06	3.33
Zinc	5	0.01	0.3	3.33
Nitrate	45	0.09	1.6	5.63

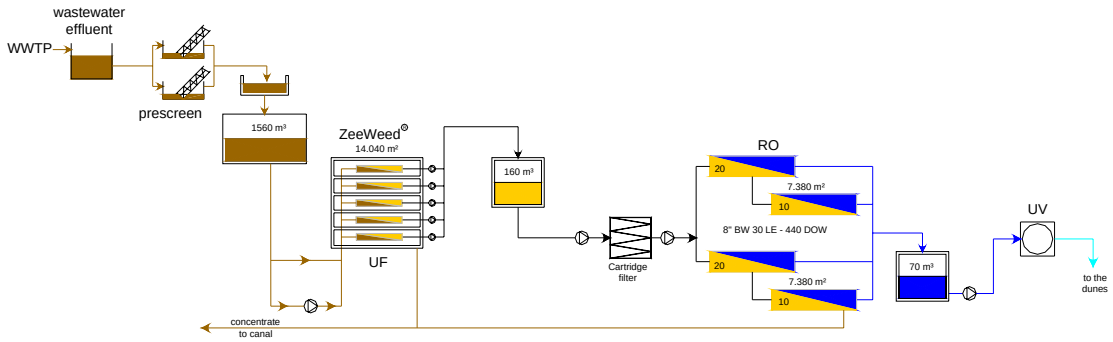
For this calculation the inorganic species seem to be more hazardous than the organic micropollutants. Nevertheless, for the organic micropollutants very much less is known about the chronical risks than of the well known inorganic compounds. Most of the compounds were not even detected in the observation wells, so that the limit of detection concentrations were calculated as maximum effluent concentration (worst case scenario). More effort is needed concerning the chemical analytical data. As well the mixture effects bear unknown effects and more effort according to effect data is required.

In conclusion, the effluent of the Dan Region Reclamation Centre (DRRC) is not supposed to be reused as drinking water without further treatment. The DRRC water is only used as good quality, safe source for agricultural irrigation.

5.7. Case study 2: IWVA Torrele (Wulpen) in Belgium – groundwater recharge

In order to reduce the extraction of natural groundwater for potable water production and hold back the saline intrusion, an average of 285 m³/h tertiary treated municipal effluent is infiltrated in an infiltration pond in the dunes at the Belgian coast. The surface area of the infiltration pond is 18200 m² (Figure 23).

Figure 23 Scheme of the IWVA Torrele Treatment Plant and Groundwater Recharge



The effluent from a municipal activated sludge treatment plant is polished with microfiltration (MF), reverse osmosis (RO) and ultraviolet irradiation (UV) before infiltration. First the produced RO filtrate was reconditioned by a bypass of 10 % of MF-fitrate to match the natural salt content in the dune water. Since May 2004 this bypass was stopped to exclude compounds such as pesticides from the infiltration water. The recharged water is recaptured after a minimum residence time of 40 days in the dune aquifer.

Figure 24 Relation of conductivity of infiltration water and 're-extracted' groundwater

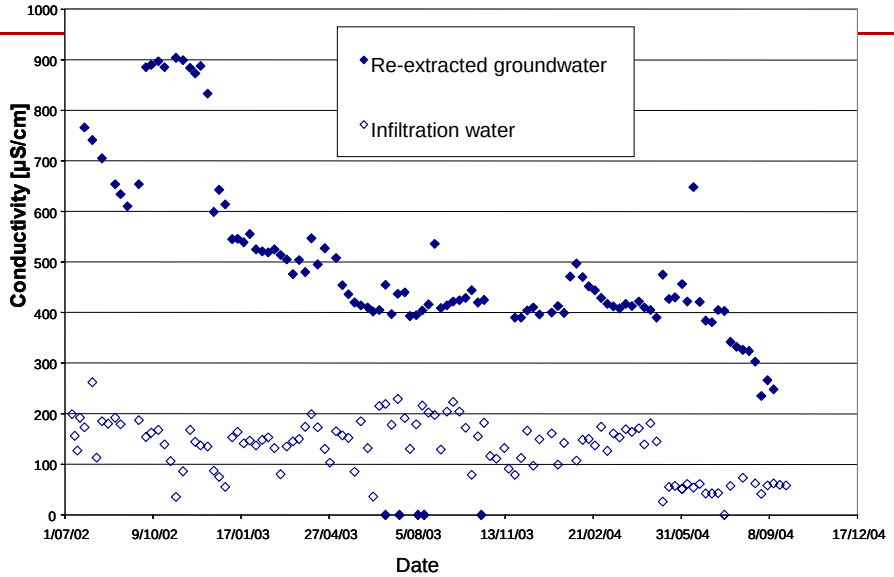


Table 42 Quality of different water between 16th of August 2002 and 1st of October 2002 compared to groundwater before infitration

		MF Filtrate			RO Filtrate			Infiltration water			Groundwater			Groundwater before infiltration		
		Mean	*Min*	*Max*	*Mean*	*Min*	*Max*	*Mean*	*Min*	*Max*	*Mean*	*Min*	*Max*	*Mean*	*Min*	*Max*
Conductivity	µS/cm	1298	473	1591	26	21		185	179	192	638	610	654	882	814	958
pH			7.64	8.22		5.42	7.49		6.08	6.35		7.09	7.29	7.14	7	7.3
Chloride	mg/L	226	70	289	2	2	3	28	26	30	42	42	42	64	53	78
Sulfate	mg/L	106	44	127	<1	<1	3	12	12	17	73	73	73	100	92	106
Total hardness	ºF	32.2	12.8	38.1	<1	<1		4.3	3.9	4.6	31.9	31.9	31.9	36.4	32.9	40
Bicarbonate	ºF	24.3	8.3	32.4	<1	<1		3.2	3.1	3.3	25.1	25.1	25.1	28.6	27.5	29.7
Total Nitrogen	mg/L	10.8	5	16	<2	<2		2	2	2						
Nitrate	mg/L							7.1	2.7	9.6	2.9	2.7	3.3	6.7	<6	7.6
Nitrite	mg/L							0.11	0.05	0.16	0.06	0.05	0.06		<0.1	0.06
Ammonia	mg/L							0.22	0.17	0.27	0.5	0.47	0.53	0.6	<0.4	0.8
Total phosphorous	mg/L	2	1.1	3.2	<0.1	<0.1		0.3	0.2	0.3	0.2	0.2	0.2			
TOC	mg/L	8.9	4.8	10.9	<0.2	<0.2		1.1	1.1	1.1	4	4	4			
Sodium	mg/L	140.7	47.7	186	4.6	4.2	4.9	21.9	20.4	23.4	28.8	28.8	28.8	41.8	26	58
Calcium	mg/L							13	13	13	122	122	122	136	122	149
Magnesium	mg/L							1.7	1.7	1.7	5.4	5.4	5.4	8.2	6.8	9.1
Potassium	mg/L							4.2	4.2	4.2	3.9	3.9	3.9	5.7	4.3	6.9
Fluoride	mg/L							21	21	21	109	109	109			
Silicon	mg/L	21.5	8.9	25.5												
Total THMs	µg/L								<0.1	3.2	0.5	0.5	0.5			
Total pesticides	µg/L							0.024	0.021	0.027	<0.01	<0.01				

Table 43 Quality of different waters between 1st of January 2003 and 1st of June 2004

		MF filtrate			RO filtrate			Infiltration water			Groundwater		
		Mean	*Min*	*Max*	*Mean*	*Min*	*Max*	*Mean*	*Min*	*Max*	*Mean*	*Min*	*Max*
Conductivity	µS/cm	1,290	627	1.798	26	20		146	26	229	448	390	555
pH			7.47	8.11		4.82	7.72		5.62	7.27		7.08	7.57
Chloride	mg/L	225	18	370	2	1	3	20	1	36	42	31	75
Sulfate	mg/L	103	54	139	1	1	3	10	7	17	42	32	56
Total hardness	°F	30.5	11.2	38.8	0.7	0.3	1.0	3.4	1.8	4.9	19.9	14.2	24.9
Bicarbonate	°F	21.1	7.3	28.6	0.8	0.2	1.3	2.4	1.3	3.2	15.8	12.6	20.5
Total nitrogen	mg/L	12.9	3.7	25.4	0.7	0.0	2.2	3.0	2.0	3.9			
Nitrate	mg/L							7.0	1.5	16.1	1.9	1.0	3.4
Nitrite	mg/L							0.12	0.02	0.29	0.03	0.02	0.04
Ammonia	mg/L							0.30	0.07	0.84	0.35	0.22	0.56
Total phosp.	mg/L	1.1	0.3	3.4	<0.1	<0.1	0	<0.1	<0.1	0.2	0.2	<0.1	0.4
TOC	mg/L	7.9	4.8	10.9	<0.2	<0.2	0.4	0.9	0.5	2.0	2.7	2.1	3.9
Sodium	mg/L	160.7	78.1	249.5	2.8	1.0	5.4	16.4	6.0	26.1	24.9	21.6	28.4
Calcium	mg/L							10.1	6	13	72.3	55	94
Magnesium	mg/L							1.4	0.7	1.9	3.3	2.4	4.3
Potassium	mg/L							2.5	0.3	4.2	3.0	2.4	3.7
Fluoride	mg/L							26	17	47	156	132	196
Silicon	mg/L	20.3	8.4	26.0									
Total THM	µg/L							6.0	3.0	17.7	0.8	0.2	2.4
Total pestic.	µg/L										<0.1	<0.1	

Table 44 Quality of different waters between 1st of June 2004 and 15th of September 2004

		MF filtrate			RO filtrate			Infiltration water			Groundwater		
		Mean	*Min*	*Max*	*Mean*	*Min*	*Max*	*Mean*	*Min*	*Max*	*Mean*	*Min*	*Max*
Conductivity	µS/cm	1,246	657	1.505	27	20		55	41	73	363	235	648
pH			7.88	8.22		5.19	5.50		6.27	7.16		7.37	7.68
Chloride	mg/L	233	115	294	3	2	4	3	2	4	34	19	78
Sulfate	mg/L	73	45	84	<1	<1	3	<1	<1		29	25	37
Total hardness	°F	26.9	16.2	31.3	<1	<1		<1	<1		15.8	13.5	19.0
Bicarbonate	°F	23.5	12.3	28.6	<1	<1		2.6	2.1	2.8	14.1	12.9	15.3
Total nitrogen	mg/L	7.8	3.7	11.8	<2	<2		<2	<2				
Nitrate	mg/L							2.3	1.1	3.6	1.5	1.0	2.0
Nitrite	mg/L							0.30	0.03	0.51	0.02	0.02	0.03
Ammonia	mg/L							0.21	0.05	0.61	0.27	0.10	0.74
Total phosp.	mg/L	1.3	0.6	3.0	<0.1	<0.1		<0.1	<0.1		0.2	<0.1	0.2
TOC	mg/L	8.7	6.5	10.1	0.3	0.2	0.4	0.2	0.2	0.2	2.2	2.1	2.3
Sodium	mg/L	160.7	79.6	203.2	3.9	1.7	5.6	12.8	9.6	14.5	21.5	19.8	24.2
Calcium	mg/L							<2	<2		58.7	50	71
Magnesium	mg/L							<0.1	<0.1		2.6	2.3	3.1
Potassium	mg/L							0.7	0.5	0.8	3.0	2.9	3.1
Fluoride	mg/L							<20	<20		147	132	162
Silicon	mg/L	19.1	11.0	22.8									
Total THM	µg/L							4.9	4.4	5.4	<0.15	<0.15	
Total pestic.	µg/L							0.02	0.02	0.02	<0.1	<0.1	

The infiltration occurs in an unconfined sandy aquifer. From the top to the depth of pumping, the sands are fine to medium in size containing shells. There are some small clayey and peaty layers present but their occurrence is irregular. Iron and manganese are present in the groundwater and thus in the soil; peat and clayey sediments probably act as receptors.

It has been observed that since the artificial recharge started the iron and manganese content dropped. When the infiltrated water is extracted coli bacteria no longer are present in the water.

The groundwater level in all wells is monitored every 14 days. Initially the water quality was monitored frequently, but as stabilized very quickly it is now done on a yearly basis only.

Before the start-up of the infiltration project, the conductivity of the groundwater extracted from the 'infiltration area' was 700 to 800 µS/cm with a chloride content of approximately 50 mg/l and a total hardness of around 35 °F (Table 42). The last data were taken 6 weeks before the infiltration started. The first groundwater samples, taken respectively 2 and 3 weeks after start-up of infiltration (8th of July 2002), showed similar results. It took some 2 more weeks before the quality of the extracted groundwater changed. As Figure 24shows, 4 periods can be distinguished: In the start-up period from July 2002 until September 2002 the salt content and further parameters (Table 42) gradually declined to around 600 µS/cm. In September 2002 the infiltration rate was lowered until December 2002 due to technical problems. During this period more groundwater had to be extracted from the infiltration area. The salt content of the extracted groundwater immediately increased to a maximum of just above 900 µS/cm. Since December 2002 a normal infiltration period led to a gradual decline of the salt content until an equilibrium was reached (Table 43). This happened after 5 months of 'normal' infiltration. The hardness of the groundwater followed the same pattern with an equilibrium around 15 °F. Comparing the other parameters listed in Table 42 and 43 the most show a significant decrease from the first infiltraton period to the stable operation from 2003 to 2004. After stopping the bypass of MF filtrate in May 2004 the salt content of the infiltration water consequently dropped further (Table 44)

Comparing the values given for the infiltration water in Table 42 - 44 with the proposed chemical limits in Table 12 given for the application indirect aquifer recharge, no value of the infiltration water failed this limit indicating a good water quality.

During a 5-day period in May 2003 a special investigation was done towards the presence of pesticides into the effluent and the process waters. It was seen that atrazine, simazine and especially diuron were present in the effluent (Table 45). It was observed that MF did not remove the pesticides; however RO removed the pesticides by around 98 %.

Despite the presence of small amounts of pesticides in the infiltration water (Table 45) the pesticide level in the groundwater is below the detection limit (0.01 µg/l). The drinking water quality standards (0.1 µg/l for an individual pesticide and 0.5 µg/l for total pesticides) are met; the recharge system performs as expected and resulted already in softer water adding to the comfort of the customers. In order to improve the water quality further the bypass of MF filtrate was stopped and RO filtrate was pumped to the infiltration pond after dosing sodium hydroxide to correct the pH of the water.

Table 45 Level of pesticides in effluent during a 5 day period [µg/L]

	Day 1	Day 2	Day 3	Day 4	Day 5	Median
Atrazine	0.10	0.08	0.06	0.05	0.04	0.06
Diuron	0.79	0.72	0.53	0.46	0.73	0.72
Simazine	0.38	0.23	0.14	0.09	0.06	0.14

We calculated as a worst case scenario the potential human lifetime input of the three pesticides found in the infiltration water if the IWVA Wulpen effluent is reused for drinking water and no elimination takes place during the treatment process (membrane leakes and no removal during groundwater recharge) (Table 46). For no pesticide the ADI level would be reached by drinking water consumption of 2 L per day and person.

Table 46 Human daily intake of three pesticides of IWVA Wulpen effluent reused for drinking water

Organic compound	max detected effluent concentration [µg/L]	Daily human intake effluent [mg/day]	ADI [mg/kg day] * 60 kg BW	% of RfD
Atrazine	0.08	0.00016	2.10	0.01
Diuron	0.79	0.00158	0.12	1.32
Simazine	0.38	0.00076	0.30	0.25

6. RISK MANAGEMENT CONCEPT (HACCP)

6.1 General introduction

It is proposed that a quantitative risk management system such as the Hazard Analysis at Critical Control Points (HACCP), a preventive risk management system, would be most suitable for applications of wastewater reuse. The HACCP concept originally from NASA space program is now mostly integrated in food quality control management strategies and related guidelines [151]. The full details of a HACCP approach and analysis are well beyond the scope of this study. However, some specific aspects pertaining to the presence and implications of chemical contaminants have been investigated and are described below.

In order to establish an integrated HACCP concept for Reclaimed water reuse treatment processes the most critical hazards have to be first identified. By quantitative chemical risk assessment (QCRA) [152] and quantitative microbial risk assessment (QMRA) the most critical parameters may be identified and the high number of control parameters will be reduced.

The HACCP concept includes seven steps:

- Identification of the scale and of the objective, identification of the hazards, risk analysis and control options
- Identification the critical control points (CCP)
- Establish the control condition
- Establish the control procedure
- Establish the corrective actions when breach of conditions
- Establish revision procedures and efficiency control procedures
- Control of the established procedures and documentation.

6.2 Identification of CCP (Critical control points)

The proper identification of CCPs is an important issue in HACCP because the major efforts in process control and monitoring will be directed towards these steps. In many food-processing operations a single step can be identified that is a major and ultimate barrier (and thus a CCP) to pathogen [153]. Figures 25, 26 and 27 present the basic scheme of a wastewater treatment system. Health sanitation CCP are shown in Figure 25. technical CCP are included in Figure 26 and ecological CCP are presented in Figure 27.

The implantation of a HACCP system has different benefits, because it is a preventive and dynamic system. Its implantation in Reclaimed water chain would have, as a principal benefit, the increase of the safety of this kind of water.

Additionally, HACCP gives other benefits in comparison with traditional quality and inspection control systems, because it focuses its interest in those factors which directly influence above the harmlessness and quality of final product. It is to note the following benefits:

- Reduction of the number of inspections and final product analysis: economical cost is reduced.
- Treated wastewater with better quality.

Figure 25 Basic scheme of a wastewater treatment system. Possible CCP/Health sanitation [154]

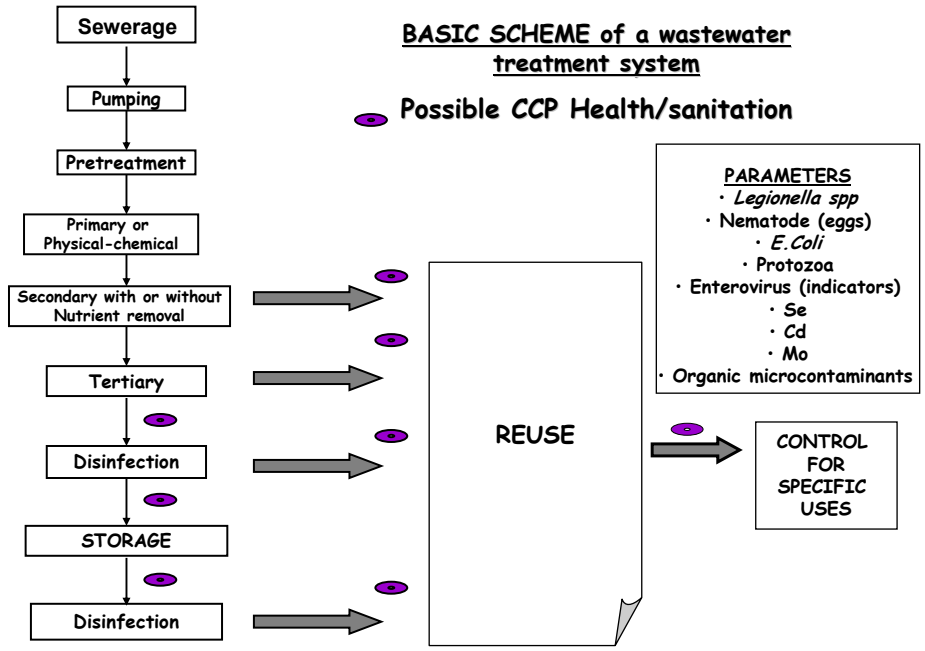
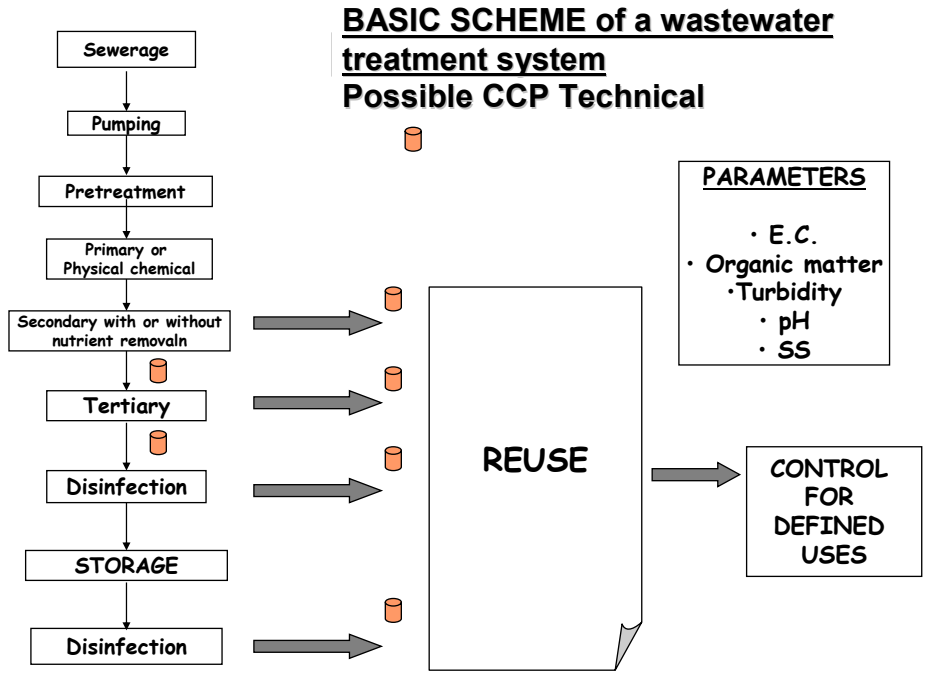


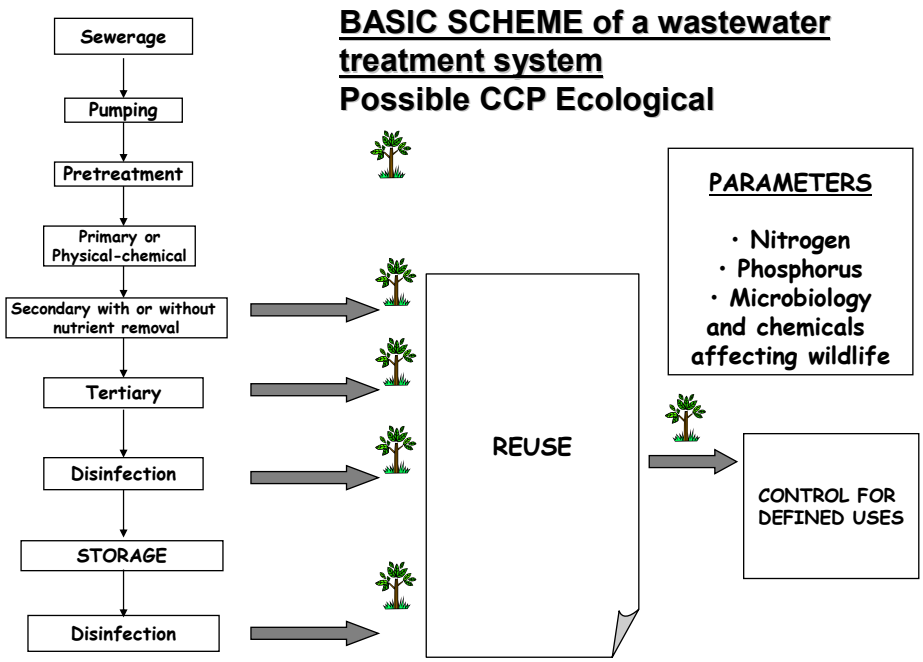
Figure 26 Basic scheme of a wastewater treatment system. Possible CCP Technical [154]



- The system permits to do controls in line, obtaining information in real time. Thus, when dangers appear, corrective actions can be applied immediately.

- It can be an useful tool not only in hygienic quality achievement but even of obtaining quality products.
- It can be used to evaluate wastewater treatment processes.

Figure 27 Basic scheme of a wastewater treatment system. Possible CCP ecological [154]

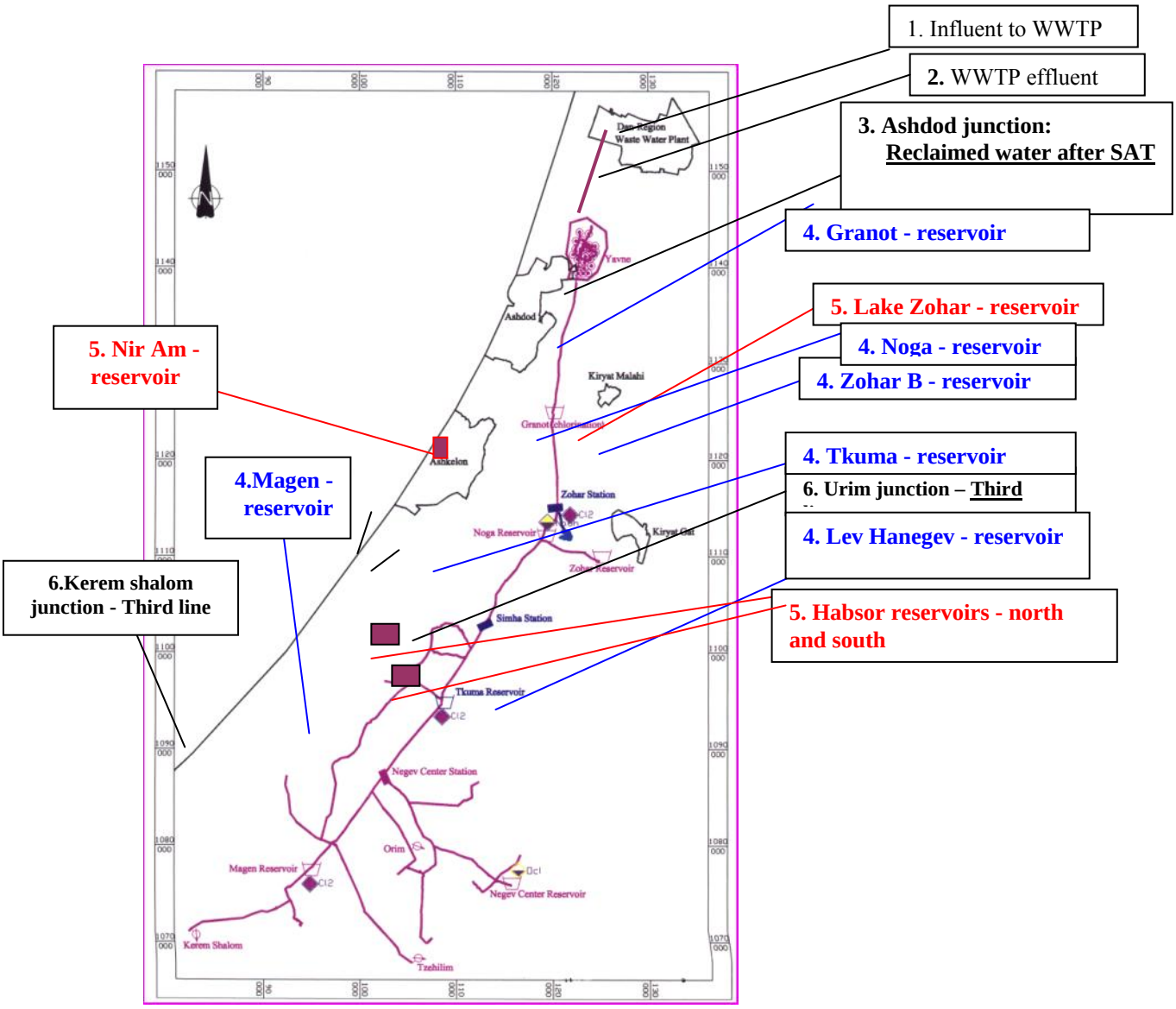


6.2.1. CCP Dan Region reclamation centre Israel

The different critical control points (CCP) can be seen in Figure 39.

1. Influent to WWTP 2. Effluent from WWTP 3.Reclaimed water after SAT
4. Operational reservoirs 5. Seasonal reservoirs 6.Distribution line (to consumer)

Figure 28 Scheme of Dan Region with CCPs



Black – WWTP, Reclaimed water after SAT, Distribution system "The third line"

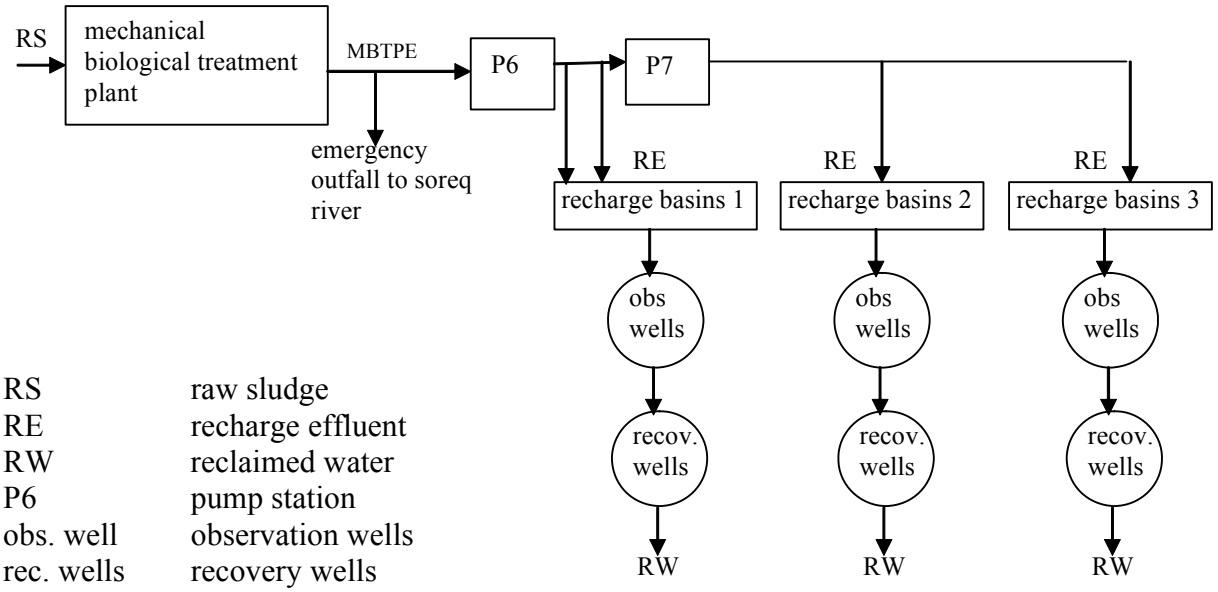
Blue – Operational reservoirs Red – Seasonal reservoirs

The Dan Region Reclamation Project (DRRC, Shafdan) is the largest wastewater treatment and reclamation project in Israel. Today it reclaims approximately 135 Mm^3/yr of wastewater from the Tel Aviv Metropolitan area and several other neighbouring municipalities. The wastewater is treated in an activated sludge plant with single-stage simultaneous nitrification-denitrification. After secondary treatment, the effluent is then further treated through recharge of the aquifer and subsequent recovery using a special SAT system developed for the Dan Region Reclamation Project. The SAT system improves the effluent quality to an unrestricted irrigation and accidental drinking water quality standard. The water is then pumped 100 km to the south of the country, where it is used to irrigate a variety of crops. Extensive water quality monitoring is performed to keep an efficient and safe wastewater reuse

system (Figure 28). Separate monitoring systems and reliability criteria exist for all three parts of the Dan Region Project.

The influent wastewater collected from the Dan Region consists of ≈ 90% domestic sewage and ≈ 10% industrial sewage. The treatment plant consists of pre-treatment (bar screens, grit removal and scum removal), four anaerobic selectors (for filament control and enhanced biological phosphorous removal), four 55,000 m³ aeration tanks each equipped with 36 horizontal Mammoth rotors, and twelve 52 m diameter clarifiers. There are no primary clarifiers. The aeration tanks consist of alternating aerobic and anoxic zones, so that simultaneous nitrification-denitrification occurs together with carbonaceous BOD removal (see Figure 29).

Figure 29 Scheme of the Dan Region Wastewater Treatment Plant and Recharge Basins, Israel



RS	raw sludge
RE	recharge effluent
RW	reclaimed water
P6	pump station
obs. well	observation wells
rec. wells	recovery wells
MBTPE	mechanical biological treatment plant effluent

Effluent from the Dan Region WWTP is conveyed to four recharge basins covering a total area of 80 ha. Hydraulic loading to the basins varies between 80 and 150 m/yr, depending on the infiltration capacity of the basins. The infiltration into the groundwater is carried out by alternate flooding and drying, a method designed to maintain aerobic conditions in the soil aquifer treatment. The effluent percolates vertically through 15 to 30 m of the unsaturated zone, and spreads horizontally (radially) through the saturated zone, outward from the recharge basins to a series of recovery wells surrounding the recharge area. Passage through the soil aquifer filters the effluent and extends the biological treatment by means of the additional contact with oxygen present in the upper soil layers. Physical-chemical processes such as adsorption, ion-exchange and sedimentation also take place in the unsaturated zone. The long retention time in the saturated zone destroys harmful bacteria and viruses.

Approximately 100 recovery wells, located 300 to 1,500 m from the recharge basins, pump the recharged water from a depth of 100 to 200 m. In addition to the water quality improvement, the SAT system provides seasonal and multi-year water storage. Water recovered from the SAT system is of extremely high quality and can be used for unrestricted agricultural irrigation.

Effluent Distribution System –Third Line

The water produced from the SAT system is pumped through a single 87 km long pipeline to the southern part of the country, where it is used for unrestricted irrigation. Since this water is used for agricultural purposes, there is a need to regulate the water supply according to seasonal changes. Several local storage reservoirs were built for this purpose, with a total volume of about 10 Mm³ (seasonal reservoirs and operational reservoirs).

The Third line system includes besides the production wells, for the extraction of the reclaimed water from the aquifer, distribution lines to distribute the water to the customers and supporting systems that include water pumping stations, water disinfection plants, six operational reservoirs along the line each 10,000 -200,000 m^3 and three peripheral reservoirs (1.5 – 3.5 MCM each). There is also a seasonal large reservoir (Zohar "lake") with a volume of 6 MCM.

The main pumping stations of the Third Line are Granot Station located near the Granot reservoir and Almog Station, each with a pumping capacity of 20,000 m^3per hour.

The main disinfection plant (chlorination) is located at the Granot Pumping Station. This plant chlorinates all the reclaimed water of the Third Line. There are other disinfection plants located at the outlets from the seasonal reservoirs.

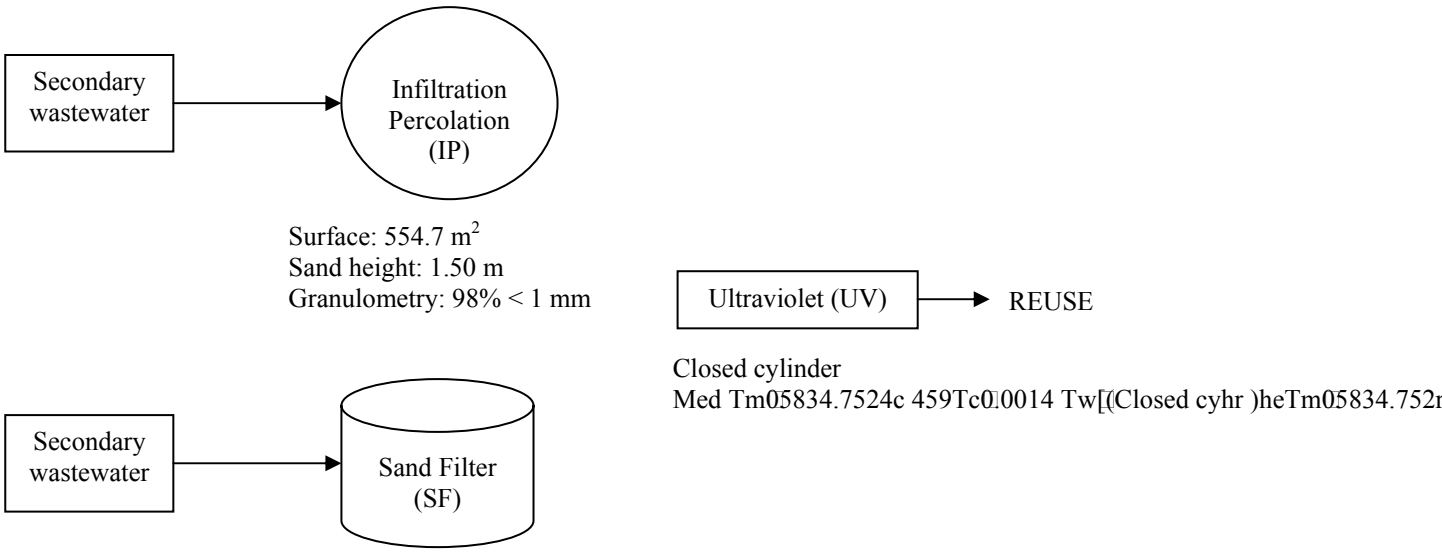
A summary of bacteriological characteristics of the water for all six CCP is given in Table 47 below.

Table 47 Microbiological quality of the water at different stages in the Dan Region Project (5 years average)

Microbiological Parameter	Units	RAW WASTEWATER	EFFLUENT from WWTP	RECLAIMED WATER from SAT#	RECLAIMED WATER from OPERATIONAL RESERVOIRS*	RECLAIMED WATER from SEASONAL RESERVOIRS**	RECLAIMED WATER from DISTRIBUTION LINES***
Total Bacteria	No./1 mL	1.77E+07	5.58E+05	100-1000	40722	10681	3956

The secondary effluent coming from this plant was reclaimed by using different pretreatments before disinfection, as infiltration–percolation and sand filter. Those systems were followed by ultraviolet disinfection among other. Figure 30 represents the diagram flow and main characteristics of the two treatment trains considered (IP + UV; SF + UV).

Figure 30 Diagram flow of Palamós advanced treatment



RESULTS OBTAINED AT PALAMÓS CASE STUDY

Water after IP has suffered, if comparing with the influent, a removal of suspended solids of 64.19% and COD and TOC removal of 31.78 and 56.91%, respectively. On the other hand, BOD_5 is below the detection limit (<5 mg/L). In relation with microbiological parameters, IP improves the microbiological quality as follows: 3.28 Ulog/100 mL of faecal coliforms are removed, somatic coliphages and F-specific bacteriophage determination shows a reduction of 2.52 Ulog/100 mL and 1.62 Ulog/100 mL, respectively. Sand filter removes only suspended solids, and consequently organic matter content is also reduced. This filtration system is mainly not effective for bacteria and virus removal (see Table 48).

UV treatment removes from 2.5 to 3.0 Ulog/100 mL of faecal coliform contents in both cases. It is known that UV efficiency depends on suspended solids contents and turbidity. The best disinfection performances are obtained with the effluents from IP (characterised by its low contents in suspended solids and turbidity as indicated before). Table 48 presents the microbiological results from ultraviolet combined with infiltration-percolation and sand filter systems.

Table 50 Risk evaluation matrix applied to infiltration-percolation + ultraviolet train in the Palamós case study

Parameter	Probability	Consequence seriousness	Risk evaluation
Faecal coliforms	Not much probable	Moderate	6
Somatic coliphage	Not much probable	Important	8
Bacteriophage F-specific	Not much probable	Important	8
Giardia	Not much probable	Important	8
Cryptosporidium	Not much probable	Important	8
		Total risk train	38

Table 51 Risk evaluation matrix applied to sand filter + ultraviolet train in the Palamós case study

Parameter	Probability	Consequence seriousness	Risk evaluation
Faecal coliforms	Probable	Moderate	12
Somatic coliphage	Probable	Important	16
Bacteriophage F-specific	Probable	Important	16
Giardia	Moderate	Important	12
Cryptosporidium	Moderate	Important	12
		Total risk train	68

Risk calculation

Quantitative risk assessment

Quantitative risk assessment models attempt to estimate the risk of infection for an exposed individual. It should be noted that only a fraction of individuals that become infected will actually exhibit symptoms of disease. Investigators attempting to use quantitative risk assessment models for human pathogens have standardized the use of an acceptable risk threshold of 1×10^{-4} for reuse.

Table 52 Quantitative risk assessment applied to Palamós trains

		Faecal coliforms	*Giardia*	*Cryptosporidium*
Model		Beta-Poisson $\alpha = 0.1705$ $\beta = 1.61 \times 10^{6}$	Exponential $r = 0.0198$	Exponential $r = 0.00467$
IP + UV		1.74 (ulog/100mL)	< 1 (cysts/L)	< 1 (oocysts/L)
SF + UV		2.65 (ulog/100mL)	1 (cysts/L)	2 (oocysts/L)
Estimated risk	IP+UV	1.84×10^{-7}	$< 1.96 \times 10^{-2}$	$< 4.66 \times 10^{-3}$
	SF+UV	2.81×10^{-7}	1.96×10^{-2}	9.29×10^{-3}

In order to calculate the quantitative risk assessment of the two Palamós trains considered, models applied are the related to *Giardia* (Exponential), *Cryptosporidium* (Exponential) and *E. coli* (Beta-Poisson). It is

to note that *E. coli* was not determined in this study, but we will considerate that Faecal coliforms contents corresponds to *E. coli*. Table 52 presents the results of the quantitative risk assessment.Faecal coliforms-related estimated risk is below 1 x 10^{-4}; consequently both trains (IP + UV and SF + UV) present an acceptable risk for human health when reusing. *Giardia* and *Cryptosporidium* risk assessment does not present a value below 1 x 10^{-4}; therefore managers must control this parameter in order to avoid health problems in the case of sand filter train. It is to note that the analytical method for *Giardia* and *Cryptosporidium* determination presents a minimum value of < 1 cysts/L and < 1 oocysts/L respectively, which indicates a need to improve the analytical methods or use more sample to strictly accurate health risk calculations.

Epidemiological evidence

Health risks from wastewater use in agriculture have been investigated through quantitative microbial risk analysis (QMRA), as well as through epidemiology [10].

Mara *et al.* (2005) estimated the mean risks per person per year for Rotavirus, *Campylobacter* and *Cryptosporidium* infections resulting from the consumption of 100 g of wastewater-irrigated lettuce on alternate days (unrestricted irrigation). The risks were estimated for one fixed value, 1000, and eight single-log ranges, 1-10 to 10^{7}-10^{8}, of *E. coli* numbers per 100 mL of wastewater [10, 156].

Table 53 Unrestricted irrigation: mean infection risks from the consumption of wastewater-irrigated lettuce estimated by 10,000-trial Monte Carlo simulations [10]*

Wastewater quality (*E. coli* per 100 mL)	**Mean infection risk per person per year**		
	Rotavirus	***Campylobacter***	***Cryptosporidium***
10^{7}-10^{8}	0.99	0.28	0.50
10^{6}-10^{7}	0.65	6.3 x 10^{-2}	6.3 x 10^{-2}
10^{5}-10^{6}	9.7 x 10^{-2}	2.4 x 10^{-3}	6.3 x 10^{-3}
10^{4}-10^{5}	9.6 x 10^{-3}	2.6 x 10^{-4}	6.8 x 10^{-4}
10^{4}	2.2 x 10^{-3}	1.3 x 10^{-4}	4.5 x 10^{-4}
10^{3}-10^{4}	1.0 x 10^{-3}	2.6 x 10^{-5}	3.1 x 10^{-5}
10^{3}	2.2 x 10^{-4}	5.6 x 10^{-6}	1.4 x 10^{-5}
100-1000	8.6 x 10^{-5}	3.1 x 10^{-6}	6.4 x 10^{-6}
10-100	8.0 x 10^{-6}	3.1 x 10^{-7}	6.7 x 10^{-7}
1-10	1.0 x 10^{-6}	3.0 x 10^{-8}	7.0 x 10^{-8}

* 100 g lettuce eaten per person per 2 days; 10-15 mL wastewater remaining on 100 g lettuce after irrigation; 0.1-1 Rotavirus and *Campylobacter*, and 0.01-0.1 oocysts, per 10^{5} *E. coli*; 10^{-2}-10^{-3} Rotavirus and *Campylobacter* die-off, and 0-0.1 oocysts die-off, between harvest and consumption; ID50=6.17 ± 25% and a = 0.253 ± 25% for Rotavirus; ID50= 896 ± 25% and a = 0.145 ± 25% for *Campylobacter*, r= 0.0042 ± 25% for *Cryptosporidium*.

The estimated risks, given in Table 53 are ~10^{-4} – 10^{-6} per person per year (pppy) for a wastewater quality of 1000 *E. coli* per 100 mL, and ~10^{-2} – 10^{-4} pppy for a wastewater quality of 10^{4} – 10^{5} *E. coli* per 100

mL – i.e., the risks derived of eating wastewater-irrigated lettuce are not high than 10^{-2} pppy when the wastewater quality exceeds the level of ≤ 1000 *E. coli* per 100 mL by one order of magnitude or more. Table 52 also shows that (i) when untreated wastewater is used, the risks are very high; and (ii) even where a tolerable risk is set at 10^{-4} pppy, this can be achieved by treating wastewater to 1000 *E. coli* per 100 mL [10].

The mean infection risk from the consumption of wastewater-irrigated lettuce in Palamós case studies (IP + UV and SF + UV trains) is presented in Table 54. IP + UV train presents a Faecal coliforms concentration between 10-100 *E. coli* per 100 mL. SF + UV train shows a Faecal coliforms concentration that corresponds to a range from 100 to 1000 *E. coli* per 100 mL. Both trains present a median risk per person per year less than 10^{-4}; in other words, an acceptable risk.

Table 54 Palamós case study. Application of unrestricted irrigation: mean infection risks from the consumption of wastewater-irrigated lettuce estimated by 10,000-trial Monte Carlo simulations [10]*

Treatment train	Faecal coliform/100 mL (Palamós)	*E. coli* per 100 mL [10]	Mean infection risk per person per year		
			Rotavirus	*Campylobacter*	*Cryptosporidium*
IP + UV	54.95	10-100	8.0×10^{-6}	3.1×10^{-7}	6.7×10^{-7}
SF + UV	446.68	100-1000	8.6×10^{-5}	3.1×10^{-6}	6.4×10^{-6}

Determination of Critical Control Points (CCP)

Ideally, CCP are monitored on-line so that corrective action can be taken by a direct feedback system. Today, microbiological monitoring systems do not fulfil these requirements and are therefore not well suited for monitoring purposes. Figure 31 presents the CCP in Palamòs case study. Table 55 includes a general description of CCP established in Palamòs scheme.

CCP1 corresponds to infiltration-percolation (IP) system outlet. IP must be controlled in order to avoid system clogging, because this phenomenon does not permit the entrance of oxygen to the biofilter. Consequently, biological and physical-chemical parameters present a removal reduction. In order to control the oxygen characteristics, the effluent nitrification must be controlled, because IP has the capacity to transform ammonia into nitrates when the system works correctly (in aerobic conditions). Nitrates content could be an indicator of performance.

CCP2 corresponds to sand filter (SF) system. SF clogging must be controlled by the turbidity control of treated water. In order to avoid this technical problem is important to follow the backwash period and adjusting the programme if necessary.

CCP3 is related to ultraviolet (UV) system for both reclamation lines (IP and SF + UV). The UV radiation is calculated for a specific water quality after filtration systems (IP or SF). If the organic matter removal is lower than designed, the final removal of microorganisms will not comply with the expected values. Consequently, there will be an increase of risk. In order to avoid this problem, the turbidity, the UV transmittance and the lamp current and/or age must be controlled.

CCP4 is related to the network system. It is important to maintain water quality reached after both reclamation trains. In order to control the microorganisms recontamination it will be useful to monitor the pressure through the reclamation network.

Figure 31 CCP in Palamós case study

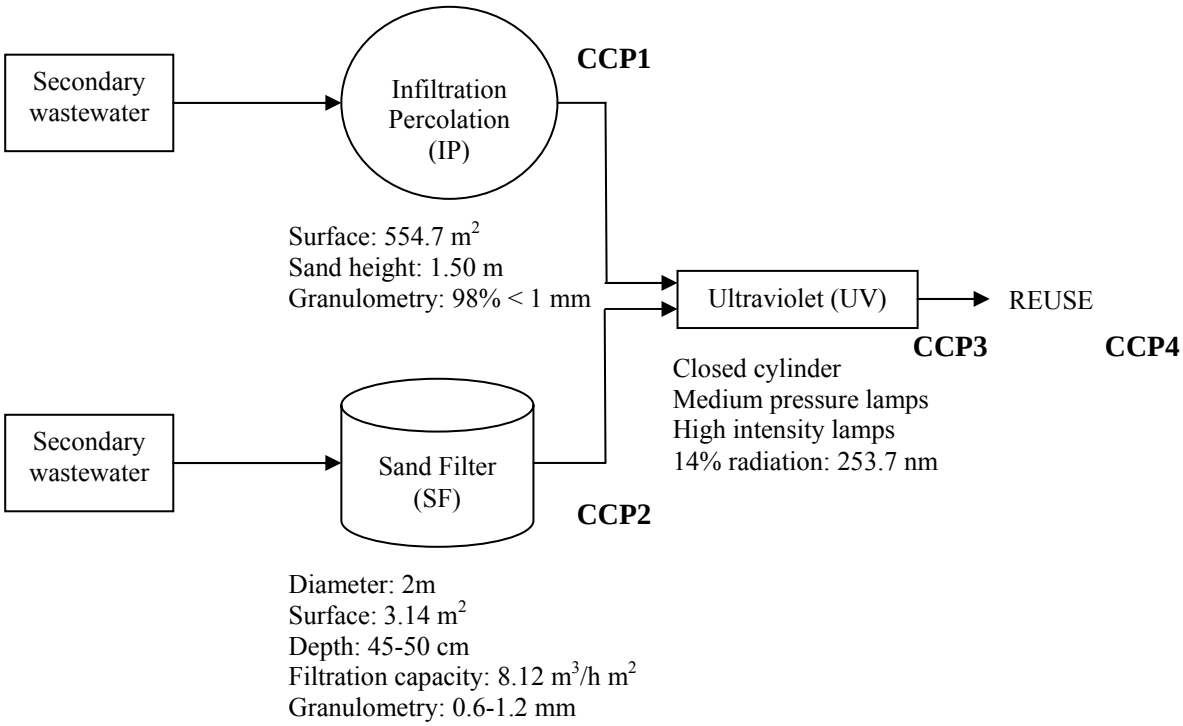


Table 55 Hazards, monitoring strategies and corrective actions per identified CCP

CCP	Process step	Hazard	Monitoring	Corrective action
CCP1	Infiltration-Percolation	Clogging	Effluent nitrification	Stop system/Surface recovery
CCP2	Sand Filter	Clogging	Turbidity	Backwash
CCP3	Ultraviolet	Absorbance Power loss of lamp	Turbidity UV transmittance Lamp current/lamp age	Direct feedback Check IP/SF Check/Replace lamp
CCP4	Distribution network	Recontamination	Pressure changes	Isolate part of the system; purge

7. REFERENCES

1. York, D. W.; Walker-Coleman, L. (2000). Pathogen standards for reclaimed water. *Wat. Environ. Technol.*, 12 (9): 58-61.

2. Bouwer. H. Predicting infiltration and mounding, and managing problem soils. In: Peters. Editor. Artificial Recharge of Groundwater. 1998. pp. 149-154.

3. Höll, K. Wasser. 8 ed. Nutzung im Kreislauf. Hygiene. Analyse und Bewertung. Ed. Grohmann. A. 2002. Berlin. New York. Walter de Gruyter.

4. Grohmann, A.; Hässelbarth, U. and Schwerdtfeger, W. eds. Die Trinkwasserverordnung: Einführung und Erläuterungen für Wasserversorgungsunternehmen und Überwachungsunternehmen. 4 ed.. Ed. Grohmann. A. Erich Schmidt Verlag GmbH & Co. Berlin. 2003.

5. Haarhoff, J. and Van der Merwe, B. (1996). Twenty-five years of wastewater reclamation in Windhoek, Namibia. *Wat. Sci. Tech.*, 33: 25-35.

6. The Council of the European Communities. Bathing water quality Directive 2006/7/EC. Strasbourg, 15th February 2006.

7. CEDEX. Propuesta de calidades mínimas exigidas para la reutilización directa de efluentes depurados según los distintos usos posibles, así como de aspectos relativos a la metodología, frecuencia de muestreo y criterios de cumplimiento de los análisis stablecidos, para incluir en una normativa de carácter estatal. Madrid, Spain. Draft. 1999.

8. Inbar Committee. Quality criteria for reuse of reclaimed water for unrestricted irrigation and disposal to river bodies in Israel. 2003.

9. US EPA. Guidelines for Water Reuse. EPA/625/R-04/108. Environmental Protection Agency and U.S. Agency for International Development. Washington. DC. U.S. 2004.

10. Kamizoulis, G. The new draft WHO guidelines for water reuse in agriculture. Technical Sessions. La integración del agua regenerada en la gestión de los recursos: El papel dinamizador del territorio. Girona, October, 2005.

11. Mahin,T. and Pancorbo, O.. (1999). Wastewater pathogens. *Wat. Environ. Tech.*, 11: 51-55.

12. Pascual L. Calidad microbiológica del aire en diferentes ambientes laborales: aplicación de métodos analíticos moleculares. Ph. D. Thesis. University of Barcelona. 2001.

13. Campos C. Indicadores de contaminación fecal en la reutilización de agua residual regenerada en suelos. Ph. D. Thesis. University of Barcelona. 1999.

14. Olivieri. A.W.; Eisenberg, D.M. and Cooper, R.C. City of San Diego health effects study on potable water reuse. Ed. Asano, T. Wastewater reclamation and reuse. Technomic Publ. Co. 521-580. Lancaster. 1998.

15. Haas, C.; Rose, J.B. and Gerba, C. Quantitative microbial risk assessment. John Wiley & Sons. Inc. USA. 1999.

16. Salgot, M.; Campos. C.; Galofre, B. and Tapias, J.C. (2001). Biological control tools for wastewater reclamation and reuse. A critical review. *Wat. Sci. Tech.*, 43: 195-201.

17. Health guidelines for the use of wastewater in agriculture and aquaculture. WHO Technical Report Series 778. Geneva, Switzerland. 1989.

18. Cooper, R.C. and Olivieri, A.W. Infectious disease concerns in wastewater reuse. In: Ed. Asano. T. Wastewater reclamation and reuse. Technomic Publ. Co.489-520. Lancaster. 1998.

19. Bouwer, H. (2000). Groundwater problems caused by irrigation with sewage effluent. *J. Environ. Health.*, 63: 17-20.

20. Ministero dell´Ambiente e della Tutela del Territorio. Regolamento recante norme techniche per il riutilizzo delle acque reflue. Rome. Decree 12 giugno 2003, n. 185.

21. Mekorot, Israel National Water Co. Dan Region Project – Groundwater Recharge with Municipal water. 2003.

22. Olivieri, A.W.; Eisenberg, D.M.; Cooper, R.C.; Tchobanoglous, G. and Gagliardo, P. (1996). Recycled water - A source of potable water: City of San Diego health effects study. *Wat. Sci. Technol.*, 33: 285-296.

23. Chang, A.C.; Genexing. P.; Page, A.L. and Asano, T. Developing Human Health-related Chemical Guidelines for Reclaimed Water and Sewage Sludge Applications in Agriculture. World Health Organization. Geneva. 2001.

24. Zühlke. S.; Dünnbier, U.; Heberer, T. and Fritz, B. (2004). Analysis of endocrine disrupting steroids: Investigation of their release into the environment and their behavior during bank filtration. *Ground Wat. Monit. R.*, 24: 78-85.

25. D'Ascenzo, G.; Di Corcia, A.; Gentili, A.; Mancini, R.; Mastropasqua, R.; Nazzari, M. and Samperi, R. (2003). Fate of natural estrogen conjugates in municipal sewage transport and treatment facilities. *Sci. Total Environ.*, 302: 199-209.

26. Heberer, T. (2002). Occurrence, fate, and removal of pharmaceutical residues in the aquatic environment: a review of recent research data. *Toxicol. Lett.*, 131: 5-17.

27. Clara, M.; Strenn, B. and Kreuzinger, N. (2004). Carbamazepine as a possible anthropogenic marker in the aquatic environment: investigations on the behaviour of Carbamazepine in wastewater treatment and during groundwater infiltration. *Water Res.*, 38: 947-54.

28. Mitch, W.A. and Sedlak, D.L. (2004). Characterization and fate of N-nitroso-dimethyl-amine precursors in municipal wastewater treatment plants. *Environ. Sci. Technol.*, 38: 1445-54.

29. Mitch, W.A.; Gerecke, A.C. and Sedlak, D.L. (2003). A N-Nitrosodimethylamine (NDMA) precursor analysis for chlorination of water and wastewater. *Wat. Res.*, 37: 3733-41.

30. European Commission, Joint Research Centre, Institute for Environment and Sustainability, Soil and Waste Unit, UMEG Center for Environmental Measurements. Environmental Inventories and Product Safety. Organic contaminants in sewage sludge for agricultural use. 2001.

31. Nadal, M; Schuhmacher, M. and Domingo, J.L. (2004). Levels of PAHs in soil and vegetation samples from Tarragona County, Spain. *Environ. Pollut.*, 132: 1-11.

32. Knepper, T.P.; Barcelo, D.; Lindner, K.; Seel, P.; Reemtsma, T.; Ventura, F.; De Wever, H.; van der Voet, E.; Gehringer, P. and Schonerklee, M. (2004). Removal of persistent polar pollutants through improved treatment of wastewater effluents (P-THREE). *Wat. Sci. Technol.*, 50: 195-202.

33. Rodriguez-Mozaz, S.; de Alda, M.J.L. and Barcelo, D. . Monitoring of estrogens. pesticides and bisphenol A in natural waters and drinking water treatment plants by solid-phase extraction-liquid chromatography-mass spectrometry. *J. Chromatogr. A.*, 1045: 85-92.

34. Vassilakis. I., Tsipi, D. and Scoullos, M. (1998). Determination of a variety of chemical classes of pesticides in surface and ground waters by off-line solid-phase extraction, gas chromatography with electron-capture and nitrogen-phosphorus detection, and high-performance liquid chromatography with post-column derivatization and fluorescence detection. *J. Chromatogr. A.*., 823: 49-58.

35. McCulloch, A. (2002). Trichloroacetic acid in the environment. *Chemosphere*, 47: 667-686.

36. Altissimo, L.; Andreoli, E.; Giacometti, A. and Marcomini, A. (2002). Time trends of 1.1.1-trichloroethane, trichloroethylene, and perchloroethylene in confined and unconfined aquifers of a groundwater system in northern Italy. *Ann Chim*, 92: 61-71.

37. Baronti, C.; Curini, R.; D'Ascenzo, G.; Di Corcia, A.; Gentili, A. and Samperi, R. (2000). Monitoring Natural and Synthetic Estrogens at Activated Sludge Sewage Treatment Plants and in a Receiving River Water. *Environ. Sci. Technol.*,34: 5059-5066.

38. Kuch, H.M. and Ballschmiter, K. (2001). Determination of endocrine-disrupting phenolic compounds and estrogens in surface and drinking water by HRGC-(NCI)-MS in the picogram per liter range. *Environ. Sci. Technol.*, 35: 3201-6.

39. Ternes, T.A.; Stuber, J.; Herrmann, N.; McDowell, D.; Ried, A.; Kampmann, M. and Teiser, B. (2003). Ozonation: a tool for removal of pharmaceuticals, contrast media and musk fragrances from wastewater?. *Wat. Res.*, 37: 1976-82.

40. Rohweder, U. Ergebnisse des Untersuchungsprogramms "Arzneimittel in der Umwelt". Bericht an die 61. Umweltministerkonferenz am 19./20.11.2003 in Hamburg. Untersuchungen im Auftrag des Bund/Länderausschusses für Chemikaliensicherheit (BLAC). Freie und Hansestadt Hamburg. Behörde für Umwelt und Gesundheit. 2003.

41. Motelay-Massei, A.; Ollivon, D.; Garban, B.; Teil, M.J.; Blanchard, M. and Chevreuil, M. (2004). Distribution and spatial trends of PAHs and PCBs in soils in the Seine River basin, France. *Chemosphere*, 55: 555-565.

42. Inquiry Centre at Environment Canada. Assessment Report N-Nitroso-dimethylamine. Quebec, Canada. 2002.

43. Egorov, A.I.; Tereschenko, A.A.; Altshul, L.M.; Vartiainen, T.; Samsonov, D.; LaBrecque, B.; Maki-Paakkanen, J.; Drizhd, N.L. and Ford, T.E. (2003). Exposures to drinking water chlorination by-products in a Russian city. *Int. J. Hyg. Environ. Health.*, 206: 539-51.

44. McCulloch, A. (2003). Chloroform in the environment: occurrence, sources, sinks and effects. *Chemosphere*, 50: 1291-1308.

45. Johnson, A.C. and Sumpter, J.P. (2001). Removal of endocrine-disrupting chemicals in activated sludge treatment works. *Environ. Sci. Technol.*, 35: 4697-4703.

46. Soto, A.M.; Michaelson, C.L.; Prechtl, N.V.; Weill, B.C.; Sonnenschein, C.; Olea-Serrano, F. and Olea, N. (1998). Assays to measure estrogen and androgen agonists and antagonists. *Adv. Exp. Med. Biol.*, 444: 9-23; discussion 23-8.

47. Körner, W.; Bolz, U.; Sussmuth, W.; Hiller, G.; Schuller, W.; Hanf, V. and Hagenmaier, H. (2000). Input/output balance of estrogenic active compounds in a major municipal sewage plant in Germany. *Chemosphere*, 40: 1131-42.

48. The International Cooperation of Rhine Water Power (IAWR). Water power specific guidelines for drinking water treatment close to nature. Nieuwegein. 2003.

49. Rowe, D.R. and Abdel-Magid, I.M. Handbook of wastewater reclamation and reuse. Lewis publishers. 1995.

50. Asano, T. and Levine, A. Wastewater reclamation, recycling and reuse: an introduction. Cap. 1 a: Asano, T. (ed.). Wastewater reclamation and reuse. Technomic Publ. Co.. Lancaster, PA. (1998).

51. NRC-National Research Council. Use of reclaimed water and sludge in food crop production. National Academy Press. Washington, D.C. (1996).

52. Florida Department Environmental Protection. Risk impact statement. Unpublished version. 1998.

53. Water Science and Technology Board and Comission on Geosciences, Environment, and Resources. Ground water recharge using waters of impaired quality. National Academy Press, Washington, D. C. Commitee on Ground Water Recharge. 1994.

54. World Bank. World development report: Investing in health. 1993.

55. World Health Organization Global comparative assessment in the health sector. Disease Burden, expenditures and intervention packages. In Murray, C.J.L. and Lopez, A.D. eds. Switzerland. 1994.

56. Anand, S. and Jonson, K. Disability adjusted life year: A critical review. Harvard center for population and development studies working papers series (95.06). Harvard, Boston. 1995.

57. California. Department of Health Services (2004). Title 22. California Code of Regulations. Division 4. Environmental Health. Chapter 3. Recycling Criteria. Draft 1 Dec 2004.

58. http://www.pitt.edu

59. Homedes, N. (2000). The disability-adjusted life year (DALY) definition, measurement and potential use. HCO. www.worldbank.org/html

60. Hollinghurst, S.; Beban, G. and Bowie, C. Disease by disability adjusted life years. LSE Health, London School of Economics & Political Science. 1999.

61. Chang, A.C.; Page, A.L. and Asano, T. Developing human health-related chemical guidelines for Reclaimed water and sewage sludge applications in agriculture. World Health Organization. Geneva. 1995.

62. U.S. Environmental Protection Agency. Guidelines for water reuse (Manual). EPA/625/R-92/004. Washington, D.C. 1992.

63. Salgot, M (2000). Normas de buena práctica en la reutilización de aguas. *Tecno Ambiente*, 98/99: 5-10.

64. Feachem, D.G.; Bradley, D.J.; Garelick, H. and Mara, D.D. Sanitation and disease: health aspects of excreta and wastewater management. John Wiley and Sons. Bath. 1983.

65. Teunis, P.F.M.; van der Heijden, O.G.; van der Giessen, J.W.B. and Havelaar, A.H. The dose-response relation in human volunteers for gastro-intestinal pathogens. Report no. 284550002. National Insitute of Public Health and the Environment. Bilthoven, The Netherlands. 1996.

66. Metcalf, T.G.; Melnick, J.L. and Estes, M.K. (1995). Environmental virology: from detection of virus inseage and water by isolation to identification by molecular biology – a trip of over 50 years. *Ann. Rev. Microbiol.*, 49: 461-487.

67. Gerba, C. P.; Rose, J.B.; Haas, C. and Crabtree, K.D. (1996). Waterborne rotavirus: a risk assessment. *Wat. Res.*, 30: 2929-2940.

68. Ward, L.R.; Bernstein, D.I. and Young, E.C. (1986). Human rotavirus in volunteers: determination of infectious dose and serological response to infection. *Jour. Infec. Dis.*, 154: 871-877.

69. WA State Water Strategy Securing our water future: A state water strategy for Western Australia. Western Australian Government. (can be accessed from http://www.ourwaterfuture.com.au/community/ statewaterstrategy.asp). February 2003.

70. Bushnak, A.A. Future Strategy for Water Resources Management in Saudi Arabia. Proceedings of A Future Vision for the Saudi Economy Symposium. Riyadh. pp. 37. Oct 12-23, 2003.

71. Shereif, M.M.; El-S Easa, M.; El-Samra, M.I. and Mancy, K.H. (1995). A demonstration of wastewater treatment for reuse applications in fish production and irrigation in Suez, Egypt. *Wat. Sci. Tech.*, 32: 137-144.

72. Dillon, P.J.; Pavelic, P. ; Gerges, N.Z.; Armstrong, D. and Emmett, A.J. Artificial recharge of groundwater. Proceedings of the 2nd International Symposium on Artificial Recharge of Ground Water. pp. 426-435. 1994.

73. Anderson, J.M. (1996). Current water recycling initiatives in Australia: scenarios for the 21st century. *Wat. Sci. Tech.*, 33: 37-43.

74. Guillaume, P. and Xanthoulis, D. (1996). Irrigation of vegetable crops as a means of recycling wastewater: applied to Hesbaye Frost. *Wat. Sci. Tech.*, 33: 317-326.

75. Toze, S. Microbial pathogens in wastewater. CSIRO Land and Water Technical report 1/97. 1997.

76. Toze, S. (1999) PCR and the detection of microbial pathogens in water and wastewaters. *Wat. Res.*, 33: 3545-3556.

77. Wilson, R. and Fujioka, R. (1995). Development of a method to selectively isolate pathogenic Leptospira from environmental samples. *Wat. Sci. Tech.*, 31: 275-282.

78. Fliermans, C. B. (1996). Ecology of *Legionella*: from data to knowledge with a little wisdom. *Microb.l Ecol.*, 32: 203-228.

79. Neumann, M.; Schulze-Röbbecke, R.; Hagenau, C. and Behringer, K. (1997). Comparison of methods for isolation of mycobacteria from water. *Appl. Environ. Microbiol.*, 63: 547-552.

80. Toze, S. and Hanna, J. The Survival Potential of Enteric Microbial Pathogens in a Treated Effluent ASR Project. In: Management of Aquifer Recharge for Sustainability. (ed) P. Dillon. Balkema Publishers. Australia. pgs 139-142. 2002.

81. Carey, C.M.; Lee, H. and Trevors, J.T. (2004). Biology, persistence and detection of *Cryptosporidium parvum* and *Cryptosporidium hominis* oocyst. ***Wat. Res.***, 38: 818-862.

82. Peasey, A.; Blumenthal, U.; Mara, D. and Ruiz-Palacios, G. (2000). A review of policy and standards for wastewater reuse in agriculture: a Latin American perspective. WELL Study. http://www.iboro.ac.uk/well/.

83. Blumenthal, U.J.; Peasey, A.; Ruiz-Palacios, G. and Mara, D.D. (2000). Guidelines for wastewater reuse in agriculture and aquaculture: recommended revisions based on new research evidence. Water and Environmental Health at London and Loughborough. http://www.lboro.ac.uk/well/.

84. Petterson, S.R.; Ashbolt, N.J. and Sharma, A. (2001). Microbial risks from wastewater irrigation of salad crops: a screening-level risk assessment. ***Wat. Environ. Res.***, 72: 667-672.

85. Sheikh, B.; Jaques, R.S. and Cort, R.P. (1987). Reuse of tertiary municipal wastewater effluent for irrigation of raw-eaten food crops: a five year study. *Desalination*, 67: 245-254.

86. Po, M.; Kaercher, J.D. and Nancarrow, B.E. Literature review of factors influencing public perceptions of water reuse. Report to Australian Urban Water Conservation and Reuse Research Program. (2003).

87. Hartley, T.W. Water Reuse: Understanding public perception and participation. Water Environment Research Foundation. Alexandria, Virginia. (2003).

88. Nancarrow, B.; Kaercher, J.D. and Po, M. Community attitudes to water restrictions policies and alternative sources: A longitudinal analysis 1988 – 2002. Australian Research Centre for Water in Society Report. November 2002.

89. Marks, J. The sociology of "disgust" towards the use of reclaimed water. In: Proceedings of 2nd National Conference. Water Recycling Australia. Brisbane, Australia. 1st - 3rd September 2003.

90. Crute, K.; Toze, S.; Pritchard, D. and Penney, N. Proposed research to determine risk from microbial pathogens in grains due to land application of biosolids. Proceedings of Biosolids Specialty II Conference Sydney Australia. 2-3 June 2004.

91. Gennaccaro, A.L.; McLaughlin, M.R.; Quintero-Betancourt, W.; Huffman, D.E. and Rose, J.B. (2003). Infectious *Cryptosporidium parvum* oocysts in final reclaimed effluent. ***Appl. Environ. Microbio.***, 69: 4983-4984.

92. Sakaji, R. H. and Funamizu, N. Microbial risk assessment and its role in the development of wastewater reclamation policy. Cap. 15 a: Asano. T. (ed.). Wastewater reclamation and reuse. Technomic Publ. Co.. Lancaster, PA. (1998).

93. Webb, S.; Ternes, T.; Gibert, M. and Olejniczak, K. (2003). Indirect human exposure to pharmaceuticals via drinking water. ***Toxicol. Lett.***, 142: 157-67.

94. Rajapakse, N.; Silva, E. and Kortenkamp, A. (2002). Combining xenoestrogens at levels below individual no-observed-effect concentrations dramatically enhances steroid hormone action. *Environ Health Perspect.*, 110: 917-21.

95. Silva, E.; Rajapakse, N. and Kortenkamp; A. (2002). Something from "nothing" - eight weak estrogenic chemicals combined at concentrations below NOECs produce significant mixture effects. *Environ. Sci. Technol.*, 36: 1751-6.

96. US EPA. Integrated Risk Information System (IRIS). http://www.epa.gov/iris/intro.htm.

97. Koh, C.H.; Khim, J.S.; Villeneuve, D.L.; Kannan, K. and Giesy, J.P. (2002). Analysis of trace organic contaminants in sediment, pore water, and water samples from Onsan Bay, Korea: instrumental analysis and in vitro gene expression assay. *Environ. Toxicol. Chem.*, 21: 1796-803.

98. Mackay, D. and Paterson, S. (1991). Evaluating the Multimedia Fate of Organic Chemicals - a Level-III Fugacity Model. *Environ. Sci. Technol.*, 25: 427-436.

99. Mackay, D.; W.-Y.S. and K.-C. Ma. Illustrated Handbook of Physical-Chemical Properties and Environmental Fate for Organic Chemicals. V. Lewis Publishers. Florida. 1997.

100. US EPA. EPI Suite (v. 3.12). 2000.

101. Khan, S.J. and Ongerth, J.E. (2004). Modelling of pharmaceutical residues in Australian sewage by quantities of use and fugacity calculations. *Chemosphere*, 54: 355-67.

102. Chang, A.C.; Page, A.L.; Asano, T. and Hespanhol, I. (1996). Developing human health-related chemical guidelines for Reclaimed water irrigation. *Wat. Sci. Technol.*, 33: 463-472.

103. Hansch, C.; Hoekman, D.; Leo, A.; Zhang, L. and Li, P. (1995). The expanding role of quantitative structure-activity relationships (QSAR) in toxicology. *Toxicol. Lett.*, 79: 45-53.

104. Chu, W.H.; Wang, J.Y. and Kao, C.M. (2002). A simplified risk-based approach for process screening in municipal wastewater reclamation and reuse. *Wat. Sci. Technol.*, 47(1): 257-262.

105. Van der Oost, R.; Beyer, B.; Nico, P.E. and Vermeulen, C. (2003). Fish bioaccumulation and biomarkers in environmental risk assessment: a review. *Environ. Toxicol. and Pharm.*, 13: 57-149.

106. Suter, G.W. Ecological Risk Assessment. Lewis Publishers. Boca Raton. FL, USA. p.538. (1993).

107. TerraSys (2002). www.sanexen.com/terrasys/demo/Biotests.htm.

108. Ahtiainen, J. Microbiological tests and measurments in the assessment of harmful substances and pollution. Monographs of the Boreal Environment Research 22. (2002).

109. Bjorklund, L.; Woode, E. and Ullstrand, S. Method for toxicity testing with floating plant *Lemna minor*. ITM Report 7. Institute of applied Environmental Research. Solna, Sweden. 1993.

110. Blinova, I. (2000). The perspective of microbiotests application to surface water monitoring and effluent control in Estonia. *Environ. Toxicol.*, 15(5): 385-389.

111. NRC. Biologic markers in reproductive toxicology. National Academy Press. Washinghton, D.C. (1989).

112. McCarty, L.S. Relationship between toxicity and bioconcentration for some organic chemicals. Parts I & II. In: Kaiser, K.L.E. and Reidel, D. (Eds.). QSAR in Environmental Toxicology. Tensen Scientific Publ. Co.. Dordrecht. The Netherlands. pp. 207-/229. (1987).

113. Spacie, A. and Hamelink, J.L. (1982). Alternative models for describing the bioconcentration of organics in fish. *Environ. Toxicol. Chem.*, 1: 309-320.

114. Loonen, H.; Parsons, J.R. and Govers, H.A.J. (1994a). Effect of sediment on the bioaccumulation of a complex mixture of polychlorinated dibenzo-p-dioxins (PCDDs) and polychlorinated dibenzofurans (PCDFs) by fish. *Chemosphere*, 28: 1433-1446.

115. Clark, T.; Clark, K.; Paterson, S.; Mackay, C. and Norstrom, R.J. (1988). Wildlife monitoring, modelling and fugacity. *Environ. Sci. Technol.*, 22: 120-127.

116. Cabana, G. and Rasmussen, J.B. (1994). Modelling food chain structure and contaminant bioaccumulation using stable nitrogen isotopes. *Nature*, 372: 255-257.

117. Vermeulen, N.P.E. Role of metabolism in chemical toxicity. In: Ioannides, C (Ed.). Cytochromes P450: Metabolic and Toxicological Aspects. CRC Press. Boca Raton. FL, USA. p. 29-53. (1996).

118. Valberg, P.A.; Drivas, P.J.; McCarthy, S. and Watson, A.Y. (1996). Evaluating the health impacts of incinerator emissions. *J. Hazard. Mater.*, 47: 205-227.

119. Butterworth, F.M.; Corkum, L.D. and Guzman-Rincon, J. Biomonitors and Biomarkers as Indicators of Environemntal Change. Environmental Science Research. Vol.50. Plenum Press. 348pp. 1995.

120. Munawar, M.; Hanninen, O.; Roy, S.; Munawar, N.; Karenlampi, L. and Brown, D. Bioindicators of Environmental Health. SPB Academic Publishing. Amsterdam. 265pp. 1995.

121. Markert, B.A.; Breure, A.M. and Zechmeister, H.G. (eds.) Bioindicators & Biomonitors. Principles, concepts and adaptations. Elsevier. Amsterdam-Tokyo. pp.997. 2003.

122. Kelly, M.G. (1998). Use of the trophic diatom index to monitor eutrophication in rivers. *Water Res.*, 32: 236-242.

123. IBGN. In: Indice biologique global normalisé I.B.G.N NF T 90-350. Guide technique. 2ème édition. 2000. **http://213.186.39.110/eaufrance/francais/etudes/pdf/IBGN.pdf**.

124. AFNOR - Association Francese de Normalisation. Essais des eaux. Détermination de l'indice biologique global (IBGN). NFT: 90-350. 2ème édition. 2000.

125. Sladecek,V. (1973). System of Water Quality from the Biological Point of View. *Arch. Hydrobiol. Beih.*, 7: 1-218.

126. Lange-Bertalot, H. Bentische Diatomeen – Gesselschaften in Zuge veränderter Wasserqualitäten im Rhein zwischen Ludwigshafen und Lorch von 1974 bis 1993. Diplomarbeit. Fachbereich Biologie der J.W. Goethe – Universität Frankfurt am Main. 1-157. 1994.

117. Rott, E.; Hofmann, G.; Pall, K.; Pfister, P. and Pipp, E. Indikationslisten für Aufwuchsalgen in österreichischen Fliessgewässern. Teil 1: Saprobielle Indikation. Wasserwirtschaftskataster. Bundesminsterium für Land- und Forstwirtschaft. Stubenring 1. 1010 Wein, Austria. 1997.

128. Plafkin, J.L.; Barbour, M.T. ; Porter, S.K. ; Gross, S.K. and Hughes, R.M. Rapid bioassessment protocols for use in streams and rivers: benthic macroinvertebrates and fish. EPA 440-4-89-001. U.S. Environmental Protection Agency, Office of Water. Washington, D.C. 1989.

129. Lowe, R. L. Environmental Requirements and Pollution Tolerance of Freshwater Diatoms. US. Environmental Protection Agency. EPA-670/4-74-005. Cincinnati, Ohio, USA. 1974.

130. Schneider, S.; Krumpholz, T.; Melzer, A. (2000). Trophäeindikation in Fließgewässern mit Hilfe des TIM (Trophäe-Index Macrophyten) – Erprobung eines neu entwickelten Index im Inniger Bach. *Acta Hydrochim. Hydrobiol.*, 28(5): 241-249.

131. Palmer, M. A. (1992). Incorporating lotic meiofauna into our understanding of faunal transport processes. *Limnol. Oceanogr.* 37: 329–341.

132. Saprobien Water Quality Assessment. ÖNORM M6232: Richtlinien für die ökologische Untersuchung und Bewertung von Fließgewässern. österr. Normungsinstitut. Wien. 1995.

133. Saprobienindex. 1992. In: BAYERISCHES LANDESAMT FÜR WASSERWIRTSCHAFT - Bestimmungsschlüsselfür die Saprobier-DIN-Arten Makroorganismen) 2. Aufl. 274 pp. München.

134. Shannon, C.E. and Weaver, W. The mathematical theory of communication. The University of Illinois Press. Urbana. 19-107. 1949.

135. Margalef, R. (1951). Diversidad de especies en las comunidades naturales. *Publnes. Inst. Biol. Apl.*, 6: 59-72. Barcelona.

136. Armitage, P.D.; D, Moss.; Wright, J.F. and Furse, M.T. (1983). The performance of a new biological water quality score system based on macroinvertebrate over a wide range of unpolluted running-water sites. *Wat. Res.*, 17: 333-347.

137. Quality Rating System. In: Extence, C.A.; Bates, A.J.; Forbes, W.J. and Barham, P.J. (1987). Biologically based water quality management. *Environ. Pollut.*, 45: 221-236.

138. Ecological Integrity. In: Austrian Standards M 6232 - Guidelines for the ecological study and assessment of rivers - Member of the Working Group Österreichisches Normungsinstitut. Vienna. 1997.

139. AMOEBA. In: Van Dijk, G.M. and Marteijn, E.C.L. (eds.). Ecological rehabilitation of the river Rhine, the Netherlands research summary report. 1988-1992. Report nº. 50. 1991.

140. Rapid Bioassessment Protocols for Use in Wadeable Streams and Rivers. Periphyton, Benthic Macroinvertebrates, and Fish. U.S. Environmental Protection Agency, Office of Water, Washington. D.C/EPA-841-B-99-002. 1999.

141. Birkett, J.W. and Lester, J.N. Endocrine disrupters in wastewater and sludge treatment processes. Lewis Publisherrs. IWA Publishing. Washington, D.C. 293pp. 2003.

142. Chovanec, A.; Hofer, R.; Schimer, F. Fish as bioindicators. In: Markert, B.A. Breure, A.M. and Zechmeister, H.G. (eds.) Bioindicators & Biomonitors. Principles, concepts and adaptations. Elsevier. Amsterdam-Tokyo. pp.997. 2003.

143. Karr, J.; Fausch, K.D.; Angermeier, P.L.; Yant, P.R. and Schlosser, I.J. Assessing Biological Integrity in Running Waters. A Method and Its Rationale. Illinois Natural History Survey. Special Publication 5. 1986.

144. Barbour, M.T.; Stribling, J.B. and Karr, J.R. The multimetric approach for establishing biocriteria and measuring biological condition. In:.Davis, W.S & Simon, T.P. (eds). Biological assessment and criteria: tools for water resource planning and decision making. Lewis Publishers. Boca Raton. Florida. 1995.

145. Plafkin, J.L.; Barbour, M.T. ; Porter, S.K. ; Gross, S.K. and Hughes, R.M. Rapid bioassessment protocols for use in streams and rivers: benthic macroinvertebrates and fish. EPA 440-4-89-001. U.S. Environmental Protection Agency, Office of Water. Washington, D.C. 1989.

146. RavAcha, C.; Duchovni, I.; Kanarek, A.; Izakson, N. and Rebhun, M. (1996). Retardation factors and breakthrough times of organic micropollutants in groundwater recharged with effluents. *Wat. Sci. Technol.*, 33: 391-399.

147. Mekorot, Israel National Water Co. The long-term performance of Soil Aquifer Treatment (SAT) for effluent reuse. 2003.

148. US EPA. WSKOWWIN vs. 1.40 Software. 2000.

149. Baker, J.R.; Mihelcic, J.R. and Sabljic, A. (2001). Reliable QSAR for estimating Koc for persistent organic pollutants: correlation with molecular connectivity indices. *Chemosphere*, 45: 213-21.

150. Simpson, C.D.; Wilcock, R.J.; Smith, T.J.; Wilkins, A.L. and Langdon, A.G. (1995). Determination of Octanol-Water Partition-Coefficients for the Major Components of Technical Chlordane. *Bull. Environ. Contam. Toxicol.*, 55: 149-153.

151. WHO. Codex Alimentarius - Guidelines for the Application of the Hazard Analysis and Critical Control Point (HACCP) System. Alinorm 93/13A. Appendix II. Genova. 1993.

152. Hoornstra, E.; Northolt, M.D.; Notermans, S. and Barendsz, A.W. (2001). The use of quantitative risk assessment in HACCP. *Food Control*, 12: 229-234.

153. Dewittinck, T.; Van Houtte, E.; Geenens, D.; Van Hege, K. and Verstraete, W. (2001). HACCP (Hazard analysis and critical control points) to guarantee safe water reuse and drinking water production - a case study. *Wat. Sci. Tech.*, 43(12): 31-38.

154. Salgot, M.; Huertas, E.; Arreciado, A.; Deocón, M. and Pascual, A. El risc associat a la reutilització. II Jornades tècniques de gestió d'estacions depuradores d'aigües residuals. ACA. Barcelona. 2005.

155. Marter, K.; Mullenger, M.; Stevens, M. and Deere, D. Application of HACCP for distribution system protection. AWWA, WQTC Conference. 2003.

156. Mara, D.D.; Sleigh, P.A. and Blumenthal, U.J. (2005). Health risks in wastewater irrigation: comparing estimates from quantitative microbial risk analyses and epidemiological studies. *J. Wat. Health* (submitted).

Acknowledgement

We appreciate the contribution of Emmanuel Van Houtte concerning the IWVA Torrele in Belgium.