WATER STORAGE IN RURAL HOUSEHOLDS:
INTERVENTION STRATEGIES TO PREVENT
WATERBORNE DISEASES

NATASHA POTGIETER
WATER STORAGE IN RURAL HOUSEHOLDS: INTERVENTION STRATEGIES TO PREVENT WATERBORNE DISEASES

by

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Submitted in partial fulfilment of the requirements for the degree

PHILOSOPIAE DOCTOR
PhD (Medical Virology)

In the Faculty of Health Sciences
Department of Medical Virology
University of Pretoria
Pretoria
South Africa

February 2007
I, the undersigned, declare that the thesis hereby submitted to the University of Pretoria for the degree PhD (Medical Virology) and the work contained therein is my own original work and has not previously, in its entirely or in part, been submitted to any university for a degree.

Signed ___________________, this the _______ day of ____________________ 2007.
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DEDICATION

Just enough light

Sometimes only the step I’m on,
or the very next one ahead,
is all that is illuminated for me.

God gives just the amount of light I need
for the exact moment I need it.

At those times I walk in surrender to faith,
unable to see the future,
and not fully comprehending the past.

And because it is God who has given me
what light I have,
I know I must reject the fear and doubt
that threaten to overtake me.

I must determine to be content where I am,
and allow God to get me where I need to go.

I walk forward,
one step at a time,
fully trusting that the light God sheds,
is absolutely sufficient.

(Stormie Omartian, 1999)

I dedicate this work to my Lord and Saviour, Jesus Christ
He is shaping and building my character each second of my life.
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My students at the Department of Microbiology, University of Venda who assisted and encouraged me with their enthusiasm and interest in the field of Health and Water related Microbiology since 1997.

My parents, for believing in me and encouraging me to reach for the top, and for always making me feel like a winner. Johan, Jaun Pierre, Johan Jr, Andre, Adel, Ruan, Dinky, Marius, Edelweiss and Rene for loving me and bringing joy, happiness and love to my life.
WATER STORAGE IN RURAL HOUSEHOLDS: INTERVENTION STRATEGIES TO PREVENT WATERBORNE DISEASES

by

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DEPARTMENT: Medical Virology, Faculty of Health Sciences
DEGREE: PhD (Medical Virology)

SUMMARY

Poor sanitation, unhygienic practices and close living associations between people and animals in rural communities increase the risk of zoonoses and add to faecal contamination of stored drinking water. Point-of-use interventions can improve the microbiological quality of household drinking water and a combination of microbial and chemical indicator tests could identify the origin of faecal pollution. The improvement of the microbiological quality of drinking water in rural households by the implementation of intervention strategies which included the use of traditional storage containers as well as an improved safe storage container (CDC, USA), with or without the addition of a sodium hypochlorite solution were determined. The origin of faecal contamination in the water sources and household stored water were determined using male specific F-RNA subgroup genotyping. This study attempted to assess the survival of indicator microorganisms and selected bacterial pathogens and viruses in the improved safe storage container in borehole and river water samples.

An intervention study was conducted in two rural villages utilising different source water. Results indicated that the improved safe storage container without the addition of a stabilized sodium hypochlorite solution did not improve the microbiological quality of the stored drinking water and had counts of indicator microorganisms similar to that found in the traditional storage containers. However, the households using the 1% and the 3.5% sodium hypochlorite solutions have shown an effective reduction in the counts of indicator microorganisms in both the traditional and the improved safe storage containers. The compliance with the use of the sodium hypochlorite interventions ranged between 60% and 100%, which was in agreement with similar studies carried
out in other developing countries. One village complied with the intervention while the other village did not. Reasons for this included financial factors, an unsupportive infrastructures and lack of education and knowledge on health risks by the households.

Male specific F-RNA bacteriophage genotyping showed that faecal contamination in the water source samples and both the traditional and improved safe storage containers at the point-of-use were primarily of animal origin (Subgroup I). Households using river water had subgroup II F-RNA bacteriophages present in the stored household water, which was associated with human faecal pollution. However, subgroup II F-RNA bacteriophages has been isolated from faeces of cattle and poultry, which indicated that F-RNA subgroup typing might not be a specific tool to determine the origin of faecal pollution in water sources.

Laboratory seeding experiments indicated that 1% sodium hypochlorite solution were less effective in reducing heterotrophic bacteria, *Escherichia coli*, *Salmonella typhimurium*, *Clostridium perfringens*, F-RNA bacteriophages and coxsackie B1 virus counts in the improved safe storage containers filled with river water with a high turbidity. However, the 1% sodium hypochlorite solution did reduce the indicator and seeded microorganisms within 60 min in containers filled with borehole water with a low turbidity. The 3.5% sodium hypochlorite solution effectively decreased the numbers of microorganisms to undetectable limits within 60 min in both the borehole and river filled storage containers irrespective of the turbidity values. This study has showed that a combination of intervention strategies can provide rural communities with microbiologically safe drinking water.

**Keywords:** improved safe storage container, F-RNA genotyping, intervention strategies, microbiological quality; compliance, sustainability, sodium hypochlorite solution, waterborne diseases.
DIE STOOR VAN WATER IN PLATTELANDSE HUISHOUDINGS:
INTERVENSIE STRATEGIEË OM WATEROORDRAAGBARE SIEKTES TE VOORKOM

deur

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DEPARTEMENT: Geneeskundige Virologie, Fakulteit Gesondheidswetenskappe
GRAAD: PhD (Geneeskundige Virologie)

OPSOMMING

Swak sanitasie, higiene en ‘n noue verblyf verhouding tussen mense en diere in plattelandse gemeenskappe veroor- draag van soonosis en dra by tot die fekale besoedeling van gestoorde drinkwater. Intervensies in die huishoudings en ‘n kombinasie van chemiese en mikrobiologiese indikatore toetse kan moontlik ‘n aanduiding gee van die oorsprong van fekale besoedeling. Verbeteringe in die mikrobiologiese kwaliteit van die huishoudelike drinkwater met die instelling van intervensies soos ‘n verbeterde huishoudelike stoorhouer (CDC, VSA) en die gebruik van ‘n natrium hipochloriet oplossing was ondersoek. Die oorsprong van die fekale besoedeling van die water was bepaal deur gebruik te maak van molekulêre hibridisasie van die F-RNA bakteriofaag isolate. Hierdie studie het ook die oorlewing van indikator en geselekteerde patogene mikroorganismes in die verbeterde huishoudelike stoorhouer gevul met boorgat- en rivierwatermonsters bepaal.

‘n Intervensie studie in twee plattelandse dorpe met verskillende waterbronne was onderneem. Die resultate het gewys dat die verbeterde huishoudelike stoorhouers sonder die gestabiliseerde natrium hipochloriet oplossing nie die mikrobiologiese kwaliteit van die gestoorde water in die huishoudings verbeter nie en het dieselfde mikrobiologiese tellings getoon as die traditionele stoorhouers. Desnieteenstaande het die houers waarby die 1% en die 3.5% natrium hipochloriet oplossings gevoeg is, bewys dat die mikrobiologiese tellings van indikator organismes afgeneem het in beide die verbeterde huishoudelike en die traditionele stoorhouers. Die gebruik van die
natrium hipochloriet oplossings in die huishoudings het gewissel tussen 60% en 100% wat in ooreenstemming was met soortgelyke studies in ander ontwikkelende gemeenskappe. Die intervensie was volhoubaar met een van die studiegroepe maar nie met die ander studiegroep nie. Redes hiervoor het faktore soos onvoldoende finansies, swak infrastrukture en onvoldoende kennis aangaande gesondheids risikos in die huishoudings ingesluit.

Die manlik spesifieke F-RNA bakteriofaag geentiperking het bewys dat fekale besoedeling hoofsaaklik van dierlike oorsprong (supgroep I) was in die waterbron en ook in beide die verbeterde huishoulike en die traditionele stoorhooiers. Huishoudings wat water vanaf die rivier gebruik het, het ook supgroep II faag isolate gehad wat gassosieer word met menslike fekale oorsprong. Nie te wel, supgroep II faag isolate is al geïsoleer uit beeste en pluimvee se mis monsters en dit bewys dat F-RNA bakteriofaag molekulêre hibridisasie nie sodanig ’n spesifiek genoeg metode is om te gebruik om die oorsprong van fekale besoedeling in watermonsters te bepaal nie.

Oorlewings studies in die laboratorium het bewys dat 1% natrium hipochloriet oplossing nie effektief was om Escherichia coli, Salmonella typhimurium, Clostridium perfringens, F-RNA bakteriofage en coxsackie B1 virus tellingsin die verbeterde huishoulike stoorhooiers wat gevul was met rivierwater met ’n hoë turbiditeit, te verminder nie. Die 1% natrium hipochloriet oplossing het wel die tellings van indikatore en geselekteerde patogene in boorgatwater met ’n lae turbiditeit binne 60 min verminder. Die 3.5% natrium hipochloriet oplossing het suksesvol die tellings van indikatore en geselekteerde patogene in beide rivier- en boorgatwater binne 60 min verminder ongeag die turbiditeits waardes van die waterbronne. Hierdie studie het bewys dat ’n kombinasie van intervensie strategiêe wel mikrobiologies veilige drinkwater kan verskaf aan plattelandse gemeenskappe.

**Kern woorde:** verbeterde huishoulike stoorhooier, F-RNA molekulêre hibridisasie, intervensie strategiêe, gebruik; volhoubaarheid; mikrobiologiese kwaliteit; natrium hipochloriet oplossing, wateroordraagbare siektes.
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<td>Amplified Fragment Length Polymorphism</td>
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<td>Avian Myeloblastosis Virus</td>
</tr>
<tr>
<td>AOC</td>
<td>Assimilable Organic Carbon</td>
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<tr>
<td>ARDRA</td>
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<td>Clostridium perfringens</td>
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<td>CDP</td>
<td>disodium 2-chloro-5-4 (methoxyspiro{1,2-dioxetane-3,2’,5’-chloro) tricycle [3.3.1.1.3.7] decan}-4-yl)-1-phenyl phosphate</td>
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<td>Fluorescent Whitening Agents</td>
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<td>HAV</td>
<td>Hepatitis A Virus</td>
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<td>HCl</td>
<td>Hydrochloric Acid</td>
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<td>HH</td>
<td>Household</td>
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<td>ISO</td>
<td>International Standardization Organization</td>
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ITS-PCR - Internal Transcribed Spacer Polymerase Chain Reaction
KCl - potassium chloride
km - kilometer
l - litre
LAB - Long Chain Alkylbenzenes
mg - milligram
MgCl₂ - Magnesium Chloride
min - min
ml - millilitre
mm - millimeter
mM - milli Molar
MUG - 4-methyl-umbelliferyl-β-D-glucuronidase
ng - nanogram
NaCl - Sodium Chloride
NaOH - Sodium Hydroxide
NCTC - National Culture Typing Collection
NGO - Non Govermental Organisation
nm - nanometer
NTU - Nephelometric Turbidity Units
PAHO - Pan American Health Organization
PBS - Phosphate Buffered Saline
PCA - Plate Count Agar
PCR - Polymerase Chain Reaction
PEG - Polyethylene Glycol
PFGE - Pulsed Field Gel Electrophoresis
pfu - plaque forming unit (s)
PLC/PRF/5 - Primary Liver Carcinoma continuous cell line
pmol - picomol
% - percentage
RFLP - Restriction Fragment Length Polymorphism
RNA - Ribonucleic Acid
rpm - revolutions per minute
RSA - Republic of South Africa
RT-PCR - Reverse Transcriptase Polymerase Chain Reaction
s - second
SABS - South African Bureau of Standards
SDS - Sodium Dodecyl Sulfate
SSC - Saline Sodium Citrate
S. typhimurium - Salmonella typhimurium
<table>
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<tr>
<td>2.1</td>
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Potgieter N, Musie E, Obi CL and Du Toit PJ (2000) Evaluation of different growth media for the recovery of sulfide reducing anaerobic Clostridium perfringens from the environment. Poster presentation at the WISA Biennial Conference, 28 May-1 June, Sun City, South Africa.


