Evaluating the epidemiological evidence on the effects of calcium and magnesium in drinking water on cardiovascular disease rates

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A Review for the Global Water Research Coalition

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Contents

1 Executive Summary

In November 2003 in response to requests from a number of member states to provide guidance on the use of desalination for production of drinking water, the World Health Organisation initiated a process to review the potential health effects of nutrients in drinking water. An expert meeting reviewed the scientific literature on a number of relevant issues including the relationship between water hardness and cardiovascular disease risks. The final version of the WHO report *Nutrients in Drinking Water* was published in late 2005. The conclusions and recommendations in this report in relation to cardiovascular disease included:

- *On balance, the hypothesis that consumption of hard water is associated with a somewhat lowered risk of CVD was probably valid, and that magnesium was the more likely contributor of those benefits.*
- *In the revisions of the Guidelines for Drinking-water Quality, WHO should consider the beneficial roles of nutrient minerals including water hardness characteristics.*
- *This subject is of such potential general public health significance that a detailed state-of-the-art review should be prepared prior to consideration in the next revision of the WHO Guidelines for Drinking Water Quality.*

While the WHO review was initiated in the context of concerns over desalinated water, it is evident that any health-based guideline value for minimum levels of calcium and/or magnesium in drinking water should also logically be applied to drinking water from conventional sources. This issue is therefore of importance not only for those water suppliers which utilise desalination processes but also for the broader international water industry. This review was subsequently undertaken for the Global Water Research Coalition to assess the quality of the epidemiological evidence on this issue.

The scope of this review is limited to epidemiological studies of analytical design published in the English language literature since 1979 which have examined the relationship between water quality parameters and cardiovascular health outcomes. A summary of epidemiological study designs and factors affecting the interpretation of epidemiological evidence is included to assist the reader.

The term “cardiovascular diseases” refers to a diverse group of diseases and conditions involving the heart and blood vessels. The classes of cardiovascular disease most frequently studied in relation to water quality are Acute Myocardial Infarction (AMI, more commonly known as heart attack) or the broader category of Coronary Heart Disease (including AMI and Angina). A few studies have investigated cerebrovascular diseases (specifically Stroke). Several factors are known to be important in determining the risk for development of these cardiovascular diseases. Factors associated with an increased risk of AMI include smoking, abnormal blood lipid levels, a history of hypertension (high blood pressure), diabetes, abdominal obesity, and psychosocial factors (eg stress at work or home, depression). Factors associated with a decreased risk of AMI include daily consumption of fruits and vegetables, regular alcohol consumption, and regular physical activity. A number of these risk factors are influenced by the behavioural choices of individual people, their socioeconomic status and the nature of the health care system that exists in their society.

Mortality from AMI and other cardiovascular events is influenced by the nature of medical assistance given to the victim and the rapidity with which it is administered after the event.

This review examined three ecological studies which used small scale geographic measures to classify population exposures, two cohort studies and seven case-control studies undertaken on the issue of water hardness and cardiovascular disease and published since 1979. It was found that the studies did not provide consistent or strong support for the protective effect of magnesium or water hardness. Most of the studies on this topic must be considered as being of low quality as they have not assessed the impact of individual risk factors or socioeconomic factors on the apparent relationship between water quality parameters and cardiovascular disease incidence or mortality rates. Among the studies examining mortality, none have attempted to account for the effects of medical intervention on death rates.

The epidemiological studies which have included adjustment for socioeconomic factors and/or individual risk factors in their statistical analysis have generally not observed a significant effect of water quality. This suggests that the apparent protective effect of water hardness commonly seen in ecological studies may be mostly or entirely attributable to differences in the distribution of socioeconomic factors and/or individual risk factors in the populations being compared, rather than any effect of water quality. In addition, some epidemiological studies have raised the possibility of alternative explanations relating to either protective or adverse effects of other substances in water.

In conclusion, the current body of epidemiological evidence contains a relatively small number of analytical studies, the quality of most of these studies is poor, and the findings are inconsistent. The available evidence is not sufficiently strong to infer that low levels of calcium and/or magnesium in drinking water are significant factors in the causation of cardiovascular disease.

This review also highlighted a number of important omissions and design flaws in past epidemiological studies which need to be addressed if future studies are to make a worthwhile contribution to the knowledge base on this issue. Collaborative projects between water researchers and cardiovascular researchers are likely to be the most effective way to overcome the limitations of past research and rapidly advance our knowledge and understanding of this important public health issue.

2 Introduction

2.1 Background

In November 2003, the World Health Organisation (WHO) convened an Expert Meeting on '*Nutrient minerals in drinking-water and the potential health consequences of consumption of demineralized and remineralized and altered mineral content drinking-water'* as part of the rolling revision of the Guidelines for Drinking-water Quality. The workshop was prompted by requests from a number of member states to provide guidance on the use of desalination for production of drinking water as a consequence of the rapidly increasing adoption of this technology worldwide. The meeting addressed health considerations potentially arising from long-term consumption of water derived from water that has undergone major alteration in its mineral content, such that it must be remineralized to avoid corrosion of piped distribution systems. Other topics examined included the role of drinking water as a source of trace minerals, the relationship between water hardness and cardiovascular disease risks, and the dental health impacts of fluoride in drinking water.

The Expert Meeting produced thirteen draft papers summarising the scientific evidence relevant to the issues assessed, and provided the collective opinion of the group in a consensus report. With respect to the issue of cardiovascular disease, the draft consensus report stated that:

> *'The group concluded that there is sufficient evidence of an inverse relationship between calcium and magnesium concentrations in drinking water and ischaemic heart disease mortality, and that consumption of water containing calcium and magnesium, and therefore also the reintroduction of Mg and Ca into demineralised water in the remineralization process would likely provide health benefits in those consumer populations'.*

While the WHO review was initiated in the context of concerns over desalinated water, it is evident that any health-based guideline value for minimum levels of calcium and/or magnesium in drinking water should also logically be applied to drinking water derived from conventional surface and groundwater sources. This issue is therefore of importance not only for those water suppliers which utilise desalination processes but also for the broader international water industry. This review was undertaken for the Global Water Research Coalition to assess the quality of the epidemiological evidence on this issue. Separate reviews were also undertaken on biochemical/biomedical aspects, and reasons and criteria for softening and conditioning of drinking water.

The final version of the WHO report *Nutrients in Drinking Water* has published in late 2005. The conclusions and recommendations in this report in relation to cardiovascular disease were largely unchanged from the draft report and included:

- *On balance, the hypothesis that consumption of hard water is associated with a somewhat lowered risk of CVD was probably valid, and that magnesium was the more likely contributor of those benefits.*
- *Studies should evaluate a number of potentially relevant health outcomes (e.g., renal stone formation, CVD, hypertension incidence, osteoporosis, stroke, mineral balance,*

mineral nutritional deficiencies). Exposure assessments should include analyses for calcium, magnesium, and trace elements.

- *Studies should evaluate the issue of whether there are adverse health consequences associated with consumption of soft corrosive water due to extraction of metals from pipe. There should also be additional studies to determine whether and how softened waters differ in that respect from soft waters.*
- *Clinical trials of people at high risk of heart attacks and other illnesses such as osteoporosis, should be conducted to assess the potential benefits of mineral supplementation. Results of previous studies have been inconsistent.*
- *In the revisions of the Guidelines for Drinking-water Quality, WHO should consider the beneficial roles of nutrient minerals including water hardness characteristics.*
- *This subject is of such potential general public health significance that a detailed state-of-the-art review should be prepared prior to consideration in the next revision of the WHO Guidelines for Drinking Water Quality.*

2.2 Scope Of This Report

This report reviews epidemiological studies of analytical design published since 1979 which have examined the relationship between water quality parameters and cardiovascular health outcomes. The review is limited to relevant English-language articles identified from searches of the Medline and Current Contents databases. We did not attempt to identify unpublished studies, studies published in the grey literature or studies published in sources not indexed by the major databases mentioned above. Accordingly, this review is limited in scope but addresses the analytical studies covered by the WHO review of epidemiological studies 1979-2004 (Monarca *et al.* 2005).

3 Epidemiological Study Designs

A number of different epidemiological study designs may be used to investigate the causes of human disease and the factors which increase or decrease disease risks. Each of these designs has inherent strengths and weaknesses, and it is important to appreciate these when assessing the results of such studies. The choice of the appropriate design depends on the nature of the question under study, and the availability of data and resources.

Epidemiological studies can be broadly divided into observational and experimental studies. In an observational study, the researcher collects information about a group of subjects comparing the effect of an event /exposure on the group but does not intervene nor influence the event/exposure. By contrast in an experimental study, the researcher deliberately influences events and investigates the effects of the intervention.

3.1 Observational Studies

3.1.1 Cross-sectional Studies

Cross-sectional studies are essentially surveys of the number of existing cases of a disease in a population group at a given point in time (prevalence), and the current exposure status of individuals to potential risk factors. Cross-sectional studies cannot give information about changes in exposures or disease development over time.

These studies can provide only limited information on associations between risk factors and disease, but they may be useful in determining the frequency of diseases and exposures in populations. Cross-sectional studies are relatively straightforward to perform and vary in cost according to the type of information collected.

3.1.2 Ecological Studies

Ecological studies are observational studies, which compare the rates of disease in two or more populations. These populations are defined in terms of geographic boundaries, which are inferred to correspond to differing levels of exposure to the factor of interest. The most commonly used measures of disease are the number of newly arising cases (incidence), or (where applicable) the number of deaths from the disease in a particular time period. The rate of disease for each population is then calculated from these measures, generally as a rate per 10,000 population per year.

It is important that the rate calculation takes into account any differences in the demographic structure of the two populations. As the likelihood of disease often varies in different age groups or between the sexes, disease rates must be adjusted to account for any differences in the age and sex distribution in order to produce comparable figures. Adjustments should also be made for other factors which are known or suspected to influence the rate of disease, for example differences in socioeconomic status between the populations.

Ecological studies do not attempt to assess individual exposures, instead the level of exposure of an entire population group is inferred from their place of residence. Thus it is not

possible to determine whether the individuals who developed the disease actually had higher or lower exposure to the suspected causative agent than individuals who did not develop the disease. These studies are also unable to take into account potential confounding or modifying factors which affect disease rates.

Rates of disease or mortality may be derived from existing disease specific registries (e.g. cancer registries), death certificates, records of hospital presentations, treatment or discharges, or other databases (e.g. medical insurance organisations). Such data sources may vary considerably in their accuracy and comprehensiveness, and this will affect the resultant estimates of disease rates. Ecological studies are sometimes used to assess changes in disease rates and population exposures over time (time-series studies).

Ecological studies have the advantage of being relatively inexpensive and rapid to conduct as they are able to utilise existing databases or record sources. However the lack of individual exposure estimates, inability to measure confounding factors, and variability in the quality of data sources means that the associations between exposure and disease observed in such studies must be regarded as very tentative. Ecological studies are the first step in generating a hypothesis that an association between an exposure and a disease may exist.

The results of ecological studies cannot be used to draw conclusions that the observed association between a hypothesized risk factor and a disease represents a cause and effect relationship. There are many examples in epidemiology where a strong association found in one or more ecological studies could not be substantiated when subsequent studies of stronger design were performed. This type of erroneous conclusion about causation is commonly known as the "ecological fallacy".

3.1.3 Cohort Studies

A cohort study is an observational study where a group of individuals (the cohort) are followed over a period of time. At the beginning of the study the group is defined on the basis of presence or absence of exposure to a suspected risk factor for a disease. At this time all potential subjects must be free of the disease under investigation. Eligible participants are then followed over a period of time to assess the occurrence of the disease being studied. The cohort may be subdivided at the outset into groups with different characteristics, and cohort studies may be prospective or historical:

Prospective cohort study - a group of subjects is identified and followed prospectively perhaps for many years with detailed information on exposures and disease development collected regularly.

Historical cohort study - a past cohort is identified and their exposure experience up to the present is obtained. This is a less common type of study because detailed records are often not available. This lack of data makes it difficult to be sure about an individual's level of exposure or sometimes even whether they did or did not develop the disease under study. Historical cohort studies are most commonly used for investigation of occupational exposures using employment records. Retrospective cohorts may also be identified through census records. For example if a person with the same name, gender and date of birth lived

at a particular address for consecutive census periods it may be assumed that the person has been living there continuously during the intervening time.

The strengths of a prospective cohort study are that the level of exposure of individuals can be measured, multiple health outcomes can be followed in the same study, and disease incidence can be accurately calculated. However, due to their nature cohort studies must continue until a fair proportion of the study group develops the outcome, and good follow up rates for participants are required. For this reason cohort studies can take a long time and can be expensive, particularly when rare outcomes are studied. Cohort studies are also subject to surveillance bias (an apparent increase in disease incidence due to close observation) and confounding. If used properly however, a cohort study can be a valuable strategy to obtain a valid estimate of the association between an exposure and disease.

3.1.4 Case-Control Studies

Case-control studies are observational studies involving a comparison between a representative sample of people with a disease (cases) and another group without the disease (controls). A questionnaire is generally administered to both groups and if it is found that a certain factor is more common among the cases than among the controls, this suggests that this factor may be associated with the development of the disease. However this finding does not prove that the factor causes the disease: a case-control study provides only evidence of an association and a variety of other criteria must be met before one can conclude that a causal association is likely.

Case-control studies may not always assess individual exposures by questionnaire - instead records such as place of residence at the time of diagnosis may be used to infer exposure to an environmental factor, or employment records may be used for occupational exposures. Such inferences may be less accurate than information obtained by individual questionnaire. However, exposure assessment by questionnaire also has a number of pitfalls. Recall of possible exposures from several decades earlier may be unreliable. If the disease under study is terminal or severely debilitating, the affected person may have spent some time considering possible causes and may report past exposures in much more detail than control subjects. Media reports or advice obtained on suspected risk factors during diagnosis may also trigger exaggerated emphasis on particular exposures.

Case-control studies are used primarily to identify risk factors for disease and to attempt to find the causes of disease. The advantage of these studies is that they can be undertaken quickly and with relatively little expense. Unlike cohort studies it is not necessary to follow a large number of people for a long period waiting for disease to develop in a few. Case-control studies also allow the identification of risk factors (and/or causes) of rare diseases which are difficult to study with a cohort design (because so few cases occur). They allow the relationship between many potential risk factors and a single disease to be examined, whereas cohort studies can examine the relationship between a single risk factor and many diseases.

However, case-control studies have several disadvantages which restrict their applicability and reliability. The most important of these is their potential to generate spurious associations because of various biases that can be almost impossible to eliminate:

- Case selection bias occurs when the cases studied are only a subset of all cases and this subset is unrepresentative with respect to the risk factors being studied.
- Information bias refers to the potential to create or conceal a relationship between an exposure and a disease as a result of an error in measuring exposure. Because of the reliance on the provision of accurate information from the memory of cases and controls, this is a potentially important form of bias that can easily produce suggestive evidence of a weak association. Even if care is taken to ensure comparability of information sources, it is difficult in case-control studies to ensure the absence of bias resulting from differential recall of information.
- Control selection bias may arise in the selection of an appropriate control group. Ideally such a group should be selected from the same population or sub-population as the cases, but in practice this may be difficult to achieve.
- Confounding and modifying factors may also cause problems with case-control studies but the effect of these can usually be eliminated if they are identified beforehand and measured during the study.

Finally, because so many possible risk factors can be studied, the problem of positive findings arising by chance is a common occurrence and requires great care to be taken in interpreting studies where an unexpected finding (i.e. one not hypothesised in advance) has arisen.

3.2 Intervention Studies

3.2.1 Randomised Controlled Trials

Randomised controlled trials are intervention studies in which participants are randomly assigned to groups which receive or do not receive an experimental therapy or intervention. The results are assessed by rigorous comparison of outcome measures between the groups (e.g. rates and severity of disease, recovery or death).

This type of study frequently involves the use of “blinding” where the intervention/treatment assignment is unknown to both the participant and the researcher (double blind), or unknown to the participant (single blind). The blinded design requires the use of a sham or placebo treatment in one group of participants and aims to remove the potential for reporting bias by either the participant or the researcher. In some cases, a placebo cannot be used as the control for ethical reasons - for example, in a trial of a new drug it is not ethical to assign the control group to placebo treatment if an accepted drug treatment already exists. In such instances, the control group is assigned to the conventional treatment, and a comparison is made between new and old treatments rather than new and placebo treatment. Blinding is also ruled out where the intervention is clearly apparent to the participant, for example where changes in behaviour are required.

In the context of exposure to suspected environmental risk factors, it is generally difficult to implement randomised controlled trials, however it may be feasible to use this design where

the exposure of one group can be reduced below usual levels. Such an intervention would most likely be considered ethically acceptable, whereas deliberately increasing exposure to a potentially harmful substance would not.

The double blind randomised placebo controlled trial is generally regarded as the most scientifically rigorous method of hypothesis testing in epidemiology, and is often referred to as the "gold standard" in such studies. This is because this study design minimises a number of possible sources of bias which might influence the results, and so provides the most credible evidence of an effect of the intervention (or lack of effect).

3.3 Confounding And Modifying Factors

A major problem of observational studies is the difficulty of isolating the effect of the risk factor of interest from other possible causes of the disease which it may be linked to. Confounding is defined as the occurrence of a spurious linkage relationship between an exposure and a disease that results because the exposure of interest is linked with another variable (confounder) that is the true cause of the disease. For example if a disease is caused by Factor A, and Factor A and Factor B tend to occur together, then an association will be seen between disease risk and Factor B even though this is not the causative agent.

When potential confounders have been identified and considered in advance, their influence can generally be removed by matching cases and controls for exposure to the confounding factor, or by the use of appropriate statistical techniques during analysis. However it is not possible to allow for unknown confounders that have not yet been identified as the cause of the disease in question.

Modifying factors are factors which influence the disease risk or outcome without necessarily being linked to the exposure being studied. If these factors are not measured and included in analysis, and they happen to be unequally distributed between the groups under observation, they may have a significant influence on the result. In other words, any observed differences in disease outcome may reflect differences in the distribution of modifying variables rather than differences in the exposure of interest.

In practice the term "confounder" is often loosely applied to include both "true" confounders and modifying factors.

3.4 Judging The Quality Of Epidemiological Studies

3.4.1 Assessing Individual Studies

In the category of observational studies, cross-sectional and ecological studies are the weakest in design, and are considered to be "hypothesis generating" rather than "hypothesis testing". Due to their relatively low cost and rapid time frame, such studies are generally the first to be done in the investigation of the possible health effects of environmental exposures. Cohort studies and case-control studies are considered to be more informative and to produce evidence of a higher quality as they have the capacity to analyse exposures and

disease at the level of the individual. The randomised controlled trial is the most rigorous design that can be applied to the study of disease in human beings, and provides the highest quality of evidence. However this type of study is generally also the most difficult and expensive to carry out. Application of this design is also restricted by ethical considerations as it is not acceptable to deliberately expose human subjects to increased risks beyond what they may already experience in everyday life.

Within each type of study design, individual studies will vary in quality due to the methods employed, the quality of the data sources, and the degree to which the researchers were able to control for biases, potential confounders and modifying factors which might have influenced the results. Ideally, epidemiological studies should be carried out prospectively, with data collection mechanisms designed specifically for the study and with appropriate quality assurance measures in place. In reality many studies rely on data collected by others, which was intended for a different purpose, and which is sometimes of uncertain quality. The interpretation of the findings from any study must therefore be made with consideration of the inherent strengths and limitations of the study design, together with an assessment of the specific methodology employed in the individual study.

3.4.2 Assessing The Body Of Epidemiological Evidence

To adequately assess the strength of epidemiological evidence relating an exposure to health status it is necessary to carry out a critical review of the evidence. The desirable features of such a review include:

- a clear and concise definition of the question to be examined,
- a defined search strategy to locate relevant publications,
- efforts to locate articles published in lesser known journals, conference proceedings and the 'grey' literature which would not be found through conventional database searches,
- efforts to locate unpublished work through contacts with key researchers in the field (it is well accepted that studies showing no effect of a particular exposure are less likely to be published than those showing statistically significant associations) ,
- well-defined inclusion and exclusion criteria for publications to be included,
- assessment of the methodology of each study against a set of quality criteria, and
- an overall assessment of the strength of the evidence with most weight being placed on the highest quality studies

3.5 Causation

Epidemiological studies can provide evidence of the statistical association between a particular exposure and a disease, but do not prove that the exposure causes the disease. Before deducing that the observed association might represent a causal association, other factors must be examined:

- Consistency of association - if a causal relationship exists, the same association should be found in other independent studies.
- Strength - a strong association provides greater evidence of possible causation than a weak association.

- Specificity - an association that is specific to a single exposure, particularly when there is no association with various other exposures, provides better evidence of causation than a situation where the disease is apparently associated with many factors.
- Appropriate time relationships - some diseases require a period of time to develop and become apparent after exposure to a causative agent. An association is more suggestive if the strongest effect is seen for exposures focused on the appropriate time interval.
- Coherence with other information - a finding from an epidemiological study is more convincing if it is supported by other evidence such as animal or human studies of metabolism, toxicity or carcinogenicity. Such studies should suggest a plausible biological mechanism whereby the exposure exerts its effect.
- Dose response - differing strengths of association may be evident for different durations or intensities of exposure (assuming that studies are large enough to allow this type of statistical analysis). Generally speaking, the risks for a disease should increase as the degree of exposure to a harmful agent increases.

A strong and consistent body of scientific evidence is required to resolve the issue of causation, however there are no specific rules about how much evidence is sufficient to reach a decision on causation.

4 Cardiovascular Diseases

4.1 Classification Of Cardiovascular Diseases

The term "cardiovascular diseases" refers to a diverse group of diseases and conditions involving the heart and blood vessels. In developed countries, the main underlying cause of cardiovascular diseases is atherosclerosis. This is a condition where abnormal build-ups of fat, cholesterol and other substances are found in the inner lining of the arteries. These deposits are called "plaque". Atherosclerosis may cause an interruption or restriction to the flow of blood through the arteries (known as ischaemia), which in turn affects the function of the body tissues supplied by these blood vessels.

Cardiovascular diseases and other diseases are classified under an internationally recognised diagnostic system called the International Classification of Diseases (ICD). This system was initially developed to record causes of death (mortality), but is now also used to classify illness (morbidity). The ICD system undergoes periodic revisions, with each new version being assigned a sequential number. The current revision, ICD-10, officially came into use in WHO member states in 1994: however, there is considerable variation between individual countries as to how rapidly the changeover from the previous edition is being implemented. The published epidemiological studies reviewed in this report have used the ICD-7, ICD-8 or ICD-9 coding systems.

The major forms of cardiovascular disease are:

- **Coronary heart disease** Coronary heart disease (or ischaemic heart disease) is the most common form of heart disease in developed nations. Its major clinical forms are **acute myocardial infarction** (AMI, commonly known as heart attack) and **angina**. Both these forms of disease are caused by the build-up of plaque in the coronary arteries which supply blood to the heart muscle. Heart attacks are mainly caused by the sudden rupture of a plaque in one of the coronary arteries. This causes a blood clot that completely blocks blood flow to the heart muscle downstream of the clot, causing that area of muscle to stop functioning. If the interruption to the blood supply lasts long enough, the area of heart muscle supplied by the artery will die. Heart attacks can also be caused by severe spasms of the coronary arteries resulting in a restriction of blood flow.

  Angina is caused by the build-up of plaque within a coronary artery so that blood flow is permanently restricted. Although the flow of blood may be adequate for normal activities, the artery is unable to cope with the increased demands for blood incurred by physical activity or strong emotion, resulting in temporary chest pain.

  Coronary heart disease is classified under ICD-9 codes 410-414 or ICD-10 codes I20-I25.

  Sudden cardiac death may result from coronary heart disease. This condition is defined as an unexpected death resulting from an abrupt loss of heart function and occurring within a short time of the onset of symptoms. Most sudden cardiac deaths are caused by arrythmias (irregular rapid or chaotic heartbeat) which cause the heart to stop beating (cardiac arrest). Sudden cardiac death is generally defined as occurring within 1 hour of the initial event, although brain death may begin to occur within 4-6 minutes of cardiac

arrest if cardio-pulmonary resuscitation is not performed. Sudden cardiac death may be caused by AMI but may also occur in the absence of AMI.

- **Cerebrovascular disease** This group of conditions is characterised by a disturbance in blood flow to the brain. The most significant clinical form is **stroke**, which occurs when a blood vessel that carries oxygen and nutrients to the brain is either blocked by a clot (ischaemic stroke) or ruptures and bleeds (haemorrhagic stroke), resulting in part of the brain dying from lack of blood flow. This causes loss of function of the affected part of the brain, leading to death or impairment in any or all of a range of physical and mental functions. Cerebrovascular disease is classified under ICD-9 codes 430-438 or ICD-10 codes I60-I64.

- **Heart failure** Heart failure is a condition where the heart is unable to effectively pump blood around the body. Heart failure usually develops gradually over a period of years, however it can also occur suddenly. The causes of heart failure include heart attack, high blood pressure or a damaged heart valve. Heart failure is classified under ICD-9 code 428 or ICD-10 code I50.

- **Peripheral vascular disease** Peripheral vascular disease (also known as peripheral artery disease) refers to diseases of arteries outside the heart and brain. It occurs when fatty deposits build up in the inner walls of these arteries and affect blood circulation, mainly in the arteries leading to the legs and feet. Peripheral vascular disease is classified under ICD-9 codes 441-444 or ICD-10 codes I70-I74.

- **Rheumatic fever and Rheumatic heart disease** Acute rheumatic fever is a delayed complication of an untreated throat infection from Group A *Streptococcus* bacteria and there is some evidence that it may also be caused by streptococcal skin sores. The disease can affect the heart valves, the heart muscle and its lining, the joints and the brain. Its effect on the heart (rheumatic heart disease) is the only permanent manifestation and may be asymptomatic or may result in shortness of breath and chest pain. Chronic rheumatic heart disease is classified under ICD-9 codes 393-398 or ICD-10 codes I05-I10.

- **Congenital heart diseases** Congenital heart diseases refer to disorders of the heart or central (main) blood vessels present at birth. Congenital conditions include abnormalities of the heart or heart valves, defects of vessels such as the aorta and pulmonary artery or combinations of defects. These diseases are classified under ICD-9 codes 754-747 or ICD-10 codes Q20-Q28.

The classes of cardiovascular disease most frequently studied in relation to water quality are Acute Myocardial Infarction or the broader category of Coronary Heart Disease (including AMI and Angina). A few studies have investigated cerebrovascular diseases (specifically Stroke). The categories of rheumatic heart disease and congenital heart disease are considered not relevant to studies of water hardness.

4.2 Factors Affecting Incidence Rates And Mortality From Cardiovascular Diseases

Several factors are known to be important in determining the risk for development of ischaemic cardiovascular diseases. A recent international case-control study has established that nine risk factors account for 90% of the risk of suffering an initial AMI (Yusuf *et al.* 2004). The INTERHEART collaborative study involved over 15,000 cases in 52 countries and showed the same risk factors were important in men and women, and in developed and developing countries.

Factors associated with an increased risk of AMI:

- smoking
- raised ApoB/ApoA1 lipid ratio (a measure of "bad" and "good" cholesterol levels in blood)
- history of hypertension (high blood pressure)
- diabetes
- abdominal obesity
- psychosocial factors (e.g. stress at work or home, depression)

Factors associated with a decreased risk of AMI:

- daily consumption of fruits and vegetables
- regular alcohol consumption
- regular physical activity

The relative importance of each of these risk factors in a population depends on the frequency of the exposure as well as the Odds Ratio associated with each risk. However in all regions of the world, smoking and abnormal lipids were the most significant risk factors in determining risk of AMI. Several of these risk factors are influenced by the behavioural choices of individual people, their socioeconomic status and the nature of the health care system that exists in their society. Socioeconomic status has important impacts on health through multiple pathways including the level of knowledge about healthy and unhealthy behaviours (e.g. diet, exercise, smoking), control of social stress factors, access to diagnostic and preventive health services, and access to treatment. Disparities in socioeconomic status therefore have the potential to impact on virtually any disease outcome being measured in epidemiological studies. Place of residence (i.e. urban versus rural) also has a significant influence, partly due to socioeconomic factors, but also due to the effect of geographic remoteness from major health services.

Mortality from AMI and other cardiovascular events is strongly influenced by how quickly medical assistance can be given to the victim. AMI may result in a range of acute complications including arrhythmias, conduction abnormalities, hemodynamic disturbances (cardiogenic shock, congestive heart failure) and myocardial rupture. Arrhythmias are common in the setting of AMI and range from benign to fatal. Ventricular fibrillation is the most common mechanism of sudden cardiac death in AMI. Chances of survival are significantly increased by the use of an external defibrillator in the case of V-fibrillation or V-

tachycardia. Thrombolytic drugs and angioplasty via catheterisation are the first-line measures to manage the acute phase of the AMI in order to save as much myocardium as possible and restore contractile function of heart chambers.

The delay between the cardiovascular event and the first treatment intervention is influenced by a number of factors. These include the delay between the onset of symptoms and seeking medical help, the location of the individual at the time of the event relative to a major medical facility, the availability and travel time for ambulance services, and local practices regarding pre-hospital treatment by ambulance paramedics and in-hospital treatment. These factors may differ markedly between countries and between different areas within a country, and this may have a significant effect on cardiovascular mortality rates.

4.3 The Proposed Effects Of Calcium And Magnesium

Calcium and magnesium are both essential nutrients for humans. Calcium is the most abundant mineral in the human body, and the majority (99%) of bodily calcium content is located in the bones where it forms the main structural component. About 1% of the calcium in the body is present in extra-cellular and intra-cellular fluids and in cell membranes. Calcium is known to play a role in the regulation of many enzymes and hormonal responses, and is also involved in blood clotting, the transmission of nerve impulses, and the control of muscle contraction including the heart and arterial muscles. Magnesium is also an essential cofactor for many enzymes which are involved in the metabolism of carbohydrate, protein, lipids and nucleic acid. It is also involved in regulation of cell permeability and neuromuscular excitability, and in the development of bone tissue.

The postulated biological mechanisms for a beneficial effect of high magnesium intake on cardiovascular disease incidence or mortality include:

- a reduction of the risk of sudden death from AMI, due to reduction in arrythmias and/or coronary artery vasospasm. Such effects would be expected to relate to magnesium exposure levels in a relatively short time frame before the AMI event, perhaps in the order of only a few months.
- a reduction in systemic hypertension, via effects on vascular smooth muscle. This mechanism would also be expected to operate over a medium to long time frame, perhaps in the order of months to years.
- changes in blood lipid levels. This mechanism would be expected to relate to longer term exposure to magnesium, perhaps in the order of several years or more, since the build-up of atherosclerotic plaque is a gradual process.

Calcium has also been postulated to affect blood lipid levels, but suggestions that it may affect hypertension have not been supported by recent research.

Because of evidence of biological plausibility and promising results from work in animals, there has been considerable interest in the potential of Mg for reducing reperfusion injury and myocardial stunning and for acting as an anti-arrhythmic agent (Antman 1996). However, conflicting results have been reported in clinical trials. A multinational, multicenter trial of intravenous Mg sulphate administration in AMI (MAGIC = Magnesium in Coronaries) was

recently set up to compare short-term mortality in high risk patients with AMI who received either early intravenous Mg sulfate or placebo. Results showed no significant effect on 30-day mortality, and the MAGIC trial investigators concluded that there is no indication for the routine administration of intravenous Mg in patients with AMI in current coronary care practice (Magnesium in Coronaries Trial 2002). Other recent studies' results support this conclusion (Hassan *et al.* 2002).

5 Review Of Epidemiological Studies

This section briefly reviews individual epidemiological studies addressing the issue of water hardness (and/or calcium and magnesium content) and cardiovascular disease. Detailed summaries of each paper and an evaluation of the quality of each are contained in the Appendix.

5.1 Ecological Studies

The WHO review of epidemiological studies published between 1979 and 2004 (Monarca *et al.* 2005) identified 19 ecological (or geographic correlation) studies. These studies are not reviewed here as the strength of evidence they are capable of providing is greatly inferior to analytical epidemiology study designs.

5.2 Ecological Studies Using Small Area Statistics

In recent years the availability of data for small geographic areas and new data linkage and modelling capabilities has allowed ecological studies of a more sophisticated design to be performed. In most of these studies, the geographic areas utilised are the smallest unit used for population census data collection. Such areas generally comprise only a few hundred households. While these studies are still not able to assess exposures and risk factors at the level of the individual person, they can assign values to small population groups and thus provide a higher degree of accuracy than that achieved using the larger scale geographic boundaries employed in traditional ecological studies. Two relevant studies of this nature were identified in this review.

- Maheswaran *et al.* carried out a study examining the relationship between magnesium in drinking water supplies and mortality from AMI in people aged 45 and older in the north west of England (Maheswaran *et al.* 1999). The availability of census data on age, sex and socioeconomic deprivation permitted adjustment for these factors on a small geographic scale. The study found no evidence for a protective effect of magnesium in either sex. There was also no evidence of a protective effect in different socioeconomic classes or in different age groups. When the broader category of mortality from ischaemic heart disease was assessed, both magnesium and calcium appeared to have significant protective effects, but this disappeared after adjustment for geographic gradients in disease rates.

- Another study of this type was carried out in Finland by Kousa *et al.*, this time examining the incidence of AMI in men aged 35-74 years (Kousa *et al.* 2003). The geographic units in this study were arbitrarily defined cells measuring 10 km x 10 km covering the entire country. The study found a statistically significant relationship between water hardness and risk of AMI, with an increase of 1 dºH (German hardness unit) in hardness being associated with a 1% decline in AMI risk. The results for calcium and magnesium were not presented in the paper. Some other water parameters showed non-significant associations with AMI risk; copper was associated with a 4% increase in AMI risk for

each 1µg/L increase in concentration, and iron with a 10% increase in AMI risk for each 1 mg/L increase in concentration. Fluoride was associated with a 3% decrease in AMI risk for each 1 mg/L increase in concentration. This study did not adjust for the influence of individual risk factors on AMI incidence.

These two studies have given conflicting results on the influence of water hardness. The findings of the UK study did not support a protective role for magnesium or calcium, while the Finnish study found a protective effect of hardness but did not separately analyse calcium and magnesium. The UK study included adjustment for socioeconomic status, whereas the Finnish study did not do so.

5.3 Cohort Studies

Two cohort studies were identified in the WHO 1979 to 2004 review, and a further cohort study from the UK has since been published.

- Comstock *et al.* examined the relationship between water hardness and deaths from arteriosclerotic heart disease in a population of about 30,000 men and women in the US followed for 12 years (Comstock *et al.* 1980). Water hardness was classified on an ecological basis into 6 categories rather than by measurement at individual households. However the investigators had information on whether households used municipal supplies or private wells as their drinking water source. The study was able to adjust for socioeconomic factors and smoking at the individual level, and also looked at the effect of gender, long or short term residence (more or less than 7 years), and urban/rural location. Individual water intake was not assessed. No consistent relationship was found between water hardness and risk of death from arteriosclerotic heart disease whether considering all arteriosclerotic deaths or only those occurring within 1 hour of the precipitating event (sudden death). The paper does not provide information on magnesium and calcium levels in water: however, the authors remarked that the two water supplies with greatest variation in magnesium content (small towns and deep wells) showed opposite associations with sudden death and hardness.

- Punsar and Karvonen compared cohorts of men in two rural areas of Finland which were known to have different rates of cardiovascular mortality (Punsar and Karvonen 1979). These areas also differed in a number of water quality parameters. This study specifically evaluated copper, chromium and magnesium. The data set analysed included 366 men from the western region (lower CVD risk) and 427 from the eastern region (higher CVD risk) who had been followed up for 15 years since 1959. Water quality was classified on an ecological basis using median values for subregions. No assessment was made of individual risk factors such as smoking, and water intake was not assessed. There was also no assessment of socioeconomic factors. Median levels of water constituents were compared for men who remained alive in 1974 and those who had died during follow up. In the western region, chromium levels were significantly lower for those who had died from coronary heart disease (CHD) than for those still alive. There were no significant differences for magnesium or copper. In the eastern region, magnesium levels were significantly lower for the categories of death from all causes and death from other

causes but not for deaths from CHD. There were no significant differences for copper or chromium levels. There was also no significant association between water quality parameters and sudden CHD deaths. (Note that this paper was cited in the WHO review (section 3.3) as supporting the protective effect of magnesium, however this observation relates to the crude ecological comparison of the two regions, not to the more detailed cohort analysis).

- Morris *et al.* reported the results of a 15 year follow up of a cohort of 7,735 men from 24 towns in Great Britain chosen to represent a wide range of coronary heart disease mortality rates (Morris *et al.* 2001). This publication focused on the effects of individual risk factors (smoking, physical activity, body mass index etc) on CHD but some observations on water quality were also reported. The analysis showed that up to 75% of the variance in CHD rates between towns was explained by the effects of smoking, physical activity, systolic blood pressure, occupational social class, and height. The effect of water hardness after adjustment for these factors was non-significant (OR 0.96, 95% CI 0.88 – 1.05).

The findings of these three cohort studies do not support a significant protective role for water hardness or magnesium content for CHD mortality.

5.3.1 Case-control Studies

Seven case-control studies have been published on the topic of cardiovascular disease and water hardness between 1979 and 2004. Five of these studies did not attempt to assess individual risk factors for cardiovascular disease (e.g. smoking, diet, exercise), and only two of these five made some adjustment for socioeconomic factors. The five studies assigned exposure to drinking water on the basis of place of residence.

- Luoma *et al.* carried out a study of AMI in men aged 30-64 years in the southeast region of Finland (Luoma *et al.* 1983). AMI cases were compared to two sets of controls; one set selected from hospital patients with other illnesses and the other from the general population. The study found a statistically significant protective effect of magnesium at levels above 1.2 mg/L when comparing cases to population controls: however, it also found a stronger protective effect associated with fluoride at levels above 0.1 mg/L. Calcium did not appear to have a protective effect. The study did not adjust for socioeconomic factors or individual risk factors.

- Yang conducted a study of deaths from cerebrovascular disease in people aged 50-69 years in Taiwan (Yang 1998). Cases were compared to controls who had died of a range of other causes. This study made an adjustment for socioeconomic status on the basis of urbanization, but did not adjust for individual risk factors. The study found a statistically significant protective effect of magnesium on cerebrovascular mortality at levels above 7.3 mg/L, but no significant effect of calcium.

- Yang and Chui carried out a second study in Taiwan examining the relationship between calcium and magnesium in drinking water and deaths from hypertension-related diseases (Yang and Chui 1999). Again, the controls were people who had died from other causes.

This study showed a statistically significant protective effect of magnesium at levels above 3.8 mg/L. In the analysis, adjustment was made for socioeconomic status on the basis of urbanization, but there was no adjustment for individual risk factors.

- Rubenowitz *et al.* examined mortality from AMI among men aged 50-69 years living in southern Sweden (Rubenowitz *et al.* 1996). The control group was men in the same age group who had died of cancer. This study found a significant protective effect of magnesium at levels above 3.6 mg/L, but no effect of calcium. No adjustments were made for individual risk factors or socioeconomic factors.

- Rubenowitz *et al.* carried out a further study of AMI mortality in the same area of Sweden, this time in women aged 50-69 years (Rubenowitz *et al.* 1999). Women of the same age range who had died of cancer were used as the control group. A significant protective effect was seen for magnesium levels above 9.9 mg/L, and for calcium levels above 70 mg/L. The study did not adjust for individual risk factors or socioeconomic factors.

The remaining two case-control studies collected more detailed information from individual cases and controls by questionnaire, thus permitting a more sophisticated level of analysis looking at the effects of individual risk factors.

- Rubenowitz *et al.* performed a study in 18 municipalities in the southern part of Sweden to examine the relationship of calcium and magnesium levels in drinking water and morbidity and mortality from AMI (Rubenowitz *et al.* 2000). Cases were men and women aged 50-74 years who had suffered an AMI during a 21 month period in 1994-1996. Risk factors evaluated included smoking, levels of stress, body mass index, diabetes, hypertension, and physical inactivity: however, information on these factors was recorded only for surviving cases and their controls. Water intake was also assessed, but again only for surviving cases and their controls.

  In the analysis considering only age, calcium and magnesium levels in water, there were no significant differences in the incidence of AMI in relation to levels of calcium or magnesium. However the risk of suffering and dying of an AMI was significantly reduced among people in the highest quartile of magnesium concentration (8.3 mg/L or higher) relative to the other three quartiles combined. No significant associations were seen for calcium. When the analysis was repeated using estimated magnesium intake from water (concentration x volume consumed), the odds of suffering an AMI and surviving increased with higher magnesium intake. This analysis was restricted to surviving cases and their controls, as water consumption data were not collected for fatal cases and their controls. These results might suggest a protective effect of magnesium in drinking water against fatal AMI, although the overall risk of suffering an AMI was not reduced. However, this analysis did not take into account any differences that may have existed between high and low magnesium exposure groups in terms of smoking rates, socioeconomic status etc. The authors comment that the reduction in fatality rate was mostly for deaths occurring outside hospital, and this supports the hypothesis that magnesium may protect against sudden death from AMI, rather than deaths from all classes of ischaemic heart disease.

The failure to collect information on individual risk factors for deceased cases and their controls was a serious flaw in the design of this study. Lack of information on these groups means that the effect of these factors on the risk of suffering an AMI or dying from AMI cannot properly be assessed in the statistical analysis. Thus it is not possible to say whether differences in the prevalence of individual risk factors such as smoking or socioeconomic status might have been responsible for the observed differences in AMI mortality, rather than the differences in water composition. No information was collected on water consumption for deceased cases and their controls: therefore, it is not even certain whether they drank tap water. Furthermore, it appears that the deaths recorded in the study may have occurred up to 22 months after the AMI event, and there was no analysis for the specific subgroup corresponding to the sudden death category which is postulated by the authors to be the most relevant health outcome (see Appendix for further discussion).

- Rosenlund *et al.* carried out a case-control study in a different region of Sweden, which examined first AMI events in relation to calcium, magnesium and hardness levels in drinking water (Rosenlund *et al.* 2005). This was a sub-study of a large cardiovascular study, the Stockholm Heart Epidemiology Study, which had high quality methodology. The study included assessment of smoking, job strain, BMI, diabetes, hypertension, and physical inactivity. Cases and controls were also questioned about water consumption and diet so that intake of calcium and magnesium from various sources could be estimated. Information for deceased cases was collected from close relatives.

  The study found no significant relationship for AMI risk with Mg, Ca or hardness levels in water regardless of how exposure levels were characterised. The risk of AMI tended to increase slightly as Mg levels increased (suggestive of a harmful rather than protective effect), but there was no consistent trend for other parameters. Analysis in terms of estimated Mg or Ca intake from water (calculated from water consumption information) also showed no significant association with AMI risk.

  A subgroup analysis was carried out for cases where death occurred within 28 days of AMI diagnosis. Exposure was defined in terms of daily magnesium and calcium intake from tap water for the 2 years prior to diagnosis. Again, no significant relationship with AMI risk was observed.

Overall, the case-control studies have yielded mixed outcomes. Those which have not controlled for individual risk factors have tended to find a protective association between higher magnesium levels in drinking water and cardiovascular disease, although one of these reported an even stronger protective association with fluoride levels (Luoma *et al.* 1983). A design flaw in one of the two most recent studies (failure to collect risk factor information for deceased cases and their controls) meant that it was unable to assess the influence of individual risk factors on the apparent relationship between magnesium and AMI mortality (Rubenowitz *et al.* 2000).

It should also be noted that a number of these studies have been conducted in the same geographic area as each other, and thus they do not represent independent settings. For example, the three Rubenowitz *et al.* studies were performed in the same geographic area of

Sweden while the studies by Yang and Yang & Chui were carried out in the same geographic area of Taiwan. Therefore these five studies represent only two independent settings. If there was an uneven geographic distribution of individual risk factors in a given study area (say if there happened to be more smokers living in soft water regions of Sweden or Taiwan), this would create an underlying bias in cardiovascular disease risk that would affect all studies conducted in that area. None of these studies assessed smoking or other individual risk factors and therefore they would have been unable to detect and adjust for such sources of bias in their statistical analyses.

To date only the Rosenlund *et al.* case-control study can be regarded as having made an adequate assessment of individual risk factors, and no protective effect of magnesium or calcium was found in this instance (Rosenlund *et al.,* 2005). The range of exposures examined in this study was relatively restricted, with only 7% of people exposed to Mg levels above 8.0 mg/L in drinking water, and 16% exposed to Ca levels of 28.5 mg/L or above. The authors have suggested that the failure to find an effect may have been due to an insufficient degree of contrast in exposures. However, some of the other case-control studies outlined above have found statistically significant differences within a low range of magnesium exposures. For example, the Finnish study found a significant effect on AMI risk using a cutoff value of only 1.2 mg/L for magnesium to define low and high exposure categories (Luoma *et al.* 1983), while a study in Sweden found a significantly decreased risk of death from AMI among men exposed to magnesium at 6.9 mg/L or above compared to those exposed to 3.5 mg/L or less (Rubenowitz *et al.* 1996 ,Rubenowitz *et al.* 1999). It would appear therefore that the failure of the Rosenlund study to find an effect of magnesium may not be simply dismissed on the grounds that it had an inadequate range of exposures.

None of the studies performed to date have attempted to assess the possible effects of availability of appropriate medical care, or pre-hospital / in-hospital treatment practices on mortality from cardiovascular diseases.

5.4 Other water quality parameters

Some of the epidemiological studies reviewed here have raised the possibility of alternative interpretations of the epidemiological data relating either to protective or adverse effects of other substances in water. These suggestions include a protective effect of fluoride, or an adverse effect of some metals.

5.4.1 Fluoride

A case-control study of AMI in the southeast region of Finland observed a stronger protective relationship for fluoride than for magnesium (Luoma *et al.* 1983). A statistically significant protective effect was evident using cutoff levels in the range of 0.1 to 0.5 mg/L to define high and low fluoride exposure. More recently a small scale ecological study covering the entire country of Finland also noted a protective effect of fluoride although this was not statistically significant (Kousa *et al.* 2003). An ecological study specifically examining fluoride in drinking water and coronary heart disease in rural areas of Finland found a significant protective effect (Kaipio *et al.* 2004). In this study a J-shaped risk curve was observed with an optimum protective effect in the range of 0.15 to 0.30 mg/L fluoride and a stronger effect in younger

age groups (35-64 years) than in older people (65 years and older). The only other study reviewed here which considered fluoride was the small area ecological study carried out in the northwest of England (Maheswaran *et al.* 1999). This study did not observe any effect of fluoride on AMI risk.

A possible mechanism by which fluoride might influence the incidence of cardiovascular disease is through the prevention of dental caries and periodontal disease. Recent research has shown an association between dental infections and increased risks of cardiovascular disease and stroke, possibly mediated by inflammatory effects (Pihlstrom *et al.* 2005).

5.4.2 Metals

Calcium and magnesium in drinking water are largely responsible for water hardness, and hardness is a major factor in determining the corrosivity of water towards metal components of water distribution systems and household plumbing and fixtures. A number of these metals have the potential to play a role in cardiovascular disease.

A few of the studies reviewed here included metals in their data analysis. A small area ecological study in Finland noted a non-significant increase in the risk of AMI associated with increasing levels of copper and iron in drinking water (Kousa *et al.* 2003). The small scale ecological study conducted in northwest England found a non-significant protective effect of lead (Maheswaran *et al.* 1999). A cohort study in rural areas of Finland observed inconsistent patterns of risk in relation to chromium levels and CHD and no significant association for copper (Punsar and Karvonen 1979).

Because naturally soft waters can be very corrosive, metals in drinking water have the potential to be true confounders in studies of the health effects of hardness. The impact of specific metals is likely to vary between water supplies depending on variations in other water quality parameters which contribute to corrosivity, the degree to which metal components are used in the distribution system and the quality of materials in household plumbing. Artificially softened waters usually are not corrosive and have sufficient buffering capacity to prevent excessive leaching of metals.

5.5 Conclusions

This review has shown that the analytical epidemiological studies undertaken on the issue of water hardness and cardiovascular disease do not provide consistent or strong support for the protective effect of magnesium or water hardness. Most of the studies on this topic must be considered as being of low quality as they have not assessed the impact of individual risk factors or socioeconomic factors on the apparent relationship between water quality parameters and cardiovascular disease incidence or mortality rates.

Those studies which have included adjustment for socioeconomic factors and/or individual atherosclerosis risk factors in their statistical analysis have generally not observed a significant effect of water quality. This suggests that the apparent protective effect of water hardness commonly seen in ecological studies may be mostly or entirely attributable to

differences in the distribution of socioeconomic factors and/or individual risk factors in the populations being compared, rather than any effect of water quality. In addition, some epidemiological studies have raised the possibility of alternative explanations relating to either protective or adverse effects of other substances in water.

Among the studies examining mortality, none have attempted to account for the effects of medical intervention on death rates. Only two of the studies assessed sudden cardiac death, which is frequently cited as the outcome of most relevance for the proposed protective effect of magnesium. Both of these studies found no effect of magnesium or hardness levels on sudden cardiac death (Comstock *et al.* 1980, Punsar and Karvonen 1979).

In conclusion, the current body of epidemiological evidence contains a relatively small number of analytical studies: the quality of most of these studies is poor: and the findings are inconsistent. The available evidence is not sufficiently strong to infer that low levels of calcium and/or magnesium in drinking water are significant factors in the causation of cardiovascular disease.

6 Design Needs for Future Epidemiological Studies

This review has highlighted a number of important omissions and design flaws in past epidemiological studies which need to be addressed if future studies are to make a worthwhile contribution to the knowledge base on this issue.

Definition of the research question
Two different questions need to be addressed, and the choice of question will affect the study design that is required:

- Do high levels of magnesium in drinking water have a protective effect against the development of ischaemic coronary disease (or other cardiovascular diseases)? (Effect on incidence)
- Do high levels of magnesium in drinking water have a protective effect against sudden cardiac death among people who suffer an AMI? (Effect on mortality)

Definition and validation of health outcomes
The ICD coding system is an accepted means of recording cause of death, however some categories of cardiovascular disease are not relevant to the magnesium hypothesis and should be excluded from future studies.

A number of studies have shown that coding of cause of death on death certificates has a significant rate of error. To ensure a high degree of accuracy, consideration should be given to setting more stringent criteria to verify the cardiovascular outcomes.

The time frame between the cardiovascular event and the health outcome needs to be clearly defined. For mortality studies it would be preferable to limit the time frame so that death could be reasonably attributed to the event rather than other subsequent causes or complicating conditions.

Assessment of relevant cardiovascular risk factors
For incidence studies, the potential confounding and modifying factors that need to be addressed include the known cardiovascular risk factors such as socioeconomic status, smoking, hypertension, lipid levels etc. Given that magnesium may affect both lipid metabolism and hypertension, the statistical analysis should be performed with and without adjustment these factors to determine how strongly they may influence the result.

For mortality studies, the relevant confounding and modifying factors include variables associated with the lag time between the outset of symptoms and medical intervention, and the efficiency of the emergency care system (including the nature of the treatment). The cardiovascular risk factors listed above may also be relevant to outcomes as well as incidence.

Assessment of magnesium exposure
Given the increasing consumption of bottled water in many countries, future studies should determine that study subjects actually drink tap water, and what volume they consume. The effect of home water treatment devices should also be assessed, together with the dietary

intake of minerals. An exposure history covering perhaps 20 years or more should be recorded for incidence studies, while for mortality studies a time period of 1 year would probably be adequate. Assessment should be done at the individual level although water sampling may not be required at individual households, depending on the nature of the drinking water supply system.

Assessment of other water quality parameters

The potential influence of other substances in water should be assessed, particularly fluoride and metals as these have the potential to be true confounders. Accurate exposure assessment for metals in water may be difficult as samples need to be taken inside dwellings rather than from the distribution system, and individual behaviour patterns also need to be assessed (i.e. whether a person habitually drinks "first flush" water or not).

Selection of study design

Ecological studies where exposure is classified on a large scale cannot provide meaningful information on this issue and further studies of this nature will not be helpful in resolving the question. Small scale geographic studies may provide some useful information if they are able to adjust for some of the major cardiovascular risk factors.

Cohort or case-control studies are the most suitable designs to examine this issue:

A cohort study examining the incidence of cardiovascular diseases in a population of healthy people (i.e. not already having hypertension, high cholesterol etc) may require a very large number of subjects to be followed over time. However a smaller number would be adequate if subjects with a high risk of AMI were used as the population study base and sudden death incidence rates were compared according to the Mg level/intake from drinking water.

The case-control study design could be used to examine both fatal and non-fatal events and assess the effects of magnesium and other relevant exposures. In such a design it is important that the same information is collected for both deceased and living cases and their controls.

Intervention studies to address these research questions are not likely to be feasible due to the large number of people that would be required to provide the required statistical power. It maybe possible to address the sudden death issue in a randomised trial of a high risk group such as those who have already suffered an AMI, however the findings in this group may not be applicable to the broader population at risk of first AMI. Intervention studies on smaller numbers of subjects could be used to explore the biological effects of calcium or magnesium on risk factors such as hypertension or cholesterol levels.

Synergism with other studies

In an ideal world we might wish to design and conduct a 'perfect' study with the sole purpose of addressing this question, however large epidemiological studies are complex and costly to undertake, and before embarking on this course we need to consider how the same ends could be accomplished more rapidly and at a lower cost through other approaches. Cardiovascular disease is a leading cause of morbidity and mortality in middle-aged and older adults in the developed world, and its impact in developing nations is rapidly increasing. As a result, many clinical and epidemiological studies are currently being undertaken to assess the effect of lifestyle factors, preventive programs and treatment interventions on

disease incidence and outcomes. Some of these projects are multi-centre studies across large geographic areas in a single country while others involve international collaborations. These studies offer opportunities to address many of the questions regarding the effects of water quality on cardiovascular disease incidence and outcomes, and investigate the underlying biochemical mechanisms. To date these opportunities appear not to have been exploited by water quality researchers, with only one recently published case-control study of heart attack being conducted as a sub-study of a larger study with a primary focus on coronary heart disease. Through the analysis of existing datasets and partnership with ongoing and new epidemiological studies on cardiovascular disease, the potential exists to overcome the limitations of past research and rapidly advance our knowledge and understanding of this important public health issue.

7 Appendix - Summaries of Epidemiological Studies

Comstock GW, Cauthen GM and Helsing K J (1980).
Water hardness at home and deaths from arteriosclerotic heart disease in Washington County, Maryland. *American Journal of Epidemiology* **112**(2): 209-216.

Aim: To conduct a prospective epidemiological study of the relationship between water hardness and death from arteriosclerotic heart disease.

Design: Cohort study.

Location and population: Washington County, Maryland, USA. A resident population of an estimated 30,942 people aged 25 years or older were included in the study. The area has a number of different water supplies including two rivers with soft and moderately hard water, and both soft and very hard groundwater.

Identification of cohort: Residents aged 25 years or older in July 1963 were identified in a private census. Only those who reported drinking from municipal supplies or deep wells (over 50 feet /15 metres deep) were included. Other exclusions comprised residents of the town of Sharpsburg which had recently had a major change in its drinking water supply, and non-white people who had different rates of cardiovascular disease from other residents and lived mainly in one town. Population size was estimated at the midpoint of the study period (1971) from a resample of 5% of the original census group.

Time period: 16 July 1963 to 15 July 1975 (12 year follow up).

Drinking water parameters:
Hardness as calcium carbonate, range 0-389 ppm

Source of water quality data: Results from 1,569 water samples collected from private residences during the 1963 census were used to assign a category of water hardness to each residence based on its location and census information on well water or municipal water use. Six water hardness categories were used.

Risk factor information: Information on smoking history was available from the census. Other characteristics known at the individual level were age, sex, marital status, years of schooling, duration of residence, and frequency of church attendance (stated by the authors to have an inverse relationship with chronic illness).

Risk factor information source: Census questionnaire.

Health outcomes: Deaths occurring during the study period and coded as ICD-7 codes 420-422. Information was also available on the duration of the immediate and underlying cause of death, and this allowed the identification of the sub-category of sudden death.

Health outcome information source: Death certificates.

Results:
A total of 6,126 deaths occurred in the study population, of which 2,406 were coded as due to arteriosclerotic heart disease. These deaths were further categorised as being “instant” (n=503), not instant but having an underlying cause with duration of less than 1 hour (473), or having an underlying cause with duration of greater than 1 hour (1430).

Statistical analysis apparently included adjustment for smoking, age, marital status, years of schooling, and frequency of church attendance (this is not described very clearly in the paper). For the

analysis, the population was divided into long term residents (those who had been in residence for 7 years or longer at the 1963 census) and short term (those who had been in residence less than 7 years at the 1963 census). Females and males were analysed separately. Urban-rural differences were controlled for by separately analysing residents of the main township (Hagerston), smaller towns and rural residents served by deep wells.

There was no consistent relationship between average annual death rates and water hardness. Longer term residents had higher death rates but this was explained by their greater age. Relative risks were calculated by dividing the death rates at 0 ppm hardness with those at 200 ppm hardness. RRs for women were mostly greater than 1 (suggesting an adverse effect of soft water) while those for men were less than 1 (suggesting a protective effect of soft water).

The analysis was repeated looking only at instant deaths or deaths with less than 1 hour duration of underlying cause, but again no consistent relationship with water hardness was seen. Calculated RRs were also variable and inconsistent.

For comparison, RRs for non-cardiovascular deaths were calculated in relation to water hardness, and an inconsistent pattern was also seen with a similar range of RR values above and below 1.0.

Authors conclusions:
The study does not support the hypothesis that hard water reduces rates of cardiovascular death.
The paper does not present numerical information on specific minerals, however the authors comment that the two supplies with greatest variation in magnesium content (small town and deep wells) showed opposite associations with sudden death and hardness and hence with magnesium content. It has been postulated that magnesium protects the heart against abnormal electrical impulses (arrhythmia), therefore a protective effect against sudden death might have been expected.
The observation that non-cardiovascular deaths were as likely to be associated with soft water as were deaths from arteriosclerotic disease also argues against any specific effect of water hardness.

Strengths and Limitations

Definition of cohort

- Census records of individuals.

Water exposure

- Household water hardness was not individually measured but classified by area.
- Information was available on type of water source (private wells or municipal supplies) for individual households.
- Assessed different exposure durations (greater or less than 7 years).

Risk factors

- Adjustment was made for smoking and socioeconomic status at the individual level.
- No measurement of other risk factors.

Validation of health outcomes

- Death certificates only. Matched against individual census records.

Effects of medical treatment

- Not assessed.

Evaluation
Moderate quality.

Kaipio J, Nayha S and Valtonen V. (2004)
Fluoride in the drinking water and the geographical variation of coronary heart disease in Finland *European Journal of Cardiovascular Prevention and Rehabilitation* **11:** 56-62.

Aim: To investigate the relationship between levels of fluoride in drinking water and mortality from coronary heart disease.

Design: Ecological study of small geographical areas.

Location and population: Finland. The study included 365 rural communes and the target population was men and women over 35 years of age. The size of the total population in this age range is not stated. Only areas still classified as rural in 1976 were included in order to maximise the probability that most people in the study population used well water as their drinking water source. The average population size per commune was 5,800 in 1961 and 5,400 in 1995, however there was a large variation between individual commune sizes (120 - 24,500 people).

Time period: Deaths occurring from 1961 to 1995.

Drinking water parameters:
fluoride, range 0.00 – 2.15 mg/L
magnesium, 0.21-72.70 mg/L
calcium, 5.68 – 165.43 mg/L

Source of water quality data: Water quality data came from a national survey conducted in 1958. One primary school per 1000 inhabitants was randomly selected and one student was asked to submit a sample of well water from their home supply. A total of 2,764 samples were collected. After exclusion of communes which had been reclassified as urban by 1976 (n=110) and those with missing water quality data (n=6), there were 2,131 samples from the 365 communes in the study (1-17 samples per commune). The mean and median values of water parameters for each commune were assumed to represent the exposure level for the commune.

Risk factor information: The average of the mean monthly income by commune for the years 1970 and 1993 was used as an indicator of socioeconomic factors.

Risk factor information source: Statistics Finland.

Health outcomes: Deaths from CHD for people aged 35 years or more. ICD codes for the relevant period were 1961-1968 (ICD-7 code 420), 1969-1986 (ICD-8 code 410-414), and 1987-1995 (ICD-9 codes 410-414).

Health outcome information source: Death certificates recorded by the Statistical Office of Finland.

Results:
Negative binomial regression was used to examine the effect of sex, age, time period, fluoride, magnesium and calcium on mortality.

In the crude analysis, risk ratios for fluoride showed an inverse J-shaped association with CHD, with the lowest mortality in the second highest quintile (0.1501-0.3000 mg/L). The reduction in mortality was statistically significant for all levels of fluoride above the reference category (0.0000 – 0.0640 mg/L). Risk ratios declined as magnesium levels increased, and the decline was significant in the top two quintiles (above 7.35 mg/L). Risk ratios tended to rise slightly as calcium levels increased. Risk ratios declined significantly as income levels increased.

In the adjusted analysis, the relationships remained very similar with only minor changes to risk ratios. Fluoride remained significantly protective for all exposure categories above the reference category, with the lowest risk ratio (RR=0.85, 95%CI 0.83-0.87) seen in the fourth quintile (0.1501-0.3000 mg/L). Magnesium also was significantly protective for all exposure categories above the reference category, although risk ratios were not as low as those for fluoride (RR=0.92, 95%CI 0.90-0.95 in highest exposure quintile, 10.08-72.70 mg/L). Socioeconomic factors had a stronger influence than either fluoride or magnesium, with the lowest RR in the highest income category (RR=0.80, 95%CI 0.78-0.82).

The adjusted analysis for fluoride was then repeated after dividing the subjects into two age categories. This showed a much stronger effect in the younger age category (35-64 years) than the older category (65 years and above). For the younger age group, the risk ratio fell to 0.76 (95%CI 0.74-0.79) in the second highest fluoride quintile (0.1501-0.3000 mg/L). In the older group, the corresponding risk ratio was 0.90 (95%CI 0.88-0.93).

Overall CHD mortality in Finland has declined over the study period, however a strong geographic variation in rates still exists. The effect of fluoride during each of the 4 decades of the study period was examined, and it was found that the inverse J-shaped relationship with CHD mortality was present in all time periods, although the strength of the protective effect decreased over time. In 1961-1970 the adjusted CHD mortality for the fourth quintile of fluoride concentration was 22% lower than the first quintile, but for 1991-1995 the difference was only 13%.

Authors conclusions:
The study has demonstrated an ecological association between fluoride levels in drinking water and mortality from CHD, although causality cannot be asserted due to the study design. A possible mechanism may be via the prevention of dental caries and periodontal disease. Recent research has shown an association between dental infections and increased risks of CHD. The protective effect observed here was evident at fluoride levels well below the concentration normally recommended for water fluoridation to prevent dental caries (1 mg/L). The declining effect of fluoride in water over time may be due to increasing exposure to fluoride from other sources (eg packaged food and drinks made from fluoridated water, fluoridated toothpaste).

Strengths and Limitations
Definition of populations
- Not well defined. Populations were variable in size and the study did not achieve a fine level of scale.

Water exposure
- Assumed from place of residence.
- Household water hardness was not individually measured but classified by area.

Risk factors
- Adjusted for socioeconomic factors at level of commune.
- No adjustment for other risk factors.

Validation of health outcomes
- Death certificates only.

Effects of medical treatment
- Not assessed.

Evaluation
Low quality.

Kousa A, Moltchanova E, Viik-Kajander M*, et al.* (2003)
Geochemistry of ground water and the incidence of acute myocardial infarction in Finland. *Journal of Epidemiology and Community Health* **58:** 136-139.

Aim: To examine the association of spatial variation in AMI incidence and its putative environmental determinants in groundwater.

Design: Small area geographical study using 10 km x 10 km cells.

Location and population: Finland. The target population were men aged 35 - 74 years, the total number in this age range appears to be 3,526,852 (from summing figures in Table 2). Data on the number of people at risk in each 10 km x 10km cell were provided by Statistics Finland but the number range is not stated in the paper.

Time period: 1983, 1988 and 1993 (individual years)

Drinking water parameters:

parameter	unit	median	75th percentile
total hardness	(German degrees, ºdH)	2.8	4.9
calcium	(mg/L)	14.4	24.4
magnesium	(mg/L)	3.3	6.5
zinc	(µg/L)	11.4	36.8
aluminium	(µg/L)	10.4	61.4
copper	(µg/L)	2.4	9.8
fluoride	(mg/L)	0.1	0.3
iron	(mg/L)	0.0	0.1
nitrate	(mg/L)	1.0	5.9

Source of water quality data:
Hydrogeographic database for the Geological Survey of Finland. For most parameters the dataset comprised 3,621 measurements of groundwater. The number of samples was larger for fluoride (12,407) and nitrate (4,039). The time period of data collection was not stated.

Risk factor information: None.

Risk factor information source: N/A

Health outcomes:
First AMI (fatal or non-fatal) in men aged 35-75 during the years 1983, 1988 and 1993. Data for the three years were pooled. The text states that ICD-8 and ICD-9 codes 410-414 were included, however only code 410 specifically relates to AMI while codes 411-414 relate to other forms of ischaemic heart disease.

Health outcome information source:
National death register and hospital discharge register.

Results:
A Bayesian spatial conditional autoregressive model was used for analysis. Age group (5 year increments), total water hardness and other water parameters were included in the analysis as covariates.

When water hardness was classified into 3 categories (<1.7 ºdH, 1.71 to <5.2 ºdH, ≥5.2 ºdH), a decrease in the age standardised rate of AMI was seen as hardness increased. One unit (ºdH)

increase in hardness was associated with a 1% decrease in AMI risk and this effect was statistically significant (Bayesian 95% High Density Region -2.14% to -0.03%, approximately analogous to frequentist 95% confidence interval).

Results for calcium and magnesium were not presented.

Some other water parameters showed non-significant associations with AMI risk; copper was associated with a 4% increase in AMI risk for each 1µg/L increase in concentration, and iron with a 10% increase in AMI risk for each 1 mg/L increase in concentration. Fluoride was associated with a 3% decrease in AMI risk for each 1 mg/L increase in concentration.

Authors conclusions:
The results support earlier observations of an inverse association between water hardness and AMI incidence in Finland. Although the results for fluoride, copper and iron were not statistically significant, this may have been due to the smoothing techniques used for data manipulation. These elements may also be important for the risk of AMI.

Strengths and Limitations
Definition of populations
- Populations in small areas (10 x 10 km). Number range in areas not stated.

Water exposure
- Assumed from place of residence.

Risk factors
- Not assessed.

Validation of health outcomes
- Death records and hospital discharge records.

Effects of medical treatment
- Not assessed.

Evaluation
Low quality.

Luoma H, Aromaa A, et al. (1983).
Risk of myocardial infarction in Finnish men in relation to fluoride, magnesium and calcium concentration in drinking water. *Acta Medica Scandinavica* **213**(3): 171-6, 1983.

Aim: To examine the possible association between acute myocardial infarction (AMI) and some water constituents, primarily fluoride and magnesium.

Design: Case-control study of AMI cases using hospital controls and population controls.

Location and population: Finland, southeastern region with variability in levels of natural fluoride and magnesium in water, and variability of magnesium levels in soil. Study area comprised one central hospital district, 6 towns and 8 rural communes. Total population about 180,000.

Identification of cases: Cases were defined as male patients who were discharged (alive or dead) from Kotka Central Hospital after their first AMI. 69 cases were initially identified, 3 were then excluded due to prior history of urolithiasis (kidney stones) on hospital records, leaving 66 cases for data collection. A further 8 cases were later excluded due to prior medical history of AMI, urolithiasis or gastrectomy revealed by responses to a questionnaire, or due to missing water quality data. Information from the remaining 58 cases was used for the analysis.

Identification of hospital controls (HCs): Male hospital patients discharged from same hospital with a surgical diagnosis during the same year as cases. Two tentative controls initially selected per case, matched by age (+/- 2 years) and type of community (urban/rural). Hospital records were then examined and controls with prior AMI (number not stated but can be calculated as 3), urolithiasis (1), or gastrectomy (2) were excluded. 3 second controls could not be matched to cases. Of the remaining 129 controls, 16 were later excluded due to prior medical history of AMI, urolithiasis or gastrectomy revealed by responses to a questionnaire, or due to missing water quality data. Information from the remaining 113 HCs was used for the analysis.

Identification of population controls (PCs): 3 tentative population controls selected for each case from population register, matched by age (+/- 1 year) and municipality. After exclusion of people with missing questionnaires or water quality data, 127 PCs remained for analysis.

Time period: Cases discharged from hospital between 1 January 1974 and 30 June 1975.

Drinking water parameters:
Calcium, range 5.0 - 75 ppm
Magnesium, range 0.75 - 57.5 ppm
Fluoride, range 0.05 - 7.4 ppm

Source of water quality data: Sample bottle mailed to cases, HCs and PCs with request to send back 250ml of domestic drinking water. Levels of fluoride, calcium and magnesium were tested.
Questionnaire collected data on water source (well or pipe), and duration of use for this water source.

Risk factor information: Information on hypertension and diabetes collected from hospital records and questionnaires but apparently not used in analysis.

Health outcomes: First AMI.

Health outcome information source: Combination of hospital records and self-reported medical history for cases and HCs, self-reported medical history for PCs.

Statistical Analysis:
Response rates were 92% for cases, 87% for HCs and 70% for PCs.

Two case-control data sets were constructed for analysis:
- Series 1 – the first PC and the first HC were selected as controls (58 case-HC and 58 case-PC pairs).
- Series 2 – HCs with fractures were replaced by HCs without a discharge diagnosis or prior history of fracture. This was done due to the association seen between low fluoride levels and higher risk of fracture. This limitation resulted in only 50 case-HC and case-PC pairs being available for analysis.

Results:
Series 1 comparisons
- PCs had significantly higher mean fluoride levels in their drinking water than cases.
- There were no significant differences between the groups for mean age, mean Ca or mean Mg levels in drinking water, nor in duration of use of the water source, or percentage of well water users.
- cases were more likely than HCs or PCs to have experienced other cardiovascular diseases in addition to MI (eg angina pectoris, congestive heart failure, intermittent claudication, cerebrovascular accident) (raw numbers presented, statistical comparison not done).
- cases were also more likely to have had hypertension or diabetes but none reported fractures.
- among the 58 hospital controls there was a high rate of fractures reported in hospital discharge records (27 people) or self reported (2 additional people). Only 1 of 58 population controls reported a prior history of fracture.

Several different cut-off levels were used to define low or high exposures to elements in water, and the number of discordant case-control pairs was used to calculate a point estimate of risk and 95% confidence intervals

For Series 1 comparisons between cases and population controls, a statistically significant excess risk of MI was associated with very low fluoride (cutoff 0.1 ppm) RR = 4.17 (95%CI 1.66 – 12.51), and with low magnesium (cutoff 1.2 ppm) RR = 3.75 (95%CI 1.20-15.67). No statistically significant results were seen when cases and hospital controls were compared.

Series 2 comparisons
Fluoride Comparison of cases and hospital controls showed a statistically significant excess risk of MI was associated with very low fluoride levels in water (cutoff 0.1 ppm) RR = 3.00 (95%CI 1.12 – 9.13).
A higher excess risk was seen for low fluoride levels (cutoff 0.1 ppm) when cases were compared to population controls RR = 4.40 (95%CI 1.62-14.87). Statistically significant excess risks were also seen when cutoff levels of 0.3 and 0.5 ppm fluoride were used to define exposure levels, but not when a cutoff level of 1.0 ppm fluoride was used.

Magnesium Comparison of cases and hospital controls did not show any statistically significant associations between low magnesium levels and MI risk for cutoffs of 1.2, 1.5 or 3.0 ppm.
When cases were compared to population controls, there was a statistically significant excess risk when an exposure cutoff of 1.2 ppm was used, RR = 4.67 (95%CI 1.3 – 25.32).

Calcium
No statistically significant associations were seen between MI risk and calcium levels for cutoff levels of 16, 18, and 20 ppm, for either case/hospital control or case/population control comparisons.

Other observations
The authors note that average dietary intake of magnesium in Finland has been estimated at 440 mg/day, therefore intake of 1-2 litres of water containing 3ppm (3-6 mg) seems unlikely to play a

decisive role in prevention of MI. They note that some cases had water with high magnesium levels (up to 57.5 ppm) which elevated the mean concentration in the case group above those for both sets of controls. Average daily intake of calcium in Finland is 1.5 g/day, therefore the contribution from water is negligible.
A previous study of dietary fluoride intake in the study area estimated a total dietary intake of 1.09 mg/day for men, therefore intake from water with a fluoride level of 1ppm would be an important amount.

Authors conclusions:
The results are consistent with the hypothesis that very low fluoride - and perhaps low magnesium - intakes are associated with increased risk of atherosclerosis leading to AMI.

Strengths and Limitations
Definition of cases and controls

- Deaths which occurred before reaching hospital were not included as cases. This may be a source of bias.
- The high rate of fractures in hospital controls (27/58 people or 46.6%), suggests some kind of bias in the selection of this group.

Water exposure

- Water composition was measured at individual households.

Risk factors

- Not assessed.

Validation of health outcomes

- Hospital records and questionnaires.

Effects of medical treatment

- Not assessed.

Evaluation
Low quality.

Maheswaran R, Morris S, Falconer S*, et al.* (1999)
Magnesium in drinking water supplies and mortality from acute myocardial infarction in north west England. *Heart* **82:** 455-460.

Aim: To examine whether higher concentrations of magnesium in drinking water supplies are associated with lower mortality from AMI at a small area geographical level, and to examine if this association is modified by age, sex and socioeconomic deprivation.

Design: Small area geographical study using 13,794 census enumeration districts.

Location and population: 305 water supply zones in north west England. The resident population comprised 1,124,623 men and 1,372,036 women over 45 years of age at the 1991 census. The total population (all ages) in each water supply zone was less than 50,000.

Time period: 1990-1992.

Drinking water parameters:
Magnesium, range 2-111 mg/L, median 12 mg/L
Calcium, range 5-215 mg/L, median 36 mg/L
Fluoride, range 0.02 - 0.8 mg/L, median 0.05 mg/L
Lead, range 1-190 µg/L, median 10 µg/L

Source of water quality data:
Routine water quality monitoring by water authorities. Log transformation was used to reduce the effect of extreme values in water quality parameters. A mean score for each zone was derived by averaging within zone log values for 1990-1992. Exposure to water quality parameters was assigned by overlaying water zone boundaries with census enumeration district boundaries. Where the enumeration district crossed water zone boundaries, the value assigned was for the water quality zone containing the population centroid for the enumeration district.

Risk factor information: No information at the individual level. Data on age, sex and socioeconomic deprivation (as measured by the Carstairs index which has been shown to predict mortality at the small area level) was available at enumeration district level.

Risk factor information source: 1991 population census.

Health outcomes: Deaths from AMI (ICD-9 code 410) in people aged 45 years or older. In secondary analysis the broader category of deaths from ischaemic heart disease including AMI was also assessed (ICD-9 codes 410-414).

Health outcome information source:
Office for National Statistics.

Results:
A total of 21,339 men and 17,883 women died of AMI during the study period. Poisson regression analysis was used to assess the influence of water quality parameters. Levels of magnesium in drinking water had no effect on AMI mortality. The relative risk of mortality from AMI (standardised for age, sex and Carstairs deprivation quintile) for a quadrupling of magnesium concentration in drinking water was 1.01 (95% CI 0.99-1.03). The RR remained non-significant after adjustment for calcium, fluoride and lead concentrations and for geographic gradients in mortality.

When the other parameters were analysed separately, calcium and fluoride appeared to have no significant association with AMI mortality, while lead had a marginally significant inverse association (ie protective).

When the broader category of mortality from ischaemic heart disease was assessed, both magnesium and calcium appeared to have significant protective effects, but this disappeared after adjustment for geographic gradients.

To test whether any effect of magnesium might be dependent on socioeconomic factors, RRs were calculated for men and women categorised by Carstairs index. It has been postulated that a protective effect of magnesium in water might be restricted to those with a deficiency of magnesium in their diet, so examination of more deprived socioeconomic groups may reveal such an effect. This analysis showed no evidence of a protective effect in any socioeconomic group. Similarly, analysis of different age groups showed no evidence of an age-specific protective effect of magnesium in either men or women. It might have been expected that any effect would be enhanced in elderly people who had retired from work and would be more likely to consume most of their drinking water at home.

Authors conclusions:
The results of this study do not support an association between magnesium concentration in drinking water and mortality from AMI in the study area. Similarly the results do not support an association with mortality from ischaemic heart disease once confounding and geographical factors have been taken into account.

Strengths and Limitations
Definition of populations
- Small area populations defined from census data.

Water exposure
- Assumed from geographic location (water quality zone).

Risk factors
- Adjusted for socioeconomic factors at small area level.

Validation of health outcomes
- Death statistics only.

Effects of medical treatment
- Not assessed.

Evaluation
Moderate quality.

Morris RW, Whincup PH, Lampe FC*, et al.* (2001)
Geographic variation in the incidence of coronary heart disease in Britain: the contribution of established risk factors *Heart* **86:** 277-283.

Aim: To determine the extent to which geographic variation in the incidence of major coronary heart disease in Great Britain may be explained by established risk factors. The paper focuses on individual risk factors such as smoking, physical activity, BMI etc, however some results on water hardness are also reported.

Design: Cohort study with 15 year follow up.

Location and population: 7,735 men from 24 towns in England, Scotland and Wales. The towns were selected to represent a wide range of CHD mortality following an initial survey of cardiovascular mortality rates in 253 towns.

Identification of cohort: In each of the 24 towns, over 400 men in the 40-59 year age group were randomly selected from GP practices and invited to attend a screening session. The response rate was 78%.

Time period: Recruitment occurred from 1978 to 1980.

Drinking water parameters:
Water hardness was measured between 1969 and 1973.

Source of water quality data:
Water quality data were provided by water authorities.

Risk factor information: Information was collected on smoking, alcohol, physical activity, social class and socioeconomic indicators.

Risk factor information source: At recruitment, participants underwent a medical examination, answered a questionnaire and gave a blood sample. Data collection during the follow up period consisted of postal questionnaires to participants after 5 years and 12-14 years.

Health outcomes: Major fatal and non-fatal CHD events. Fatal events were included if they were classified as ICD-9 codes 410-414.

Health outcome information source: Fatal events were identified through the National Health Service Register. GPs in the study reported cardiovascular events, and GP records were reviewed every two years to collect information.

Results:
The analysis showed that up to 75% of the variance in CHD rates between towns was explained by the combined effects of smoking, physical activity, systolic blood pressure, occupational social class, and height.
The effect of water hardness was non-significant (OR 0.96, 95% CI 0.88 – 1.05).

Authors conclusions:
Much of the variation in CHD incidence among British towns was accounted for by variables known to be related to individual risk.

Strengths and Limitations

Definition of cohort

- Identified at individual level.

Water exposure

- Assumed from place of residence (town).

Risk factors

- Adjusted for smoking, alcohol, physical activity, social class and socioeconomic status.

Validation of health outcomes

- National statistics and GP records.

Effects of medical treatment

- Not assessed.

Evaluation

Moderate quality.

Punsar S, and Karvonen MJ. (1979)
Drinking water quality and sudden death: observations from West and East Finland. *Cardiology* **64:** 24-34.

Aim: To examine the relationship between water quality and sudden death, with particular focus on magnesium, copper and chromium.

Design: Cohort study with 15 year follow up.

Location and population: 1711 men in two rural areas in western (888 men) and eastern (823 men) Finland.

Identification of cohort: All resident men born between 1900 and 1919 were recruited in 1959 (at age 40-59).

Time period: 1959 to 1974.

Drinking water parameters:
Magnesium, maximum 108 mg/L in the west, 23 mg/L in the east
Copper
Chromium

Source of water quality data: Households in the study areas use private wells. Water samples were collected from 93 wells in the western area and 234 wells in the eastern area during 1970, and analysed for a range of parameters. The study areas were divided into 10 subareas in the west and 33 in the east, and median values for water quality parameters were calculated for each subarea based on water samples for that subarea (3-20 water samples per subarea). All men within a subarea (2-125 men) were assigned the same water exposure level.

Risk factor information:
None.

Risk factor information source:
N/A

Health outcomes:
Deaths from coronary heart disease (CHD) and from other causes. Sudden death was defined as unexpected on the day of death and occurring within one hour of the onset of acute symptoms. The health status of those still alive in 1974 was categorised as having no CHD, having CHD or having other heart disease (HD).

Health outcome information source:
Death certificates and examination of survivors.

Results:
The overall death rate for the eastern region was higher than the western region (30.5% deaths vs 26.5% deaths) over the 15 year follow up. The difference was particularly marked for coronary deaths (14.7% vs 8.7%) and other cardiac causes (2.2% vs 1.4%). The death rate from other causes (non-cardiac) was less in the eastern region than in the west (13.6% vs 16.4%).

Median magnesium and chromium levels in water were lower in the east (where CHD mortality is higher) than in the west. Copper levels showed the opposite pattern, with higher levels in the east.

Median levels of magnesium, copper and chromium in water were compared for men who remained alive in 1974 and those who had died during follow up. Water data were not available for all subjects so the numbers analysed were 366 men from the western region and 427 from the eastern region. In the western region, chromium levels were significantly lower for those who had died from CHD than for those still alive. There were no significant differences for magnesium or copper. In the eastern region, magnesium levels were significantly lower for the categories of death from all causes and death from other causes but not for deaths from CHD. There were no significant differences for copper or chromium levels. (Note: the text (p29) contradicts Table IV on this point, stating that "in the eastern cohort the concentration of magnesium was statistically lower in the drinking water of the men who had died from CHD, as compared to the 'alive' category", however the table shows significant differences for the 'all deaths' and 'other cause' groups).

The analysis was repeated, breaking down the 'alive' category into 'no HD', CHD and other HD, and breaking down CHD deaths into sudden (1 hour or less after onset) and not sudden (more than 1 hour after onset). For the western region, chromium levels in water were significantly increased for those presently alive with CHD, compared to those presently alive with no HD. However chromium levels for those who had died of CHD tended to be lower although the differences were not significant. There were no significant differences for magnesium or copper. In the eastern region, chromium levels were significantly lower for those presently alive with CHD or with other HD, compared to those presently alive with no HD. Chromium levels were also significantly lower in the water of those who had died of CHD, compared to those presently alive with no HD. There were no significant differences for magnesium or copper. When comparing sudden deaths with other deaths, no significant differences were seen for magnesium, copper or chromium levels in water.

Other observations
The authors note that many other differences exist in levels of drinking water constituents between the two areas; nitrate, potassium, cobalt, nickel and barium occur at higher levels in the east, while calcium, sodium, fluoride and silicon are higher in the west.

Authors conclusions:
These data support that hypothesis that quality of drinking water plays a part in the difference in mortality from CHD between the two study areas. However they do not support the hypothesis that the alleged water factors operate through an increase in the frequency of sudden death.

Strengths and Limitations
Definition of cohort
- Subjects identified at individual level.

Water exposure
- Water quality was not measured at an individual level but classified by subarea (region around a village or group of farmhouses).

Risk factors
- Not assessed.

Validation of health outcomes
- Death certificates, medical records and interviews.

Effects of medical treatment
- Not assessed.

Evaluation
Low quality.

Rosenlund M, Berglind N, et al. (2005).
Daily intake of magnesium and calcium from drinking water in relation to myocardial infarction. *Epidemiology* **16**(4): 570-6.
Additional information on methodology also obtained from a previous paper (Reuterwall C, Hallqvist J et al. Journal of Internal Medicine (1999) **246**; 161-174)
Note: this paper had not been published at the time of the WHO 2003 review. An early abstract of this work was included in the WHO review (Rosenlund M. (2002) Drinking water hardness and myocardial infarction in the Stockholm Heart Epidemiology Program (SHEEP). *Epidemiology* **13:** S192).

Aim: To investigate the relationship between AMI and daily intake of magnesium and calcium from drinking water.

Design: Case-control study of AMI cases and population controls. This study is based on a subset of data from the Stockholm Heart Epidemiology Program (SHEP study) which is described as having a high participation rate, high reliability of case identification, and low probability of selection bias.

Location and Population: Stockholm county, Sweden. The area was restricted to 6 out of 26 municipalities in the SHEP study where large differences in magnesium levels in water would be expected (areas with small water works and some people using private wells). To be eligible, subjects had to have lived in the area for 2 years or more before inclusion in the study.

Identification of cases: Cases comprised all nonfatal and fatal first events of AMI among Swedish citizens aged 45-70 years, who resided in the study area. According to a previous paper on the SHEP study, cases were identified from records of hospital coronary and intensive care units, hospital discharge records and the death registry.

Identification of controls: Controls were selected from the population register and matched to cases on age, sex and hospital catchment area.

Time period: 1992-1993 (males) or 1992-1994 (females).

Drinking water parameters:
Hardness, range 0.4 – 88.2 German hardness degrees (dºH)
Calcium, range 1.0 - 610 mg/L
Magnesium, range 1.0 – 23.0 mg/L

Source of water quality data: Residential addresses of cases and controls were linked to water company records to identify their tap water source. Water quality information for 1990-1994 was obtained from waterworks records. For regions receiving tap water from two sources, the mean mineral content was calculated. Subjects who reported using a private well (n= 137), were posted a water sampling bottle and returned samples were analysed for magnesium, calcium and hardness.

Risk factor information: Information was collected by questionnaire covering:
Physical and psychosocial work environment including socioeconomic status, job strain
Lifestyle factors including physical activity
Dietary intake including calcium, magnesium, alcohol, coffee, fat and fibre
Water consumption
Smoking
Diabetes mellitus
BMI
Hypertension and antihypertensive drug therapy

Risk factor information source: Individual postal questionnaires, with supplementary telephone contact to collect missing data. Where a case had died, a close relative was asked to complete the questionnaire (proxy respondent), but not until at least 6 months after the death.

Health outcomes: First AMI

Health outcome information source: Records of hospital coronary and intensive care units, hospital discharge records and the national death registry. A panel of three physicians blinded to treatment assignment and blood pressure status reviewed the documentation of new cardiovascular events over the study period and adjudicated outcome events according to predetermined criteria.

Results:
The entire SHEP study included 2,246 cases and 3,206 controls. Response rates to questionnaires were 72% for female cases, 81% for male cases, 70% for female controls, and 75% for male controls.

The group included in this sub-study of a restricted geographic region comprised 1,327 people. Due to missing data on some people, the statistical analysis was performed on 452 cases and 666 controls (1118 people, number not stated in the paper but can be calculated from Table 1).

Statistical analysis – Odds Ratios and 95% CIs were calculated using unconditional logistic regression with adjustment for matching variables (age, sex, hospital catchment area). Drinking water parameters (Ca, Mg, hardness) were categorised in quartiles, or as being above or below the median value. The main multivariate model included adjustment for smoking, hypertension, socioeconomic status, job strain, diabetes mellitus, BMI, and physical activity.

No differences were seen between men and women in any of the initial analyses, therefore the combined data for both sexes were used in the main analysis.

There was no significant relationship for AMI risk with Mg, Ca or hardness levels in water regardless of how exposure levels were characterised. Odds Ratios tended to increase slightly as Mg levels increased, but there was no consistent trend for other parameters.
Analysis in terms of Mg or Ca intake from water (calculated from water consumption information) also showed no significant association with AMI risk.

A subgroup analysis was carried out for cases where death occurred within 28 days of AMI diagnosis. Exposure was defined in terms of daily magnesium and calcium intake from tap water for the 2 years prior to diagnosis. Again, no significant relationship with AMI risk was observed.

Use of different multivariate models either omitting cardiovascular risk factors (smoking etc), or including these factors together with additional adjustments for dietary intake of Ca and Mg (from food, coffee, tea, alcohol, soup) produced little change in risk estimates.

Classification of exposure using water quality data only during the year of AMI or the preceding year did not change the results. Reweighting of controls to compensate for possible overmatching effects of geography also had little effect. Adjustment for whether the subject had a regular job in the previous 2 years (a marker for water consumption outside the home) also did not change the results.

Other observations
Calculated mean intakes of magnesium were 276 mg/day from food and 2.6 mg/day from water.
There was no significant association between calcium or magnesium intake from food and risk of MI.

Authors conclusions:
The study does not support a substantial protective effect on MI associated with consumption of drinking water at moderate concentrations of hardness, magnesium or calcium. The range of calcium and magnesium levels examined in this study were lower than in some previous studies – this may explain why no protective effect was seen. Only 7% of people were exposed to Mg levels above 8.0 mg/L, and 16% were exposed to Ca levels of 28.5 mg/L or above.

Strengths and Limitations

Definition of cases and controls

- Cases identified as part of large study on heart disease. Said to have a high participation rate, high reliability of case identification, and low probability of selection bias.

Water exposure

- Individual water intake assessed. Also assessed intake of Mg and Ca from water, other beverages and food.

Risk factors

- Adjusted for smoking, hypertension, socioeconomic status, job strain, diabetes mellitus, BMI and physical inactivity.

Validation of health outcomes

- Expert adjudication of outcomes from blinded assessment of medical records.

Effects of medical treatment

- Not assessed.

Evaluation
High quality.

Rubenowitz E., Axelsson G., Rylander R. (1996)
Magnesium in drinking water and death from acute myocardial infarction. *Am J Epidemiol* 1996;143:456-462.

Aim: to study the relationship between the amount of magnesium (Mg) in drinking water (DW) and death from acute myocardial infarction (AMI) among males.

Design: case control study using men who died from AMI as cases and men died from cancer as controls.

Location and population: Sweden; 17 municipalities in 2 counties in southern Sweden (Skane and Blekinge, excluding the city of Malmö), where the water magnesium levels were different between municipalities as well as within those municipalities, and where the water quality concerning water hardness, pH, and water treatment procedures had been basically unchanged during the last 10 years.

Identification of cases: among all men who died between ages 50 and 69 years during the period 1982-1989, cases were defined as men with the diagnosis of AMI as underlying cause of death. 1237 cases were identified, and 854 were finally included into the study (383 excluded because of insufficient information on addresses or waterworks, less than 1 year at the last address, or using own private well). The median age of death in cases was 64 years.

Identification of controls: controls were defined as men of the same age group (50-69 y) in the same study area as the cases and with the diagnosis of cancer as the underlying cause of death. 1381 controls were identified, and 989 were finally included (392 excluded because of the same reasons as above). The median age of death in controls was 64 years.

Time period: mortality data collection during the 1982-1989 period.

Drinking water parameters: for each study subject, address and information on the waterworks that supplied the drinking water to this address were obtained from parish population registers and municipalities. Information on levels of magnesium (Mg) and calcium (Ca) was obtained from each waterworks (78 waterworks as a total number; between 1 and 15 waterworks engaged within the municipality), available only for the 1990-1991 years for most of the waterworks:

- Mg : 1.3 – 20.0 mg/l
- Ca : 22- 225 mg/l

Source of water quality data: the Mg and Ca values obtained from the waterworks were compared with values of Mg and Ca from analysis of one sample of tap water from the main waterworks of each minicipality. A strong correlation were observed, demonstrating that the content of Mg in the water did not change on the way from the waterworks to the households.

Risk factor information: no information collected on smoking, cholesterol levels, hypertension, diabetes, exercise, body mass index, socio-economic status.

Health outcomes: death from AMI.

Health outcome information source: underlying cause of death from the National Central Bureau of Statistics records, according to the International Classification of Diseases, WHO, 1977.

Statistical analysis : subjects were divided into quartiles according to the levels of Mg, Ca, and Mg/Ca ratio in the drinking water. ORs were calculated in relation to the group with the lowest

exposure (≤ 3.5 Mg mg/l), with adjustment for age, and in a second step for Ca or Mg in drinking water. Multivariate analysis using logistic regression model.

Results: age and Ca adjusted OR for death from AMI were significantly lower for the 2 groups with high levels of Mg in drinking water : OR = 0.70 (95%IC 0.53 – 0.93) in the 6.9-9.7 Mg mg/l group; OR = 0.65 (0.50 – 0.84) in the ≥ 9.8 Mg mg/l group. Trend analysis results were significant.
Age adjusted OR in relation to Ca levels showed slightly lower values with higher Ca level, but when adjusted for Mg levels, the differences between groups were not statistically significant, and trend analysis showed no significance for Ca level.
For the Mg/Ca ratio, the OR was significantly lower (0.70, 95%CI 0.53 – 0.93) only for the group with the highest ratio (≥ 0.22).

Other observations : the authors make the assumption that the Mg levels in 1990-1991 were about the same as during the period when the mortality data were collected (1982-1989), because only municipalities where the water source, quality, and treatment had been stable for the last 10 years. The Mg levels in the water from the waterworks with chlorination represented all quartiles.

Authors conclusions: the authors conclude that Mg in drinking water is an important protective factor for death from AMI among males.

Strengths and Limitations

Definition of cases and controls

- Possible misclassification of controls: controls were patients who died of cancer and we cannot formally exclude AMI as the cause of death in some controls.

Water exposure

- Water exposure assumed from residential address.

Risk factors

- Not assessed.

Validation of health outcomes

- Mainly from death records. Autopsies were carried out in only 23.6% of controls compared to 40.6% of cases.

Effects of medical treatment

- Not assessed.

Evaluation
Medium quality.

Rubenowitz E., Axelsson G., Rylander R. (1999)
Magnesium and calcium in drinking water and death from acute myocardial infarction in women. *Epidemiology* 1999;10:31-36.

Aim: to study the relationship between the amount of magnesium and calcium in drinking water and death from acute myocardial infarction (AMI) among women.

Design: case control study using women died from AMI as cases and women died from cancer as controls.

Location and population: Sweden; 16 municipalities in 2 counties in southern Sweden (Skane and Blekinge, excluding the city of Malmö), where the water magnesium levels were different between municipalities as well as within those municipalities, and where the water quality concerning water hardness, pH, and water treatment procedures had been basically unchanged since 1980.

Identification of cases: among all women who died between ages 50 and 69 years during the period 1982-1993, cases were defined as women with the diagnosis of AMI as underlying cause of death. 492 cases were identified, and 378 were finally included into the study (114 excluded because of insufficient information on addresses or waterworks, or using own private well).

Identification of controls: controls were defined as women of the same age (50-69 y) in the same study area as the cases and with the diagnosis of cancer as the underlying cause of death during the same period. 1706 controls were identified, and 1368 were finally included (338 excluded because of the same reasons as above). Women with cerebrovascular, respiratory, or digestive organs disease were excluded because of possible relation to Mg.

Time period: mortality data collection during the 1982-1993 period.

Drinking water parameters: for each study subject, address and information on the waterworks that supplied the drinking water to this address were obtained from parish population registers and municipalities. Information on levels of magnesium (Mg) and calcium (Ca) was obtained from each waterworks (80 waterworks as a total number; between 1 and 17 waterworks engaged within the municipality), available only for the 1990-1993 years for most of the waterworks:

- Mg : 1.3 – 21.5 mg/l
- Ca : 8 - 230 mg/l

Source of water quality data: the Mg and Ca values were obtained from the waterworks by a questionnaire sent to all 37 municipal offices. No sampling of tap water have been made.

Risk factor information: no information collected on smoking, cholesterol levels, hypertension, diabetes, exercise, body mass index, socio-economic stats, nor on “sudden death” risk factors during AMI (arrythmias, conduction defects, …) and on pre-hospital treatment occurrence (antiarrhytmic agents, thrombolysis, …).

Health outcomes: death from AMI

Health outcome information source: underlying cause of death from the National Central Bureau of Statistics records, according to the International Classification of Diseases, 1977.

Statistical analysis : subjects were divided into quartiles according to the levels of Mg, Ca, and Mg/Ca ratio in the drinking water. OR were calculated in relation to the group with the lowest exposure, with adjustment for age, and in a second step for Ca or Mg in drinking water. Multivariate analysis using logistic regression model.

Results: age and Ca adjusted ORs for death from AMI were significantly lower for the highest quartile of Mg levels (≥ 9.9 mg/l) : OR = 0.70 (95%IC 0.50 – 0.99),but not with the two other quartiles with lower Mg levels. ORs were consistently farther from the null for the younger group (50-59 years) than for the other group (60-69 years).
Regarding Ca, ORs for the whole group were significantly lower in all three upper quartiles, compared with the referent quartile. Adjustments for age and Mg altered the ORs only slightly : OR = 0.66 (95% CI 0.47 – 0.94) for the upper quartile (≥ 70 Ca mg/l); 0.61 (0.39-0.94) in the 32-45 mg/l quartile; and 0.71 (0.49-1.02) in the 46-69 mg/l quartile. ORs were farther from the null for the younger group (50-59 years) than for the other group (60-69 years).
For the Mg/Ca ratio, there was no difference of ORs between the quartiles.

Other observations : the authors make the assumption that the Mg levels in 1990-1993 were about the same as during the period when the mortality data were collected (1982-1993), because only municipalities where the water source, quality, and treatment had been stable since 1980. The OR for the quartile with the highest Mg level in this study was about the same for women as that reported for men in the earlier study (1996) with similar design in the same area.

Authors conclusions: the authors conclude that the results suggest that Mg and Ca in DW are important protective factors for death from AMI among women.

Strengths and Limitations

Definition of cases and controls

- Possible misclassification of controls: controls were patients who died of cancer and we cannot formally exclude AMI as the cause of death in some controls.

Water exposure

- Water exposure assumed from residential address.

Risk factors

- Not assessed.

Validation of health outcomes

- Mainly from death records. Autopsies were carried out in only 16% of controls compared to 41% of cases.

Effects of medical treatment

- Not assessed.

Evaluation
Medium quality.

Rubenowitz E., Molin I., Axelsson G., Rylander R. (2000)
Magnesium in drinking water in relation to morbidity and mortality from acute myocardial infarction. *Epidemiology* 2000;11:416-421.

Aim: to investigate the importance of individual levels of magnesium in drinking water in relation to both mortality and morbidity from acute myocardial infarction (AMI).

Design: case control study using men and women who had suffered an AMI as cases and subjects from the same study area as controls.

Location and population: Sweden; 18 municipalities in the catchment area of six hospitals in the southern part of Sweden. Levels of magnesium and calcium vary substantially between and within these municipalities. Water quality concerning hardness, pH, and treatment procedures had been basically unchanged since 1980 according to the previous study (1996). The study base was men and women born between 1920 and 1946, of Scandinavian origin, who had lived in any of the 18 municipalities since April 1994.

Identification of cases: cases were defined as men and women in the study base who during the period October 1 1994 through June 30 1996, suffered an AMI and at the time of the infarction were in the range of 50-74 years of age. 1086 cases were identified, including 823 survivors and 263 deceased cases.

Identification of controls: for each case, one control was randomly selected from the study base. These were men and women born 1 day after the birth date of the cases. Controls selected for deceased cases could either be alive or deceased. 1111 controls were selected, including 853 survivors and 258 deceased controls.

Time period: AMI mortality and morbidity data collection during the 1st October 1994 – 30 June 1996 period.

Drinking water parameters: for each subject, Mg and Ca content in the drinking water were identified at the last residence .

Source of water quality data: Information on levels of magnesium (Mg) and calcium (Ca) was obtained from each waterworks (N = 79). Water samples were collected from tapwater from households representing all the waterworks (N=79) and analysed for Mg and Ca. A mean was used of the values from the waterworks and the values from the analysed water samples. Water samples from all subjects using water filters 15 of the cases and 23 of the controls) or drinking water from private wells (144 of the cases and 167 of the controls) were also analysed. A total of 470 water samples were analysed. Range of concentrations :

- Mg : 0 – 44.0 mg/l
- Ca : 0 - 235 mg/l

Estimation of intake of Mg and Ca : Individual intake of Mg and Ca from food and beverages were estimated using a food frequency questionnaire. Individuals intake of water was covered by questions on consumption of water, coffee, and tea. Subjects were asked about their drinking water sources (waterworks or private wells, and considering also water source at work) and the use of water filters.

Risk factor information: telephone interviews were conducted by a experienced nurse with the surviving cases and the corresponding controls. The subjects were asked about weight and height, marital status, number of persons in the household, education level, profession, working hours, working conditions, physical activity, stress, smoking, health status (including high blood pressure and diabetes) and medication, and family history of AMI.

Health outcomes: suffering from AMI, surviving or deceased patients.

Health outcome information source: The cases were identified from the following sources:

- The clinical departments treating AMIs at the six hospitals in the area reported cases of AMI, for patients who had given informed consent.
- Hospital treatment records from the six hospitals were used to trace AMI patients who had not been reported by the clinical departments
- Registers at the Centre for Epidemiology at the National Board of Health and Welfare were used to identify cases who died from AMI during the study period and who had not been interviewed previously.

Statistical analysis: subjects were divided into quartiles according to the levels of Mg and Ca in the drinking water. The 3 lower quartiles were used as a referent. For Mg, the upper quartile corresponds to a Mg level ≥ 8.3 mg/l. New quartiles were calculated for men and women subgroups. ORs for AMI were calculated with surviving and deceased cases together and separately. Values were adjusted for age, and for Ca or Mg in drinking water respectively. Quartiles for survivors on the basis of the intake of Mg and ca calculated from the questionnaire were also formed. Covariate were controlled using a logistic regression model.
Deceased cases who used private wells were excluded from analysis (as were their controls) because no information was available on water quality.

Results: when all cases, survivors and fatalities, were included in the analyses, age and Mg or Ca adjusted ORs for suffering an AMI in relation to Mg or Ca level in DW were not significantly different from 1.0. Only among dead cases the OR for the risk of suffering an AMI in relation to Mg in DW for the highest quartile compared with the referent was significantly reduced (OR = 0.64 (95%CI 0.42-0.97)). In survivors, ORs for the risk of suffering an AMI was not significantly lower compared with the referent.

This comparison suggests that magnesium levels in water did not affect the incidence of AMI, but that mortality was reduced in high magnesium areas. However this analysis was carried out on an ecological basis without consideration of individual risk factors, or water consumption.

When Mg intake from water was used as the exposure (higher quartile for Mg > 13 mg/l), ORs for the risk of suffering an AMI were significantly increased in survivors: 1.33 (95%CI 1.05-1.68), and in the surviving male subgroup: 1.31 (95%CI 1.00-1.73).
ORs for women were lower than for men in all categories, but were not significantly different from 1.00. The total daily intake of Mg and Ca from food and beverages did not have an important effect on ORs.

The authors then go on to calculate Odds Ratios in relation to the individual risk factors (smoking, diet, socioeconomic status etc) and magnesium intake for AMI survivors and their controls (Table 4 in the paper). However these Odds Ratios reflect the overall risk resulting from two separate probabilities – 'having an AMI' and 'surviving the AMI event' long enough to be recruited into the study. It is not possible to separate the two probabilities or determine the effect of the individual risk factors on each of them because of the failure to collect the relevant information for dead cases and their controls.

The calculated OR in the final multivariate model for magnesium intake from drinking water was 1.39 (95%CI 1.07-1.80). This means that among surviving subjects, the proportion of AMI cases with a high magnesium intake was greater than the proportion of their corresponding controls with high magnesium intake. However it is not possible to conclude that this reflects a favourable effect of high magnesium intake on mortality from AMI, since the final OR reflects the net effect of changes in incidence or mortality or both. Thus a risk factor which increases the risk of suffering an AMI but does not adversely affect subsequent survival (compared to the average probability of survival among people who suffer an AMI), could produce the same OR as a risk which does not affect the risk of AMI

incidence but improves chances of survival. This can be illustrated by considering the results of the adjusted and multivariate analysis in relation to some established strong risk factors of atherosclerosis and AMI (smoking and diabetes). The calculated ORs for the risk of "suffering an AMI and surviving" for these exposures are also significantly greater than 1.

Due to the failure to collect risk factor information on dead cases and their controls, the findings of this study are essentially limited to the ecological observations in the initial analysis. The design flaw makes it impossible to carry out a proper adjustment for individual risk factors to determine their influence on AMI morbidity and mortality, relative to the apparent effects of calcium and magnesium.

The authors discuss the sudden death hypothesis and suggest their results support this postulated mechanism for a protective role of magnesium. However the study simply assessed all deaths and did not attempt to separately assess the sudden death subcategory. Given the fact that many cases (and presumably their matched controls) were not identified until many months after the AMI event (see below), it appears that the deaths could have occurred up to 22 months after the AMI event.

Other observations : It was intended that all AMI cases would be promptly reported to the study by the treating hospitals but only 38% of cases were identified in this way. The remaining 62% were identified later by review of hospital records or national death records. There was a large difference between these two groups in the time lag between the AMI and interviews for the study. For cases identified by hospitals the average time to interview was 35 (+/- 22) days, but for other cases it was 16.7 (+/- 5.1) months. The authors could not determine whether all the criteria for the diagnosis had been fulfilled in the cases with delayed identification.

Authors conclusions: the authors conclude that "Mg in drinking water did not affect the risk of suffering an AMI, but the probability of surviving the AMI was greater for those who had high Mg levels in their drinking water". The findings for calcium in drinking water were inconclusive.

Strengths and Limitations

Definition of cases and controls

- Inadequate description of control group - selection bias cannot be excluded.

Water exposure

- For dead cases and their controls, exposure was inferred on the basis of address. Where the dead case used a private well, no data were collected and the case was excluded from analysis.
- For surviving cases, water intake data was collected.

Risk factors

- Not able to be assessed due to failure to collect information on dead cases and their controls.

Validation of health outcomes

- A diversity of the terms is used to define the health outcome variable: "suffering an AMI", "suffering an AMI and surviving", "surviving from an AMI". This may lead to misunderstanding and confusion, because two different events are involved.
- Diagnosis criteria may not have been fulfilled in all cases due to delays in identification.

Effects of medical treatment

- Not assessed.

Evaluation
Low quality study.

Yang C-Y (1998).
Calcium and Magnesium in Drinking Water and Risk of Death from Cerebrovascular Disease. *Stroke* **29**(2): 411-4.

Aim: To determine the relationship between the levels of calcium and magnesium in drinking water and death from cerebrovascular disease.

Design: Case-control study

Location and population: 252 municipalities (administrative districts) in Taiwan in which the source of the tap water supply could be unequivocally determined. 109 districts were excluded - 30 aboriginal townships and 9 islets that had different lifestyles and living environments, and 70 districts with more than one waterworks where the exact population served by each could not be determined.

Identification of cases: Cases were obtained from records of the national death registry. Cases were defined as deaths occurring between 50 and 69 years of age and having cause of death attributed to specific ICD-9 codes relating to cerebrovascular disease (see Health outcomes below).

Identification of controls: Controls were also extracted from the national death registry. They were selected from among all other deaths excluding ICD-codes corresponding to the case definition, and also excluding hypertensive disease (ICD-9 codes 401-405), ischaemic heart disease (ICD-9 codes 410 - 414), diseases of pulmonary circulation (ICD-9 codes 415-417), other forms of heart disease (ICD-9 codes 420 - 429), diseases of the arteries, arterioles and capillaries (ICD-9 codes 440 - 448). Those who had died from gastric cancer (ICD-9 code 151) were also excluded because of previously reported negative association with drinking water hardness found by the same research group in the Taiwanese population.
One control was matched to each case by gender, year of birth and year of death. The authors state that “each case and its matched control had residence and place of death in the same municipality”, but do not state this as a matching criterion, therefore this statement may mean that each person lived and died within one municipality but that a case and its control may have been in different municipalities.

Time period: Deaths occurring from 1989 to 1993.

Drinking water parameters:
Calcium, range 4-81 mg/L
Magnesium, range 1.5 - 41.3 mg/L

Source of water quality data:
Data for 1990 collected from individual waterworks by a national research centre. Mean levels of calcium and magnesium were used, calculated from 4 seasonal samples over the year. This was assumed to represent long term exposure levels for people living in each area.

Risk factor information:
No information on individual risk factors.

Risk factor information source:
N/A

Health outcomes: Deaths from cerebrovascular disease (ICD-9 codes 430 - 438).

Health outcome information source: National death registry

Results:

Statistical analysis Subjects were divided into tertiles according to levels of calcium and magnesium in drinking water. Conditional logistical regression models were used to calculate Odds Ratios and 95% confidence intervals for the association between cerebrovascular death and calcium and magnesium levels. Final models included adjustment for urbanization level of residence (urban, rural) and quintiles of calcium or magnesium levels. The group in the lowest tertile of exposure was used as the reference group.

The data set analysed consisted of 17,133 cases and 17,133 controls (10,625 males and 6,508 females in each set). There was no significant difference in mean age, mean calcium level or mean magnesium level between cases and controls. There was also no difference in the percentage of population served by waterworks in the municipalities of residence, or in the urbanization level of residence.

Calcium The crude OR (adjusted for age and sex) for cerebrovascular death was significantly reduced at the highest tertile of calcium exposure but additional adjustment for magnesium level and urbanization resulted in a loss of statistical significance.

Magnesium Crude ORs (adjusted for age and sex) showed a statistically significant decrease in cerebrovascular risk for the second and third tertiles of magnesium levels relative to the lowest (reference) tertile. After additional adjustment for calcium level and urbanization, the significant difference remained.

Magnesium levels (mg/L) in tertiles (median)			
	<=7.3 (3.8)	7.4 – 13.4 (9.1)	13.5 – 41.3 (17.3)
Adjusted OR	1.0	0.75 (0.65-0.85)	0.60 (0.52 – 0.70)

Authors conclusions:

There is a significant protective effect of magnesium intake from drinking water on the risk of death from cerebrovascular disease. However due to the ecological nature of exposure assessment in this study, future studies are needed which assess individual intake of calcium and magnesium, and possible confounding factors such as smoking and hypertension.

Strengths and Limitations

Definition of cases and controls

- Death records and population register.

Water exposure

- Assumed from place of residence.

Risk factors

- Not assessed.

Validation of health outcomes

- Death records only.

Effects of medical treatment

- Not assessed.

Evaluation

Low quality.

Yang C-Y and Chui H-F (1999).
Calcium and magnesium in drinking water and risk of death from hypertension. *American Journal of Hypertension* **12**: 894-899.

Aim: To study the relationship between levels of calcium and magnesium in drinking water and risk of death from hypertension.

Design: Case-control study

Location and population: 252 municipalties (administrative districts) in Taiwan in which the source of the tap water supply could be unequivocally determined. 109 districts were excluded - 30 aboriginal townships and 9 islets that had different lifestyles and living environments, and 70 districts with more than one waterworks where the exact population served by each could not be determined.

Identification of cases: Cases were obtained from records of the national death registry. Cases were defined as deaths occurring between 50 and 69 years of age and having cause of death attributed to specific ICD-9 codes relating to hypertension (see Health outcomes below).

Identification of controls: Controls were also extracted from the national death registry. They were selected from among all other deaths excluding ICD-codes corresponding to the case definition, and also excluding ischaemic heart disease (ICD-9 codes 410 - 414), diseases of pulmonary circulation (ICD-9 codes 415-417), other forms of heart disease (ICD-9 codes 420 - 429), cerebrovascular disease (ICD-9 codes 430 - 438), diseases of the arteries, arterioles and capillaries (ICD-9 codes 440 - 448). Those who had died from gastric cancer (ICD-9 code 151) or colorectal cancer (ICD-9 code 153) were also excluded because of previously reported negative associations with drinking water hardness found by the same research group in the Taiwanese population.
One control was matched to each case by gender, year of birth and year of death. The authors state that “each case and its matched control had residence and place of death in the same municipality”, but do not state this as a matching criterion, therefore this statement may mean that each person lived and died within one municipality but that a case and its control may have been in different municipalities.

Time period: Deaths occurring from 1990 to 1994 inclusive.

Drinking water parameters:
Calcium, range 4-81 mg/L
Magnesium, range 1.5 - 41.3 mg/L

Source of water quality data:
Data for 1990 collected from individual waterworks by a national research centre. Mean levels of calcium and magnesium were used, calculated from 4 seasonal samples over the year. This was assumed to represent long term exposure levels for people living in each area.

Risk factor information:
No information on individual risk factors.

Risk factor information source:
N/A

Health outcomes:
All hypertension-related deaths were analysed together. Causes of death comprised essential hypertension (ICD-9 code 401), hypertensive heart disease (ICD-9 code 402), hypertensive renal

disease (ICD-9 code 403), hypertensive heart and renal disease (ICD-9 code 404), secondary hypertension (ICD-9 code 405).

Health outcome information source:
National death registry.

Results:
Statistical analysis Subjects were divided into quintiles according to levels of calcium and magnesium in drinking water. Conditional logistical regression models were used to calculate Odds Ratios and 95% confidence intervals for the association between hypertension death and calcium and magnesium levels. Final models included adjustment for urbanization level of residence (urban, rural) and quintiles of calcium or magnesium levels. The group in the lowest quintile of exposure was used as the reference group.

The data set analysed consisted of 2,336 cases and 2,336 controls (1,500 males and 836 females in each set). There was no significant difference in mean age, mean calcium level or mean magnesium level between cases and controls. There was also no difference in the percentage of population served by waterworks in the municipalities of residence. Cases were significantly more likely to live in metropolitan municipalities than controls.

Calcium Crude ORs (adjusted for age and sex) showed a decreased risk for hypertension death for all calcium levels above the lowest quintile, although the difference was statistically significant only in the 4th quintile.
In the final model (adjusting additionally for urbanicity and magnesium level), the risk of hypertension death was increased in all quintiles above the reference quintile, although none of the differences were statistically significant. ORs did not show an increasing trend as calcium levels increased.

Magnesium Crude ORs (adjusted for age and sex) showed a statistically significant decreased risk for hypertension death for all magnesium levels above the lowest (reference) quintile. ORs were very similar (0.79 or 0.80) for all four higher quintiles.
In the final model (adjusting additionally for urbanicity and calcium level), the risk of hypertension death showed a statistically significant decrease for all magnesium levels above the lowest (reference) quintile. ORs were lower then for the crude analysis, and there was a significant trend for decreasing risk with increasing magnesium level.

Magnesium levels (mg/L) in quintiles (median)					
	1.5-3.8 (3.5)	3.9-8.2 (7.0)	8.3-11.1 (9.1)	11.2 – 16.3 (13.5)	16.4 – 41.3 (19.4)
Adjusted OR	1.00	0.73 (0.57-0.93)	0.66 (0.50-0.87)	0.67 (0.50-0.89)	0.63 (0.47-0.84)

Other observations
In Taiwan the mean dietary intake of calcium is 507 mg. This is about 82% of the recommended daily intake. Therefore calcium in drinking water at average levels of 34.7 mg/L may make an important contribution to total intake.
There is no information on dietary magnesium intake in Taiwan or the relative contribution from drinking water.

Authors conclusions:
There may be a significant protective effect of magnesium intake from drinking water on the risk of hypertension. However due to the ecological nature of exposure assessment in this study, future studies are needed which assess individual intake of calcium and magnesium, and possible confounding factors such as sodium, potassium and alcohol.

Strengths and Limitations

Definition of cases and controls

- Death records and population register.

Water exposure

- Assumed from place of residence.

Risk factors

- Not assessed.

Validation of health outcomes

- Death records only.

Effects of medical treatment

- Not assessed.

Evaluation

Low quality.

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