Norwegian University of Life Science
Environment and Natural Resources

Master's Thesis 2017
60 ECTS

The effect of freezing and thawing on transport of pesticides through macroporous soils and the potential risk towards the aquatic environment

Frederik Norheim Bøe

Preface and Acknowledgement

This thesis is written as a part of the Norwegian Research council (RCN) funded project SMARTCROP.

I would first and foremost like to thank my supervisors Ole Martin Eklo (NIBIO/NMBU), Marianne Stenrød (NIBIO) and Hans-Christian Teien (NMBU/CERAD) for all the help and support throughout the year. Their constructive feedback and expertise have been very helpful in all parts of my thesis. I am much obliged to have been offered a part of this project. The work on this thesis has awakened a field of interest and given me a better understanding on the fate and transport of pesticides in the environment.

Secondly, I would like to thank Roger Holten (NIBIO/NMBU), who has overseen the planning of the project and are taking the topic on the transport of pesticides in cold climate conditions a step further through his ongoing PhD thesis. Thank you for the countless hours spent in the freezing laboratory where much of the theoretical aspects of the topic were discussed and for making this year a lot more enjoyable.

I would also like to thank the employees that are working at the section of Pesticides and Natural Products Chemistry, especially Marit Almvik, who performed the pesticide analysis, Randi Bolli and Ellen Aarrestad Vartdal for giving me an introduction on operational practices and help when needed in the laboratory.

Finally, a special thanks to Stine Egeli Øvrebø, friends and family for all the support and encouragement.

Frederik Norheim Bøe

14th of May 2017

Oslo

Abstract

Pesticides and their metabolites can leach rapidly through agricultural soils through macropores, be transported further into surface waters and potentially cause negative effects in aquatic organisms. Cold climate with freezing and thawing of soils can affect the leaching. The transport of pesticides during winter and early spring have however received little attention and there is a need to assess the impact of cold climate conditions to get a better understanding of the risk of contamination.

The objective of the work has been to provide knowledge on how freezing and thawing affects leaching of pesticides through agricultural soils with well-developed macropore structures. To obtain information of water transport in the soil, bromide was included as a non-reactive tracer. Ten undisturbed soil columns were collected from an agricultural soil known to contain macropore structures in Ås, Norway. The mobile herbicide MCPA and the tracer bromide was applied to all soil columns and five soil columns were frozen at -3 ° C while five where stored cold at +3 ° C. During the experiment the soil columns were subjected to four irrigation events. The leachate was collected continuously during the leaching experiment and concentration of MCPA and bromide were measured for both frozen and unfrozen soil columns, and used to assess the effects of four consecutive freeze-thaw cycles on pesticide and bromide leaching through macroporous soils.

The results showed that there was more bromide leaching from the frozen than the unfrozen soil columns during the first irrigation, while the leaching from the unfrozen soil columns was higher during later irrigation events. The total amount of bromide leached through frozen soil columns after four irrigations varied between 35.0 % and 43.1 % of the applied amount, but between 48.1 % and 72.3 % through the unfrozen soil columns. The major part of the bromide was transported through the frozen soil columns during the 1st and 2nd irrigation, while during the 3rd irrigation through unfrozen soil columns. The earlier arrival of bromide from frozen soil columns indicated a faster transport of bromide through the frozen soil columns. However, there was no significant difference between the frozen and unfrozen soil columns during the 1st and 2nd irrigation. Compared to bromide a different trend in the proportions of leaching were observed for the transport of MCPA, were the total amount of MCPA leached after four irrigations was significantly higher from frozen than unfrozen soil columns. Total

3

amount of MCPA leached of the applied amount after four irrigations varied between 16.4 % and 32.9 % from frozen soil columns and between 0.1 % and 0.8 % from unfrozen soil columns. The leaching of MCPA were largely transported through the frozen soil columns during the 2nd and 3rd irrigation and during the 1st irrigation through unfrozen soil columns. The leaching of MCPA through unfrozen soil columns might be explained by the expected air-filled macropores caused initial macropore flow. The increased total amount of MCPA leached through frozen soil columns compared to unfrozen soil columns occurred although the total leachate was lower. This could be explained by the freezing and thawing of the soil caused efficient transport of MCPA through bypassing sorption sites. Thus, results show that freezing and thawing contribute to increased leaching of MCPA. Furthermore, it is essential to understand underlying mechanisms to reduce the potential risk in the future with the expected increase in winter cereal productions and use of pesticides in the late autumn.

Table of contents

List of abbreviations

K_{OC}	Soil organic carbon sorption coefficient
K_d	Soil sorption coefficient
K_{OW}	Octanol – water partition coefficient
K_f	Freundlich adsorption coefficient
LC_{50}	50 % Lethal Concentration
EC_{50}	50 % Effect Concentration
LOEC	Lowest Observed Effect concentration
NOEC	No Observed Effect Concentration
PEC	Predicted Environmental Concentration
TER	Toxicity Exposure Ratio
RCP 4.5	Median emission scenario
TU	Toxic unit
Eutectic point	Lowest melting point of a mixture of solutes
a.i	Active ingredient
PPP	Plant Protection Product
MF-value	Concentration level which indicates potential environmental effects in aquatic organisms
WRB	World Reference Base of Soil Resources
BTC	Breakthrough curve

1. Introduction

Pesticides and their metabolites should disappear rapidly after terminating its target whether it is weeds, fungal diseases or insect pests. Conversely, pesticides and their metabolites has been found in Norwegian surface waters at levels that could potentially harm aquatic organisms. Pesticides are mainly transported to surface water by runoff, spray drift and leaching via drains through complex interactions (Walker et al., 2012). Some main factors contributing to the leaching of pesticides are pesticide- and soil properties, rainfall, soil moisture (Flury, 1996), preferential pathways (Kladivko et al., 1991) and freeze – thaw cycles (Hayashi et al., 2003).

The Norwegian Agricultural Environmental Monitoring Program (JOVA) which aim to document effects of agricultural practices on soil and water quality in small agricultural catchments in Norway collects data on nutrient run-off, soil erosion and pesticide losses. JOVA monitors 11 catchments throughout Norway, selected to be representative of the current agricultural practices. Six of these catchments include pesticide monitoring of agricultural streams. The Norwegian Food Safety Authority (Mattilsynet) make use of the gathered information from JOVA in the evaluation and approval of pesticides used on agricultural landscape in Norway. In addition, the Norwegian government use the findings in agricultural policy. JOVAs focus on pesticides is based on its potential threat towards the aquatic environment. Concentration levels indicating potential negative effects to aquatic organisms (MF-values) are estimated for each individual pesticide (Stenrød et al., 2014). Exceedance above the MF-value indicate a potential risk towards the aquatic environment. In the period between 1995 to 2010, ten percent of the analysed water samples extracted from streams exceeded the individual pesticides limits (Stenrød, 2015).

Agricultural soils with well-developed macropore structures could be susceptible for a rapid transport (preferential flow, macropore flow) of pesticides to tile drains (Solbakken et al., 2006), possibly contaminating surface waters both heavily and rapidly (Kladivko et al., 1991; Klaus et al., 2014). One international system for classification of soil types, the World Reference Base of Soil Resources (WRB) has been implemented as national classification system in Norway (VKM, 2015). By 2008 the classification system includes 270 soil units divided into 13 soil types (WRB groups) (Sperstad & Nyborg, 2008).

Northern part of Scandinavia is characterized by cold climate conditions with seasonally frozen soils (Stenrød et al., 2016). In such areas, freezing and thawing processes influence the infiltration and transport of water and pesticides through soil (Hayashi et al., 2003). Pesticide monitoring programs in the northern zone has not been prioritizing monitoring of pesticide leaching and runoff during winter seasons (Stenrød et al., 2016). However, studies show that losses of pesticides to runoff and leaching do occur also in the winter and early spring. In a study performed on silty clay loam, traces of phenoxy-acids were detected in the runoff in the late spring, ten months after application in a field catchment area in Ullensaker, Norway. Low soil temperatures inhibit the degradation of the easily degradable pesticide. Thawing through warmer temperatures and heavy rainfall causing pesticide runoff and leaching to tile drains and subsequent surface waters (Eklo et al., 1994). This is supported by Bechmann & Eggestad (2016), who argues that losses of pesticides may be of a great concern if precipitation fall on partially frozen soil. Studies show that preferential flow could be an important factor in seasonally frozen soils in transporting solutes and pesticides through soils (Stadler et al., 2000; Derby & Knighton, 2001). However, the knowledge on leaching of pesticides in cold climate conditions is limited (Stenrød et al., 2008).

Pesticides applied in the late autumn, e.g. winter cereal applied pesticides, are especially at risk due to the short time-period between pesticide application and the impact of cold climate conditions with below zero soil temperatures, subsequently inhibiting the degradation of the pesticide. Winter cereal production in Norway has however declined by 70 % in the period between 2008 to 2012 (Bechmann et al., 2014). In eastern Norway, winter cereal, mainly winter wheat, dominated 10 % and 6 % in 2006 and 2010 of the cereal production, respectively. The plant protection products (PPP) that was the most used in winter wheat in 2011 and 2014, was the sulfonylurea Hussar OD with the pesticide active ingredient (a.i.) iodosulfuron and Express SX (a.i., tribenuron-methyl) (Aarstad & Bjørlo, 2016). However, several pesticides are relevant to the winter wheat productions including the widely-used fungicide Proline EC (a.i., prothioconazole) and the post-harvest Roundup products (a.i., glyphosate). PPPs used to control late blight in potatoes, such as Revus (a.i., mandipropamid), are also applied in the late autumn.

In a future scenario, global warming might impact freeze-thaw cycles (Bechmann & Eggestad, 2016) both in magnitude and frequency (Mellander et al., 2007). Additionally, increasing temperatures (Hanssen-Bauer et al., 2015) are expected to prolong the sowing period for winter cereal, thus increase areas with winter cereal productions and subsequent pesticides applied during the late autumn (Seehusen et al., 2016). Global warming and a change in climate towards warmer and more moist conditions, could also expedite problems such as fungal plant disease and thus increasing the use of fungicides in winter cereal. The late current trend of decrease in winter cereal production in Norway might be reversed and hence, increased use of pesticides during the autumn and an increased risk of contamination of surface waters and groundwater could be a future scenario.

There is a general lack of knowledge on the fate and transport of pesticides during winter and early spring. Pesticides applied to winter cereal and potatoes are especially at risk of leaching and the challenge may be even greater due to the changing climate. Thus, there is a need for data to be better able to tackle the present and future challenges of leaching of pesticides under the influence of cold climate conditions.

1.1 Objectives and Hypothesis

As part of the RCN funded project SMARTCROP (www.smartcrop.no), which aim to develop tools and measures to achieve successful adoption of integrated pest management as outlined in the EU-directive for sustainable use of pesticides (Directive 2009/128/EC), the main objective of this thesis is to provide knowledge on how freezing and thawing influence leaching of pesticides from agricultural soils with well-developed macropore structures. In addition, a discussion of the risk towards the aquatic environment will be addressed. The results in this study will be included in a dual-permeability model (MACRO) for optimizing the simulation model to include winter conditions. The main objective has been addressed through the following:

- A lysimeter study with undisturbed soil columns dominated by macropores (Albeluvisol) subjected to four freeze – thaw episodes using the conservative tracer bromide (Br) and the mobile herbicide 4–chloro–2–methylphenoxy acetic acid (MCPA) with moderate irrigation rate.
- Discuss the extent of the problem (if any) of the effect of freezing and thawing on leaching of pesticides through soil.

The hypothesis for this study:

- There is no significant effect of soil freezing on leaching of pesticides through soil macropores during subsequent rainfall and thawing events.

2. Background

The following sections focuses on the fate and transport of pesticides in soil, a widespread soil type in Norway, cold climate conditions and risk assessments processes relating pesticide exposure and toxicity (Fig. 1).
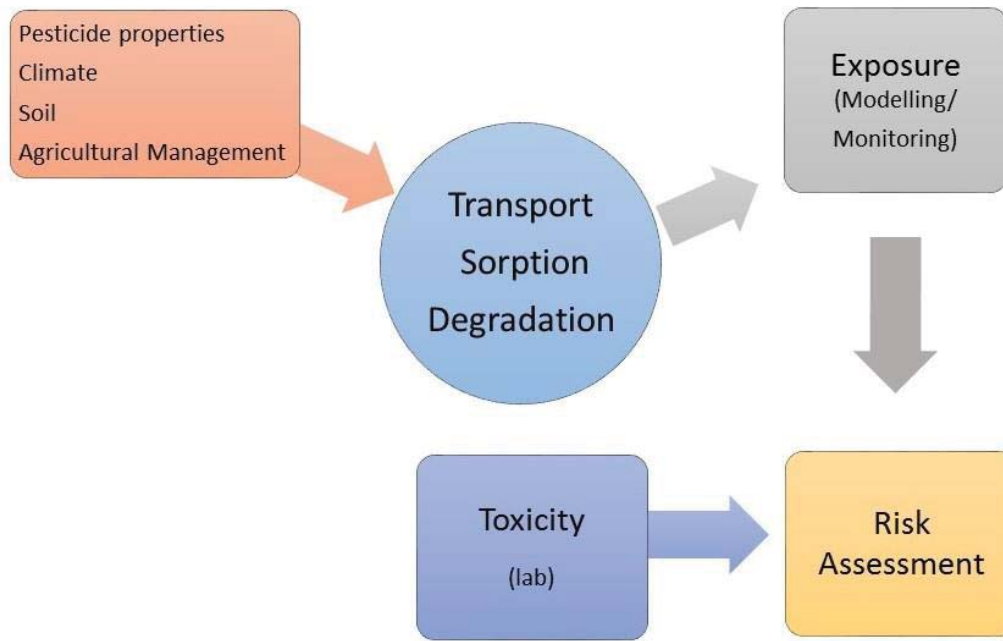
Fig. 1. Processes and factors affecting exposure of pesticides in soil. (Made by Frederik Bøe. Based on lectures in FMI310 Environmental pollutants and ecotoxicology, NMBU).

2.1 Pesticide fate in soil

The contamination of surface waters and groundwater by pesticides can originate from both regulated release (non-point source, e.g. normal application of pesticides) and unregulated release (point source, e.g. accidents and spills) (Walker et al., 2012). Further, pollution of surface waters (rivers, streams and oceans) are mainly due to surface runoff and drain flow through leaching (Fig. 2). Spray drift through air is also an alternative route for pesticides to reach surface waters (Walker et al. 2012). Annual pesticide losses to surface water can vary from 0.1 % to several percent, where pesticide losses due to leaching through soil is generally considered smaller than losses by runoff. However, pesticide leaching below the root zone after heavy rainfall shortly after application can in some scenarios reach 5 % of the applied amount (Flury, 1996).

Processes affecting the dissipation and exposure of pesticides in the environment are degradation, sorption and transport (Thorstensen & Lode, 2001a). Chemical and biochemical processes contribute to the degradation of pesticides in the environment. Hydrolysis, oxidation, photo degradation, photolysis and isomerization are chemical transformations which affect the pesticides residence time in the soil. The degradation rate is influenced by environmental factors such as climate, temperature, pH, solar radiation and properties of the pesticides. Furthermore, microorganisms metabolize pesticides and hence limit its persistence, migration and bioaccumulation (Walker et al. 2012).

Pesticides may sorb to clay, organic matter and iron or aluminium oxides possibly restricting the migration of pesticides. The most important factors contributing to sorption behaviour in soils are water solubility, polarity and acid-base constant (VKM, 2015). The top soil (A horizon) is generally rich in organic matter and therefore more fertile due to higher biological activity than the soil below. Hence, the soil microbial capacity to degrade pesticides are in general high. However, increased organic matter or organic carbon (OC) in the soil are in general associated to an increased soil sorption capacity. Studies show different response depending on the nature of the pesticides; pesticide with low sorption affinities are in general more prompt to degradation with increased OC and hence increased microbial activity, while pesticide that shows stronger sorption affinities to soil degrades less due sorption and decreased bioavailability (VKM, 2015). Clay, which is characterized with a silica-alumina crystal structure with negative charged sites (vanLoon & Duffy, 2011), are often associated with cations such as iron and magnesium. Consequently, at low pH, where iron oxides exist in its protonated form, the net negative charge of the clay soil may decrease (VKM, 2015). Pesticides are carried downward by the movement of water, especially for hydrophilic pesticides. In general, volatilization, breakdown processes and biotransformation tend to be slower in cold climate than tropical climate (Walker et al. 2012). Fig. 2 illustrates the fate and transport of pesticides in soil.
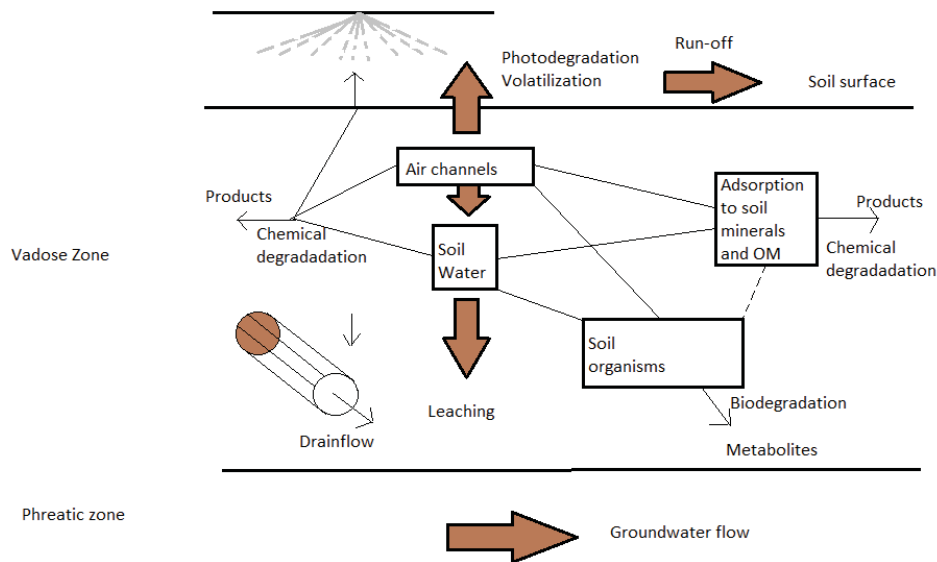
Fig. 2. Fate and transport of pesticides in agricultural soils (Made by Frederik Bøe. Modified after Walker et al., 2012).

Pesticides that are mobile and transported with waters are easily diluted and degraded (Walker et al. 2012). Lipophilic pesticides (high K_{ow}) associated with particulate matter are present in water bodies adsorbed to sediments. Conversely, hydrophilic pesticides dissolves in the water. Pesticides are in general bioavailable to water living organisms and animals when present in the free form. However, this topic is widely debated (Walker et al., 2012).

2.2 The dominating soil type Albeluvisol

The soil type Albeluvisol is one of the top dominating soil type on agricultural land in Norway and dominates 21.6 % of the mapped agricultural land (VKM, 2015). The WRB soil are widespread all over Norway, especially in boreal zones where cereal production dominates. These soils have mainly been developed from marine deposits and the texture varies from clay loam to silty clay loam. Albeluvisol dominates 31.6 % of the mapped agricultural land (2577 km^2 pr. 1.1.2014) in Eastern Norway south including; Østfold, Vestfold, Akershus, Oslo, Buskerud and parts of Telemark (VKM, 2015). In areas where clay soils are common, Albeluvisol and Stagnosol are often both present (Sperstad & Nyborg, 2008) (Appendix 6).

The most common subunit of Albeluvisol is the Epistagnic Albeluvisol (Siltic), which is periodically saturated with water due to a decreasing number of macropores with depth and dense beds (layers) stopping the vertical transport of water. Such an event often occurs after a heavy rainfall during summer or the melting of snow during spring. Trenches are needed to remove excess water. Albeluvisols are characterized by clay wash-out which follows verticals pores (Solbakken et al., 2006). Organic carbon (OC) is usually ~ 2.5 % in the Ap-layer and the pH of the soil in its natural condition is usually between 4-5 (Sperstad & Nyborg, 2008). However, flooded soils may produce reducing conditions where oxygen is depleted. A lighter-coloured bed with dark spots can be seen due to the alternating state between oxidized and reduced conditions ($Fe^{2+} \leftrightarrow Fe^{3+}$) (Sperstad & Nyborg, 2008). The Stagnic Albeluvisols are known to contain macropores (Kvernø, 2011) and are especially suitable for cereal production. Further, the degree of macropore flow through structural loam and clay soils is considered high (Larsbo et al., 2014) and well-known in loamy soils (Flury, 1996). Due to an update in WRB, the group Albeluvisol was removed. Poorly drained soils (e.g. Epsitagnic Albeluvisol) will in most cases be classified as Stagnosols (e.g. Retic Stagnosol) (Solbakken E., e-mail, 16. January 2017). "Albeluvisol" is further used.

2.3 Transport of water and pesticides through the unsaturated zone

The unsaturated zone (vadose zone) is the part of the soil that are not fully saturated with water between the surface and the top of the unsaturated zone (phreatic zone) (Fig. 2). The most important flow mechanisms in the unsaturated zone are uniform flow through the soil matrix and non-uniform flow through macropores causing stable and irregular wetting fronts, respectively (Hendrickx & Flury, 2001).

2.3.1 Uniform flow and breakthrough curves

Flow theories such as Richard's equation for flow through unsaturated soil and the convection-dispersion equations based on Fick's law for transport of solutes, are widely used to describe the transport of solutes in the unsaturated zone through the soil matrix. Processes involved in the mass flow of solutes are convection, dispersion and diffusion (Wilson et al., 1995). Convection is defined as the movement of solutes with the bulk water. Movement and reactions that occur within the soil solution are governed by processes such as diffusion and dispersion. Diffusion occurs in the direction from high to low concentration gradient and is especially important at low water velocities (Hillel, 2004). The nature of the soil (e.g. pores,

shapes, sizes etc.) causes variable flow velocities that results in mixing of solutes within the pore water. This process is denoted as dispersion. The convective stream dominates the flow regime when the flow of water is high (Wilson et al., 1995). Furthermore, convection transport is a dominating process in homogenous soils (Russo & Dagan, 1993).

Breakthrough curves (BTCs) are a useful way of describing the movement of pesticides through soil. Relative concentration and pore volume are non-dimensional parameters often used to present the BTCs (Tarradellas et al., 1997). For ideal conditions, vertical BTCs characterizes the shape of a convective dominated stream, increased dispersion rotates the BTC to the right (Fig. 3 A), sorption or retardation delay the transport of pesticides (Fig. 3 B), while BTCs characterized by degradation processes plateaus before reaching the relative concentration of one (Fig. 3 C) (Tarradellas et al., 1997).

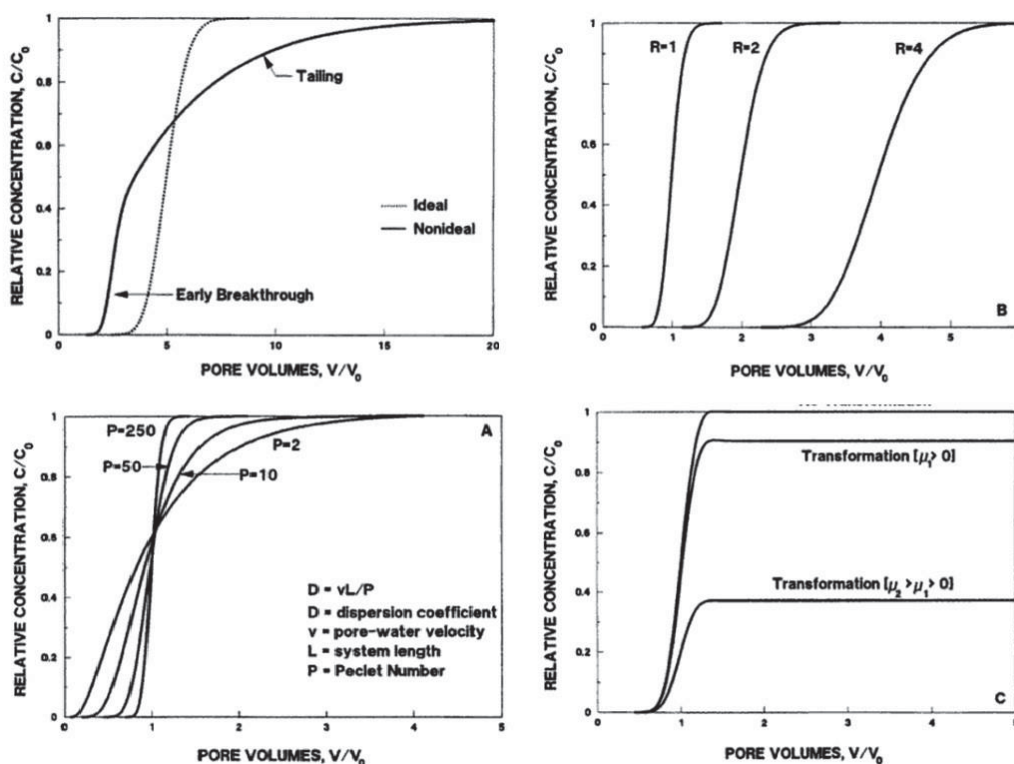*Fig. 3. Breakthrough curve illustrating ideal and non-ideal flow through soil (upper left picture) and breakthrough curves illustrating the effect of dispersion (A), retardation (B) and transformation (C) (Source: Tarradellas et al., 1997)*

The mentioned classical theories in describing transport processes in the unsaturated zone have some shortcomings with regards to leaching of pesticides through heterogenous soil

containing preferential flow pathways, especially to flow regimes in structured soils containing macropores (Beven & Germann, 1980; van Genutchen, 1987). Hence, the topic on preferential flow pathways has received increasing attention the last decade (Beven & Germann, 2013).

2.3.2 Non-uniform flow: Preferential flow through macropores

Preferential transport of water and pesticides is a preferred pathway of flow in which water and solutes moves predominately through a fraction of the soil's volume (Larsson et al., 1999) (Fig. 4). Macropores do contain microsites that are biologically active and chemically reactive, however, during transport, their small surface areas limit the degree of sorption to these sites (Jarvis, 2007). Thus, the risk of leaching increases due to a diminishing effect of adsorption and degradation (Larsson et al., 1999). Furthermore, impermeable coatings limit the degree of mass exchange between the macropores and the surrounding soil matrix (Jarvis, 2007). Preferential flow explains why pesticides have been detected in field drains within short time of field application (Larsbo, 2005). Thus, BTCs are usually characterized by having an early breakthrough and a late tail time (Fig. 3. Non-ideal) (Tarradellas et al., 1997). Simunek et al. (2003) argued that macropore flow occurs when precipitation exceeds the matrix infiltration capacity. The potential and the degree of preferential flow of water and pesticides are governed by the nature of the soil, including soil properties, biota activity, agricultural activity, precipitation and soil water content (Jarvis, 2007).
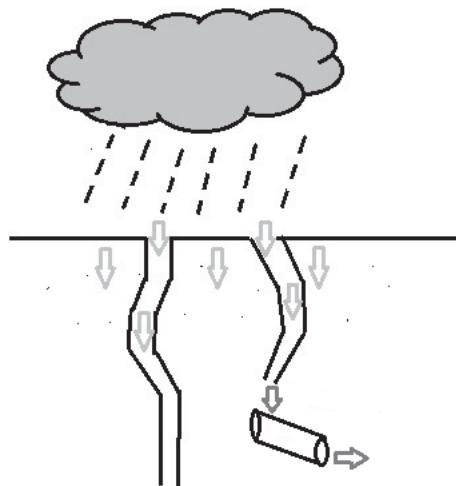
Fig. 4. Macropore flow occurs when the matrix infiltration capacity is exceeded by the rainfall. (Made by Frederik Bøe. Simplified after Larsbo, 2005)

Macropores are denoted as preferred pathways of flow. Beven & Germann (1980) defines macropores as "large continous openings in the soil" with width larger than 4 mm absent from

capillary forces. However, macropores are often discussed without stating the pore size. These preferential pathways are usually formed through biological processes such as holes made from earthworms and root networks and frost expansion (Beven & Germann, 1980). Hence, fields with no-till practice may have well-developed macropore systems (Shipitalo et al., 1989). Studies have demonstrated the importance of macropores. Clothier and White referred in Beven & Germann (2013) showed that 95 % of the water ran through larger continuous pores that only occupied 0.32 % of the soil. However, the role of macropores as preferred pathways diminishes as the wetting front penetrates deeper into the soil (Hillel, 2004). Heavy rainfall on soil that is initially wet, usually results in a rapid transport of solutes through open macropores connected to the surface (Jarvis, 2007; Larsbo et al., 2014). Macropore flow may also occur on initially dry soil (Jarvis et al., 2008). However, Shipitalo et al., (1989) demonstrated that atrazine and bromide were transported into the soil matrix, thus immobilizing the pesticide by the first rainfall event after application.

2.3.3 The influence of cold climate on the transport of water and solutes

Freezing and thawing affects the transport of water through the vadose zone in a complex manner. Natural soils contain a range of different poresizes, thus, when the soil starts to freeze, pore water in the larger pores freezes before pore water in the smaller pores. The water in the larger pores is held at a higher-pressure potential than the pore water in smaller pores, resulting in the present of both water and ice in the soil. Conversely, water present in the smaller pores will thaw before water in the larger pores (Ireson et al., 2013). Fig. 5 shows the soil freezing characteristics curve for a saturated soil which illustrates the decrease in liquid water content when the temperature decreases below zero (Spaans & Baker, 1996). However, in unsaturated soils where ice, liquid water and air coexists, the soil freezing characteristics curve are too simplistic. Further, because of the decrease in soil liquid water content when freezing (Fig. 5), the hydraulic conductivity drops (Ireson et al., 2013).
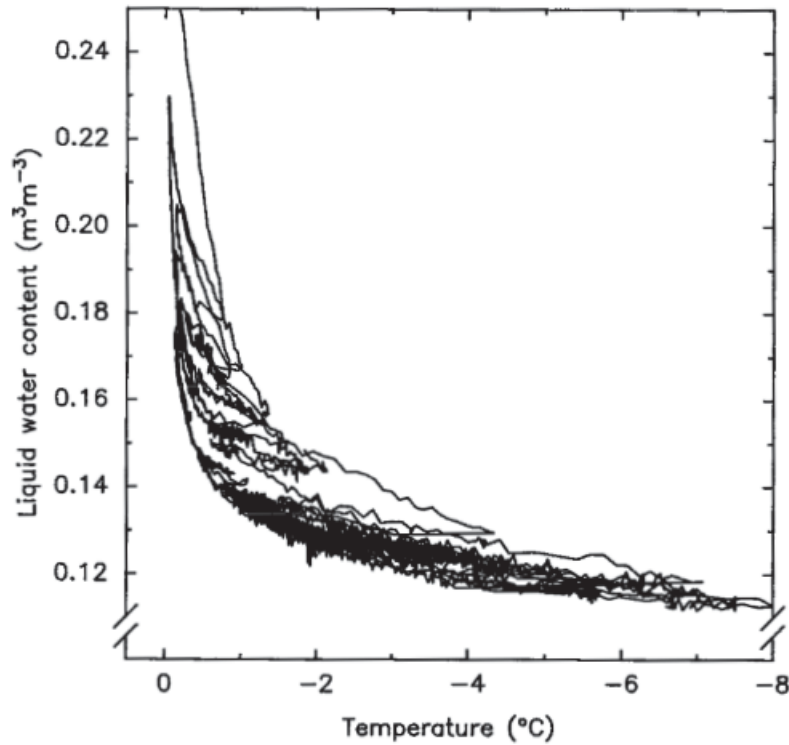
Fig. 5. Soil Freezing Characteristics Curve (SFC). Liquid water content as a function of temperature (Source: Spaans & Baker, 1996)

The presence of solutes lowers the freezing point in soil pore water (van der Kamp et al., 2003), contributing to the coexisting of both ice and liquid water (Ireson et al., 2013). Furthermore, as the soil freezes from the top-down, solutes are rejected into the unfrozen parts of the soil (Stähli & Stadler, 1997). Decreasing the temperature further, the pesticide too will freeze (Ireson et al., 2013).

In the northern hemisphere, the infiltration of snowmelt and rainfall into seasonally frozen soils are important events in the hydrological cycle. The infiltration of water into the frozen soil are affected by several factors including temperature, frost depth, snow cover, soil water content prior to freezing (Iwata et al., 2011) and degree of ice-filled pores (Ireson et al., 2013). While thick frozen soil layers will increase surface runoff of spring snowmelt, thin frozen soil layers will allow snowmelt to infiltrate the soil profile (Iwata et al., 2011). van der Kamp et al. (2003) showed that a well-developed macropore network present in the soil could increase the soil infiltrability of the frozen soil. They compared snowmelt infiltration in cultivating soils with few macropores to grass soils with larger macropores in the Canadian

prairies. During snowmelt, significant runoff occurred in the cultivating soil, whereas most of the snowmelt infiltrated through the large continuous macropores in the grass soil.

There are studies indicating that macropore structures are important flow pathways of the transport of water and solutes through partially frozen soils. Derby & Knighton (2001) reported that snowmelt in combination with heavy rainfall transported a non-reactive tracer chloride predominately through the soil as thawing progressed. Stadler et al. (2000) showed how preferential pathways in undisturbed soil columns transported the dye tracer through pores that were initially open to the air. In the same study, frozen packed sand columns that were initially wet, inhibited the infiltration of the irrigated water. In a study reported by Ulén et al. (2014), glyphosate was detected in the drainage water after a cold period with proceeding snowmelt.

2.4 Pesticide risk assessment

Pesticides leaching to surface waters and groundwater may impact both ecosystems and human health (Klaus et al., 2014), thus evaluation of exposure and effect of pre-existing and new pesticides entering the market is necessary to meet safety standards and assure proper protection. Pesticide approval and risk assessment processes are described by the regulation (EC) No 1107/2009 and the Water Framework Directive (WFD) (Commision Regulation, 2011).

2.4.1 Pesticide approval and risk assessment process

The regulation (EC) No 1107/2009 describes criteria for authorizing plant protection products (PPPs) in the EUs member state(s). Pesticide approvals are based on risk assessments of pesticide active ingredients carried out by the Authority (The European Comission, 2009). The regulation (EC) No 1107/2009 was implemented in Norway in 2015 (FOR-2015-05-06). The Norwegian Food Safety Authority (Mattilsynet) authorizes PPPs, whereas the pesticide active ingredient is approved at the EU-level. The Norwegian Scientific Committee for Food Safety (VKM) assists in the risk assessment (VKM, 2017).

Section 2.8.2 stated in the Commission Regulation (EU) No 546/2011, argues that the member states of the EU should consider long-term and short-term toxicity risk towards

terrestrial and aquatic organisms. Evaluations of exposure can be performed by calculating Toxicity Exposure Ratios (TER) for algae, daphnia and fish. The most sensitive organism is considered. Assessing the acute toxicity/exposure ratios (TER) to daphnia and fish, the ratio of the acute 50 % effect or lethal concentration (EC_{50}, LC_{50}) and the predicted short-term environmental concentration (PEC) or measured environmental concentration (MEC) is considered. PEC are values validated at the EU level. No authorization is given for the acute exposure of fish and daphnia if the TER is below 100.

$$TER_{acute} = \frac{EC, LC_{50}}{PEC} \qquad [1]$$

The no observed effect concentration (NOEC) and the predicted long-term concentrations for daphnia and fish are considered while assessing the long-term TER. No authorization is given for the long-term exposure of fish and daphnia if the TER is below 10.

$$TER_{long\text{-}term} = \frac{NOEC}{PEC} \qquad [2]$$

(Commision Regulation (EU) No 546/2011, 2011).

There are several methods to calculate the risk towards the aquatic environment. Toxic units (TU) correspond to the Toxicity Exposure Ratio (TER). There is a minimal risk towards aquatic organisms if the TU is below 0.1 and 0.01 for long-term and acute exposure, respectively.

$$TU = \frac{MEC}{EC_{50}} \qquad [3]$$

 (Petersen et al., 2013)

The European Food and Safety Authority (EFSA) provides guidance documents (GD) on how the member states should perform risk assessments on both active substances and PPPs. A tiered approach is considered in the authorization of PPPs which implies a cost-effective process with an appropriate endpoint. The risk assessments start with simple worst-case evaluations (tier 1) and increases in complexity (higher tiers) (European Food Safety Authority, 2013). The Norwegian Food Safety Authority usually carries out tier 1 by calculating the conservative TER (VKM, 2012).

In contrast to regulation (EC) No 1107/2009, the Water Framework Directive (European Commision, 2000) considers a "weight-of-evidence" approach when assessing the effect.

WFD uses environmental quality standards (EQS), which has been developed for each pesticide active ingredient to which reaching and maintaining "good ecological" and "good chemical" status is an underlying goal. The most sensitive organisms are assumed. The EQS are based on the predicted no effect concentration (PNEC) in compliance with appropriate assessment factors which are calculated according to the Technical Guidance Document on risk assessment (European Commission, 2003).

In addition to PPPs, the legislating WFD tackles a whole range of toxins. In comparison to a prospective approach in the PPP regulation, exposure assessment in WFD follows a retrospective approach. The exposure assessment is carried out when monitoring data and the EQS is compared (European Food Safety Authority, 2013).

2.4.2 Pesticide risk assessment in the JOVA program

In JOVA, evaluation of the environmental risk of measured environmental pesticide concentrations is carried out using a concentration level indicating potential environmental effects (MF-value) for each pesticide based on the predicted no-effect concentration (PNEC) from toxicity tests of aquatic organisms obtained from various databases (e.g. Pesticide Properties DataBase (PPDB) and the EU Pesticide Database) (Bechmann et al., 2014). Each MF-value is calculated based on measurements of toxicity at three trophic levels; algae/water plants, invertebrates (daphnia) and fish. The MF-values are calculated similar to the EUs quality standards (EQS). In contrast to EQS, which are based on mean year concentrations, MF-values are used as single- or mixture samples from periods of 14 days (Bechmann, 2014). Assessment factors (AF) are applied to account for different variations between different species and internal factors such as age and gender. However, validity of tests and lack of chronic designed tests may affect the accuracy of the application of AFs (Andersson & Kreuger, 2011).

3. Methods and materials

3.1 Soil type and soil sampling

Site for soil sampling was chosen to represent an agricultural soil with well-developed macropore structures. Soil was sampled from an agricultural field with winter wheat dominated by silty clay loam (Epistagnic Albeluvisol) in Kroer, Akershus County (UTM 32V PM 035 133) (Fig. 6).

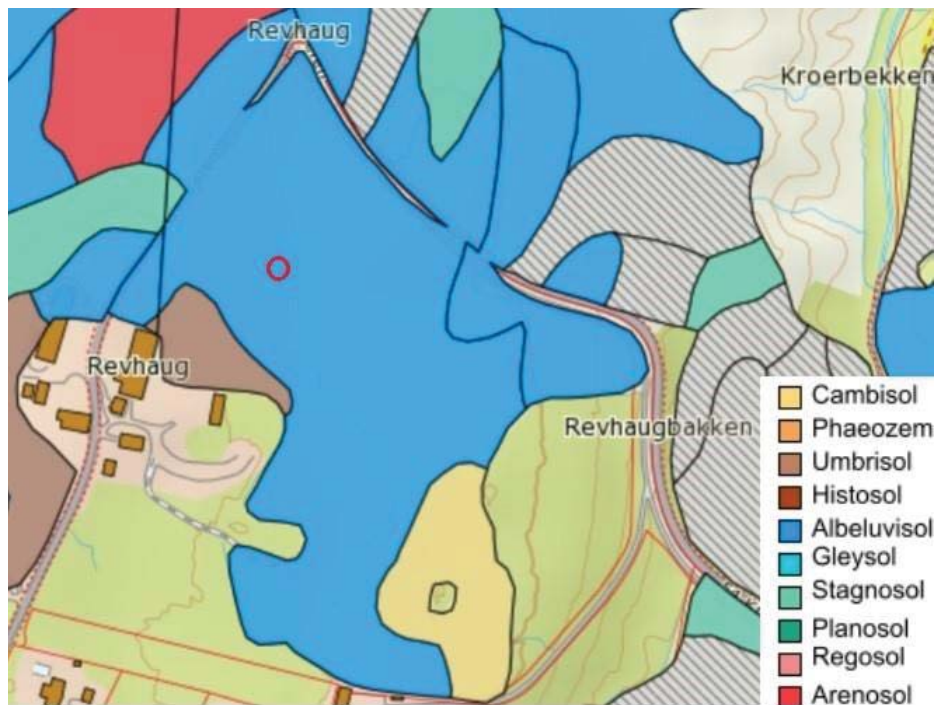*Fig 6. Map showing the sampling site at Kroer, Norway. The soil type Albeluvisol dominates the field (source: kilden.nibio.no, retrieved 28.04.2017)*

The characteristics of the sampled soil has previously been investigated and is described as a Nordic Reference Soil, representing cereal production in south-eastern Norway and Trøndelag (Greve et al., 1998). The main characteristics of the soil are given in table 1.

Table 1. Soil characteristics (grain size distribution (clay, silt and sand), soil classification, horizon, pH, organic carbon, and cation-exchange capacity (CEC)) at the sampling site in Kroer.

Property	Value
Horizon	0 – 23 cm
Soil texture	Silty clay loam
Classification WRB (2006)	Epistagnic Albeluvisol (Siltic)
Clay	19.1 %
Silt	43.8 %
Sand	37.1 %
Organic Carbon %	2.5 %
Cation Exchange Capacity (CEC)	13.2 cmol/kg
pH	5.5

Adaption from (Greve et al., 1998)

The soil samples were collected on the 13th of May 2016. Thirteen undisturbed soil columns were gathered from the topsoil (0–20 cm) using aluminium cylinders with dimensions 20 cm in height and 10 cm in outer diameter. To preserve the soil's macropore structure during sampling of the columns, the cylinders were carefully forced by hand and by use of a sledgehammer into the soil while removing the soil around the columns, until the top of the columns were at same level as the soil surface. An angled "shoe-knife" at the bottom of the columns assisted in penetrating the soil. The soil samples where gathered within a radius of approximately 10 meters. The excavations followed a straight line from where compaction from tractor wheels were not visible. The soil columns were trimmed down to a height of approximately 18 cm to assure the possibility of ponding when irrigating and sealed at the bottom using plastic caps and duct tape. Plastic was wrapped around the columns to assure no disturbances from light while storing and to preserve the soil moisture. The soil columns were transported to the fridge at NIBIO (+2–4 °C) for storage until the preparation of the soil columns. Fig. 7 illustrates a cross-section of a soil column with visible macropores and indications of rust stains.

Fig. 7. Cross section of the bottom of a soil column from Kroer of the macroporous soil type Epistagnic Albeluvisol (siltic). Macropores, produced from biological processes are visible and connected to the open air at the bottom of the 20-cm column. Indication of rust stain colours the soil. Photo: Roger Holten

The soil columns were scanned using x-ray tomography at the Department of Soil and Environment at the Swedish University of Agricultural Science in Uppsala before the experiment started. The x-ray tomography was carried out to quantify the macropores in the soil. Unfortunately, the image processing is not ready, thus no quantification of macropores were done in this thesis. However, the plug- in module SoilJ in ImageJ (Koestel, 2016) was used to construct a visual illustration of a soil column (Fig. 8). The x-ray images are being used in an ongoing PhD thesis regarding modelling.
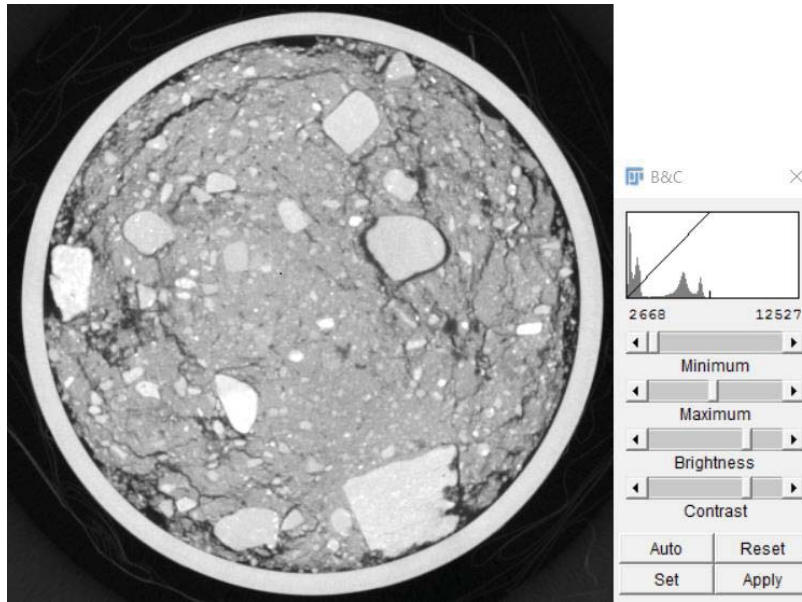
Fig. 8. Cross-section of an X-ray tomography of the soil sampled in Kroer.

3.2 Preparation of the soil columns

The ten soil columns of the best quality were used in the experiment (similar to Fig. 7). Soil columns that were considered best conserved after the soil sampling were used in the experiment. Loose soil columns with apparent weak soil structure and soil columns with larger irregularities were not used. Soil columns with smaller irregularities were adjusted using a small knife at the soil columns bottoms to ensure an even surface. Gas bands were attached at the bottom of each soil column using rubber bands to prevent soil loss. The sand box (Eikjekamp) was used in the preparation of the soil columns in two steps to ensure a comparable soil moisture content in all soil columns and to allow the macropores to become air-filled at the start of the experiment. First, the ten soil columns were pushed gently down onto one sandbox, before water was added such that the bottoms of the soil columns were covered (~ 2 cm). This was done two to four days ahead of applying the suction to allow the soil columns to absorb water due to their dry conditions after storage. The soil moisture content was measured in one soil column using a moisture meter (HH2 Delta-T Moisture Meter) as an indicator of the soil moisture (Appendix 4). The measured soil moisture indicated an increase in soil water content of approximately 5 %. Secondly, the soil columns were equilibrated at - 30 cm pressure potential (suction) for 7 to 9 days. The suction at - 30 cm was considered as sufficient pressure potential to attain equilibrium in the soil columns. The amount of water drained from the sandbox was monitored daily. Suction was removed when no water was observed in the collection vessel.

The ten soil columns where randomly divided into two groups of five, where five soil columns were chosen to be subjected to the freezing treatment. These five soil columns were inserted with thermistors into the prepared holes at 7 and 14 cm from the top. An awl assisted in carefully penetrating the soil approximately 5 cm towards the centre. The thermistors were installed to monitor the freeze-thaw cycles and to ensure that the soil columns were fully frozen at the start of irrigation. The temperature logger with thermistors were provided by ITAS "Instrumenttjenesten AS". DL2 control panel and DL2 program editor was used as recommended by ITAS. The temperature was logged every 15 minutes throughout the whole experiment. The five soil columns with inserted thermistors where then insulated with 4 cm polyethylene foam camping mats to allow the soil columns to freeze from the top-down as it would occur in the field. All ten soil columns were stored at NIBIO at +2-4 °C until the start of the experiment.

3.3 Artificial rainwater

The composition of the solution stock used to make artificial rainwater are given in table 2. The salts used are similar to the composition of natural rainwater. The solution stock was diluted 1:1000 with deionized water to produce two 20 litres' containers with artificial rainwater. The pH of the stock solution was approximately 2, while the mixed 20 litres' containers reached pH of approximately 5. Litmus paper was used as an indicator to achieve satisfying pH. The artificial rainwater was kept in the cooling room at 2-4 °C, except when the experiment was conducted.

Table 2. Overview of salts and measured weight used to make 1 litre stock solution of artificial rainwater.

Salt	Stock solution (g/L)
NaCl	0.58
$(NH_4)_2SO_4$	0.70
$NaNO_3$	0.50
$CaCl_2$	0.57
HCl (37 %)	0.95 ml/l

3.4 Selection of model pesticide

The model pesticide chosen in the experiment is the polar organic herbicide MCPA which is applied to a range of cereals (wheat, barley etc.). The phenoxyacid affects the growth regulation in weeds (EFSA, 2008). The dissipation of MCPA can be explained by its inherent properties such as low organic carbon – water partitioning coefficient K_{oc} value at 74 ml/g (mean) and relative short degradation half-life (DT_{50lab}) at 7-41 days (20 °C) (EFSA, 2008). MCPA is a pesticide rarely applied to winter cereal. However, in this study MCPA represents mobile pesticides.

3.5 Application of bromide and pesticides to soil columns

The application of bromide solution (7.85 g/L) and MCPA pesticide solution (282 mg/L) was done only once before freezing and storing of the soil columns (table 3). The bromide and pesticide solutions were placed in room temperature 30 minutes before application. Five mL of bromide solution was applied using a 5 mL Finnpipette in circular motion towards the centre of the soil columns. The same amount and procedure was used when applying the pesticide solution after the bromide solution. The amount of pesticides applied (table 3) was calculated to recommended application rates. MCPA was made from solid phase, where 100 mg of MCPA was diluted with 100 mL acetonitrile, before the amount needed was extracted using a pipette. The amount applied was somewhat high due to the area calculated was based on the outer 10 cm diameter instead of the inner diameter of 9.2 cm. In addition, the actual height of the columns might be closer to 18 cm to allow for ponding. The solutions were applied to all soil columns including the unfrozen soil columns. A certain distance (1 cm) was kept from the columns outer edge to prevent adsorption to the aluminium walls.

Table 3. Application rate, amount per column, amount weighted to make 250 mL solution and concentration.

Substace	Application rate (kg/ha)	Amount per. column (mg)	Amount weighted (mg)	Concentration (mg/L)
KBr	74.47	58.5	2922.7[1]	11691
Br	50	39.3	-	7850
MCPA	1.8	1.4	-	282

[1] Amount weighted and diluted in 250 mL deionized water

3.6 Experimental set-up for leaching experiment

Four freeze – irrigation simulations were conducted after the application of bromide and pesticide. The five insulated soil columns (4 cm polyethylene foam camping maps) were stored frozen (- 3 °C) in the dark. The soil columns were placed on Styrofoam to further ensure freezing from the top-down (see section 3.2). Correspondingly, five soil columns were stored unfrozen (+2-4 °C) in the dark. All soil columns were transported to the lysimeter lab at NIBIO when the logger displayed temperatures below 0 °C in the frozen soil columns (after approximately two weeks of storage). The soil columns were then installed onto funnels with polycarbonate bottles placed underneath to collect the leachate.

Autoclude peristaltic pump was used in the experiment. The pump was started before the soil columns were put in place so that the silicone tubes (VWR, 1.52x3.3 mm) were filled with artificial rainwater before the start of the irrigation. Whatman filter paper (GE Healthcare Life Science, 85 mm) was placed on top of each soil column to ensure an even distribution of the irrigated rainwater. Metal strings were attached to the tubes outlet to keep the irrigation centred. The Autoclude pump was calibrated through trial and errors to ensure that artificial rainwater was pumped at a rate of approximately 5 mm per hour for 5 hours (~ 196 mL). Although there was no significant difference in the irrigation rate measured between two different test-runs, some variations were observed between the tubes (Appendix 8). The irrigation rate was considered as moderate rain (National Meterological Library and Archive, 2011).

The laboratory and rainwater used held a temperature of approximately 7-10 °C and 2-6 °C, respectively during the irrigation events. The temperatures were monitored throughout the day using simple thermometers. The variation in temperatures observed was considered caused by body heat. The temperature chosen was though to represent somewhat realistic weather conditions during thaw periods. The leachate was gathered continuously using 150 mL plastic bottles with polycarbonate caps (Corning, VWR). The bottles were weighted before and after the collection of approximately 25 mL of leachate. This was done to monitor the exact amount of effluent leached through the soil columns. The number of samples per column varied from 1-8. Four mL was extracted into plastic test tubes and stored in the fridge (3 °C) for later bromide analysis. The remaining leachate was transferred to coloured medicine flask

and placed horizontal in the freezer (-20 °C) ready for pesticide analysis. The last samples were gathered the morning after onset of irrigation. After sampling the five frozen soil columns were transported back to the freezer (-3 °C), while the unfrozen soil columns were transported back to fridge (2-4°C). Fig. 9 illustrates the timeline for the experiment and treatment of the columns.
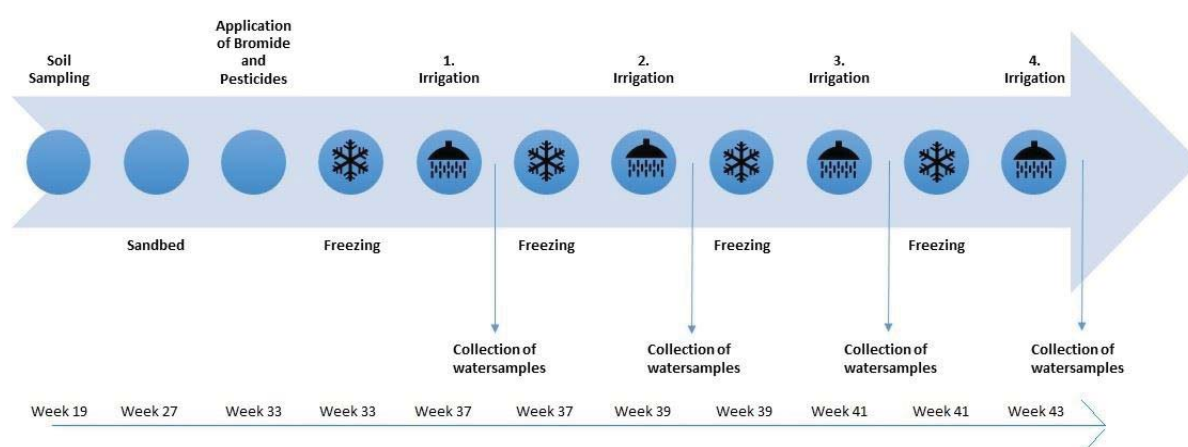

Fig. 9. Timeline and treatment of soil columns over a 24-week freezing and thawing experiment including soil sampling, preparation of the soil columns (sandbed etc.), application of bromide and pesticide and the four freezing and rainfall cycles.

3.7 Analysis of leachate samples from soil columns

3.7.1 Bromide

The Mettler Toledo pH/ion apparatus coupled with an ion selective electrode (Thermo Fisher Scientific, Orion 9635BNWP) was used to measure the bromide concentration. Reproducibility of direct electrode measurements was ±2 %. Standard curves were constructed to calibrate the instrument (Appendix 7). 0.8 ml of ISA was added to each 4 ml bromide samples (50:1 ratio recommended by NIBIO). The samples were shaken with the 'lab-dancer' for 10 seconds before the samples were measured directly. Deionized water was used to rinse the electrode between each measurement. Results are reported as the

accumulated amount of bromide leached as percentage of the applied amount of bromide 39.25 mg from frozen and unfrozen soil columns. Breakthrough curves (BTCs) are presented as relative concentration (C/Co) with respect to accumulated volume leached (mL) after the total of four irrigation. Relative concentrations were calculated as measured concentration of bromide to initial concentration of bromide 7850 mg/l.

3.7.2 Pesticide

Pesticide analysis of MCPA and the main degradation product 2-MCP were performed using LC-MS/MS by Marit Almvik at the section of Pesticides and Natural Products Chemistry at NIBIO. The limit of quantification for MCPA and 2-MCP was 0.125 µg/L and 5 µg/L, respectively. The pesticide samples were thawed in a dark room to prevent photo degradation. Medicine flask with cracks were placed in beakers. 1000 µl of the samples were filtered using a 1-ml syringe and 100 µl from the filtered samples were transferred to glass vials using pipette before deionized water (MiliQ) and an internal standard (2.4-D) were added. Some samples had to be analysed undiluted due to low concentrations. 475 µl of undiluted sample were then added to 25 µl of internal standard (2.4-D). 200 µl were extracted and transferred to glass inserts with vials (dilutant factor 1.05). Crimper tool was used to attach the crimp caps before the vials were stored in the freezer (-20 °C) ready for MCPA analysis on the LC-MS/MS (See appendix 9 for procedure). Results are reported as accumulated amount of MCPA leached as percentage of the applied amount MCPA 1.4 mg from frozen and unfrozen soil columns. Breakthrough curves (BTCs) are presented as relative concentration (C/Co) with respect to accumulated volume leached (mL) after the total of four irrigation. Relative concentrations were calculated as measured concentration of MCPA to initial concentration of MCPA 282.60 mg/l

3.8 Evaluation of exposure and potential effects of MCPA at the field scale

Environmental monitoring during winter and early spring has not been conducted regularly in the JOVA monitoring program (www.nibio.no/jova). However, existing monitoring data of pesticide use and measured environmental concentrations during the growing season were obtained from the JOVA database and used to provide a base line for the evaluation of exposure and potential effects at the field scale during winter and early spring. Data from the small agricultural catchment Skuterud in Ås, Norway, were chosen based on the predominance of cereal production and the soil type Albeluvisol, which dominates 75 % of the agricultural land (Deelstra et al., 2005). There has been a continuous development with

regards to the substances included in the pesticide analysis (Stenrød, 2015) and information on the currently applied multi-methods (M101 and M15) is available online (www.nibio.no/lab). The maximum MCPA concentration measured during the growing season each year were used when evaluating the potential risk towards the aquatic environment. Available aquatic toxicity data (EC_{50}, LC_{50}, NOEC) and formula [3] was used in the calculations of toxic units (TU) for each trophic level (See section 2.4.1 for details). The database used for toxicity tests in JOVA are gathered from The Pesticide Properties DataBase (PPDB) (University of Hertfordshire, 2017), the SANCO database and the EU/EFSA Review reports for active substances in the EU Pesticides database (2013). The MF-value for MCPA was last updated in 2014 and has a calculated MF-value of 1.4 µg/L based on the lowest 14 days NOEC on Lemna gibba (www.bioforsk.no/miljofarlighetsverdier). The calculated results are reported as colour grading according to table 4 of the VKMs grading of risk towards aquatic organisms.

Table 4. Risk evaluation with colour grading of potential effects towards organisms in the environment.

Extremely high risk	> 500 % of the limit values
Significant risk	300 – 500 % of the limit values
High risk	150 – 300 % of the limit value
Moderate risk	100 – 150 % of the limit value
Minimal risk	No exceedance above the limit value

Adaption from (VKM, 2012)

3.9 Statistical method for analysing leaching data

A "t-test: Two sample Assuming unequal variances" was used to determine the p-value for comparisons between the accumulated amount of bromide leached from frozen soil columns in comparison to unfrozen soil columns. A statistically significant difference with 95 % certainty was accepted if $p < 0.05$. The p-value is given in brackets where statistical analysis has been performed.

4. Results

4.1 Leaching Experiment

4.1.1 Soil column temperatures during the leaching experiment

Due to a delay in the start of the 1st irrigation, soil temperatures were at zero. Insufficient insulation caused even higher soil temperatures in one soil column (Fig. 10 A). Soil column temperatures during the 2nd to 4th irrigation was below 0 °C at the start of irrigation (Fig. 10 B to D). For unknown reasons, the logger stopped during the 3rd irrigation, hence, only the first 500 min after start of irrigation is showed (Fig. 10 C). Accumulated water on top of the frozen soil columns (ponding) were observed during the 2nd to 4th irrigation.
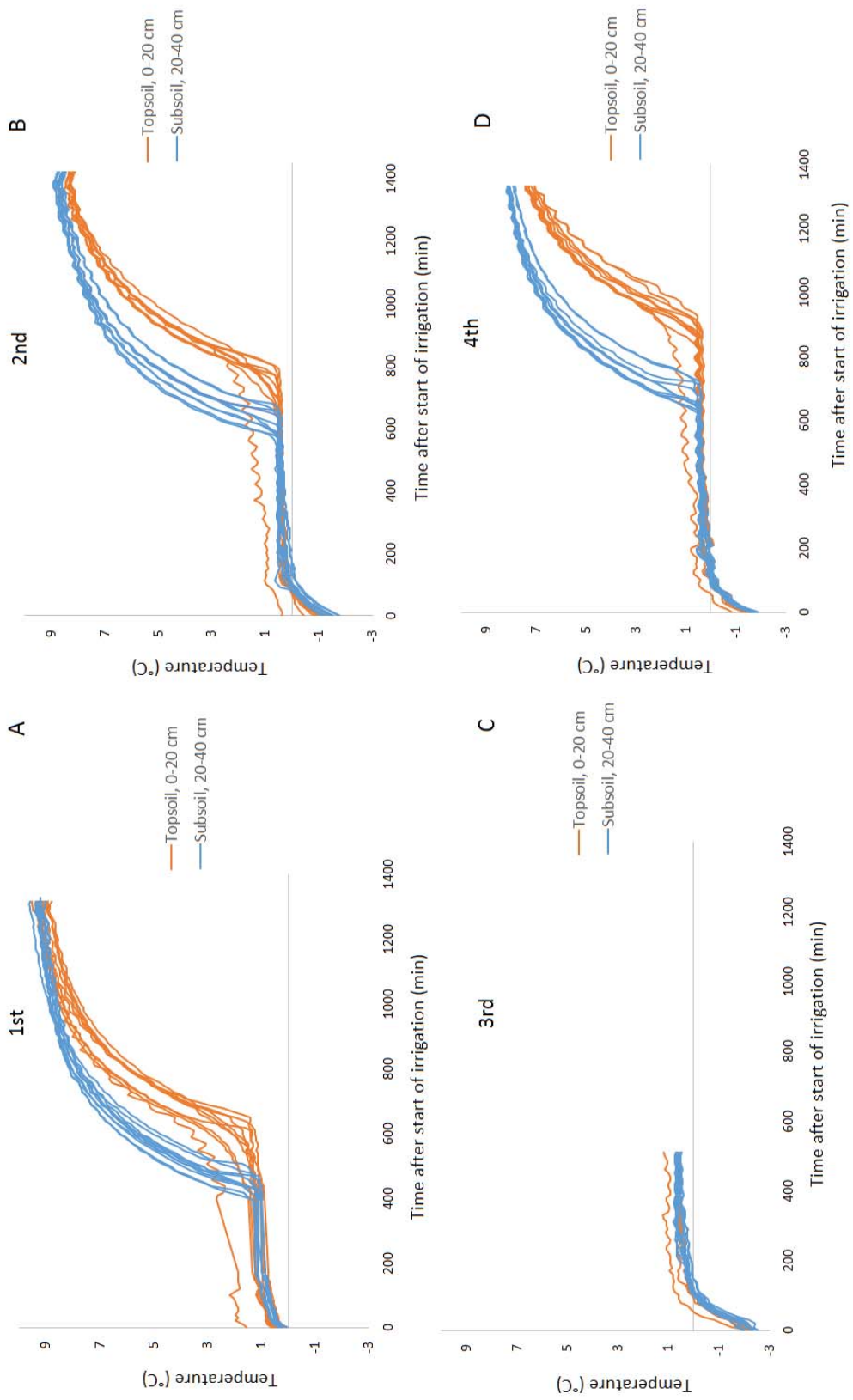
Fig. 10. Soil column temperatures measured at 7 cm and 14 cm from the top of the soil columns in the topsoil (0–20 cm) (brown) during the 1st (A), 2nd (B), 3rd (C) and 4th (D) irrigation. Subsoil (20-40 cm) blue are not included in this thesis.

34

4.1.2 Accumulated amount of bromide leached from soil columns

Total accumulated amount of bromide and water leached from frozen soil columns (2, 5, 6, 12, 27) and unfrozen soil columns (4, 8, 9, 15, 26) were on average 15 ± 1 mg and 23 ± 4 mg, respectively (table 5). The recovery of bromide was higher from unfrozen soil than frozen soil after all four irrigations. Less leachate accumulated from frozen soil columns in comparison to unfrozen columns.

Table 5. Total volume of water leached (ml), amount of bromide leached (mg) and recovery of bromide (%) from frozen (2, 5, 6, 12, 27) and unfrozen soil columns (4, 8, 9, 15, 26) after four irrigations.

Treatment	Soil column no.	Leached water (mL)	Bromide leached	
			Amount (mg)	% of applied
Frozen (-3 °C)	2	462	16.9	43.1
	5	378	14.4	36.7
	6	410	14.2	36.2
	12	363	14.0	35.8
	27	445	13.8	35.0
Unfrozen (+2-4 °C)	4	605	21.1	53.8
	8	622	19.5	49.8
	9	607	24.9	63.3
	15	628	28.4	72.3
	26	568	18.9	48.1

There was a statistically significant difference between bromide leached from frozen soil columns compared to unfrozen soil columns ($p < 0.05$). This can also be observed when comparing Fig. 11 A and B, where accumulated volume leached is plotted versus total accumulated amount of bromide leached of the applied amount. Bromide leached from frozen soil columns varied from 35 to 43 % of the applied amount, while bromide leached from unfrozen soil columns varied from 48 to 72 % of the applied amount.
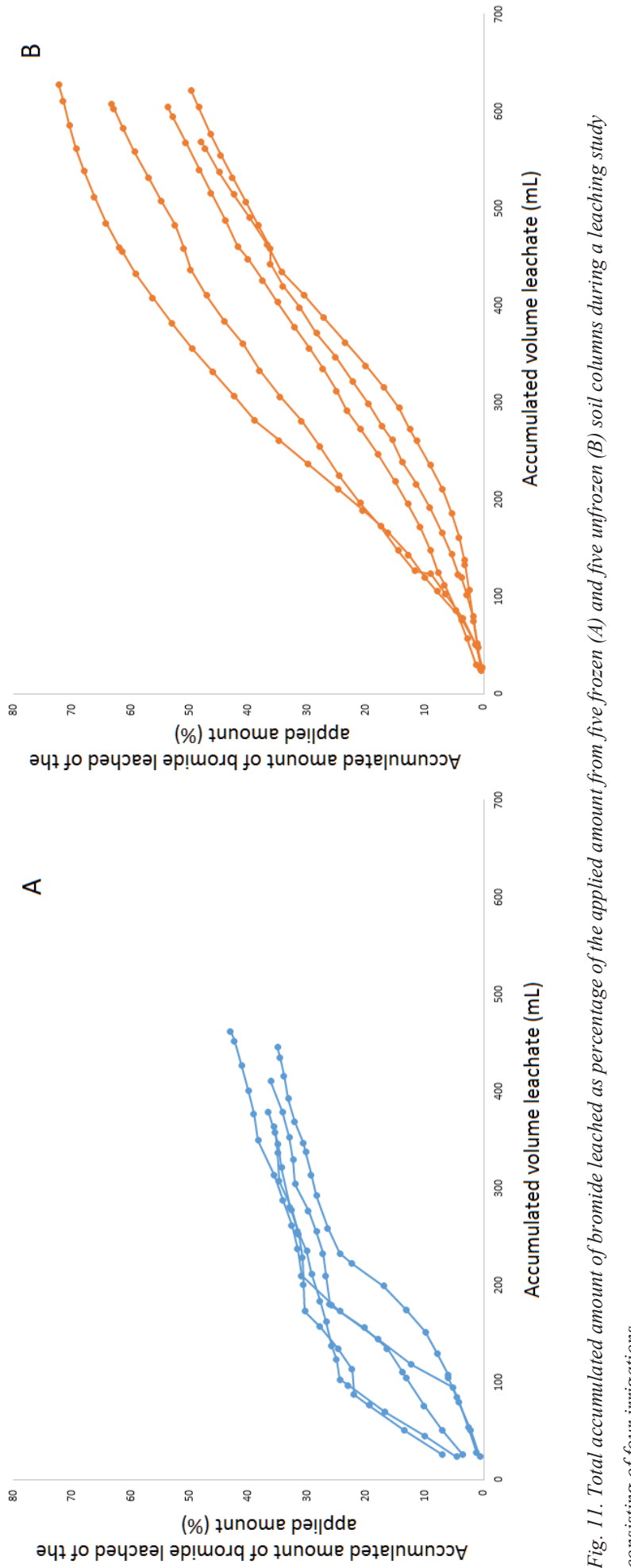
Fig. 11. Total accumulated amount of bromide leached as percentage of the applied amount from five frozen (A) and five unfrozen (B) soil columns during a leaching study consisting of four irrigations.

Results from the study of the accumulated amount of bromide leached from frozen and unfrozen soil columns with respect to time during the 1st, 2nd, 3rd and 4th irrigation is displayed in Fig. 12 A, B, C and D, respectively. There was no statistically significant difference between the amount of bromide leached from frozen soil in comparison to unfrozen soil during the 1st irrigation ($p > 0.05$). However, in 3 out of 5 columns, more bromide leached from frozen soil columns than unfrozen columns (Fig. 12 A). The maximum amount of bromide leached from frozen soil columns was approximately 25 % of the applied amount, while the maximum amount of bromide leached from unfrozen columns were approximately 10 % of the applied amount.

The leaching of bromide from the 2nd irrigation showed less clear differences between the frozen and unfrozen soil columns (Fig. 12 B). The amount of bromide leached from frozen soil columns was approximately at the same level as the amount of bromide leached in the 1st irrigation, while the amount of bromide leached from unfrozen soil columns was higher. Only two samples were collected from the frozen soil column with the lowest accumulated amount of bromide leached. Small amounts of water were observed transported via temperature wires outside the collection vessel. However, the amount is considered negligible. There was no statistically significant difference in bromide leakage from frozen soil columns in comparison to unfrozen columns ($p > 0.05$) during the 2nd irrigation. Mitigations measures were taken during the 2nd irrigation and before the start of the 3rd irrigation due to the accumulation of irrigation water that was observed on top of the frozen soil columns (ponding). Cylinder walls were extended and additional isolation measures were performed.

The amount of bromide leached from the unfrozen soil columns during the 3rd (Fig. 12 C) ($p < 0.05$) and 4th (Fig. 12 D) ($p < 0.05$) irrigation was significant higher than from the frozen soil columns. Thus, results demonstrated that there was more bromide leaching from the frozen than the unfrozen soil columns during the 1st irrigation, while the leaching from the unfrozen soil columns was higher during later irrigation events, indicating that the transport was faster in the frozen soil columns. Due to ponding, samples from frozen soil columns were collected later than samples from unfrozen soil columns during the 2nd to 4th irrigation.

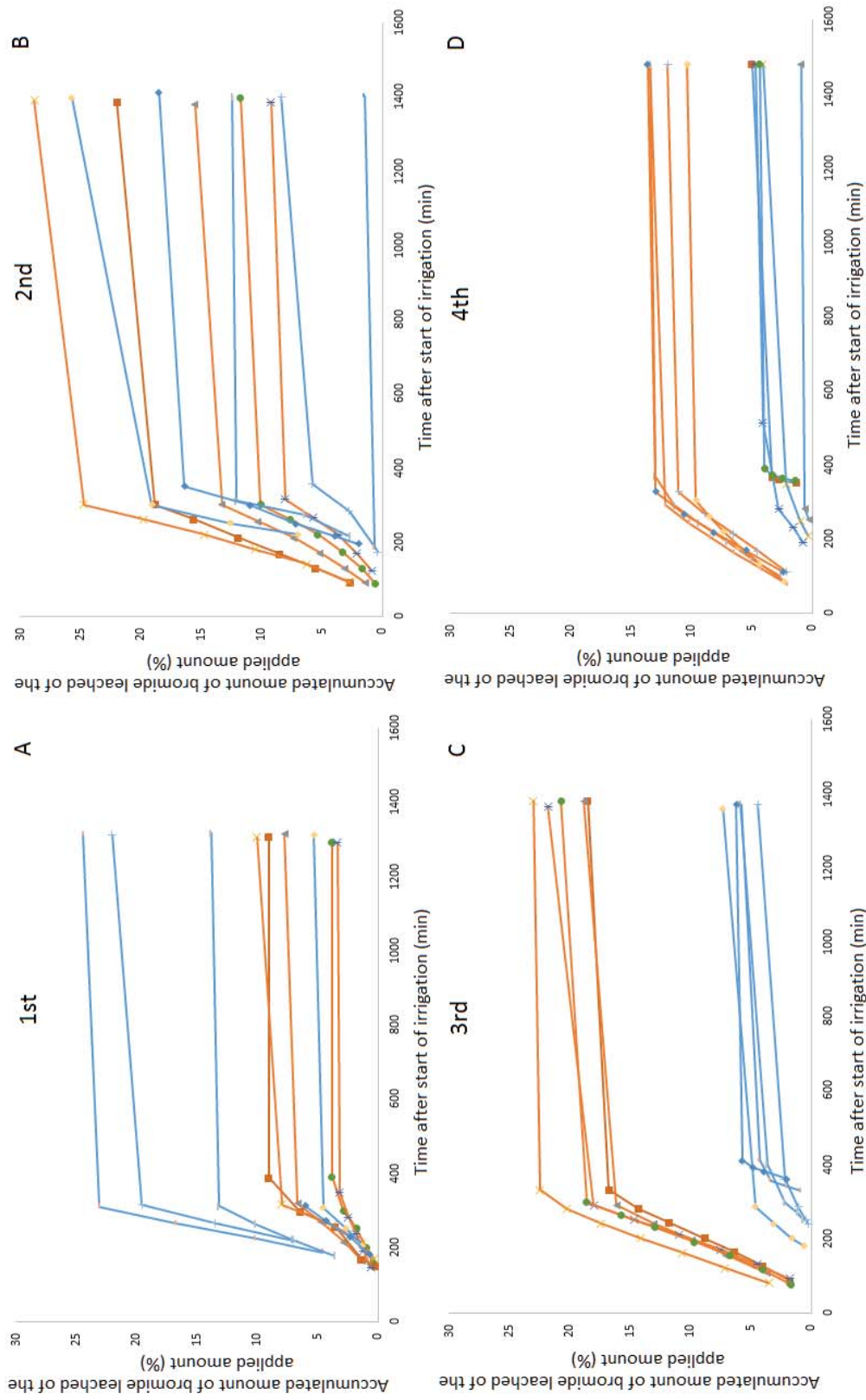*Fig. 12. Accumulated amount of bromide leached as percentage of the applied amount from each of the five frozen (blue) and five unfrozen (brown) soil columns during the 1st (A), 2nd (B), 3rd (C) and 4th (D) irrigation.*

38

4.1.3 Accumulated amount of MCPA leached from columns

Total accumulated amount of MCPA leached from frozen (2, 5, 6, 12, 27) and unfrozen (4, 8, 9, 15, 26) soil columns were on average 346±97 µg and 5±4 µg, respectively (table 6). Higher amounts of MCPA was recovered from frozen than unfrozen soil columns despite that more water leached from unfrozen soil columns.

Table 6. Total volume of water leached (ml), amount (µg) and recovery of MCPA (%) from frozen and unfrozen columns after the total of four irrigations.

Treatment	Soil column no.	Leached water (mL)	Leached MCPA	
			Amount (µg)	(%) of applied
Frozen (-3 °C)	2	462	424.5	30.0
	5	378	283.0	20.0
	6	410	327.8	23.2
	12	363	231.3	16.4
	27	445	464.4	32.9
Unfrozen (-3 °C)	4	605	11.1	0.8
	8	622	5.6	0.4
	9	607	2.6	0.2
	15	628	1.0	0.1
	26	568	5.5	0.4

Fig. 13, presented the volume dependent changes in leaches of MCPA frozen (A) and unfrozen (B) soil columns. Total amount of MCPA leached from frozen soil columns varied between 16 and 33 % of the applied amount after four irrigations. The five frozen soil column replicates in Fig. 13 A appears to continue to rise. Total amount of MCPA leached from unfrozen soil columns was less than 1 % of the applied amount after four irrigations. The maximum amount recovered was 0.8 %. The five unfrozen soil column replicates rise quickly before a flattening of the curves can be seen (Fig. 13 B). There was a statistically significant difference between the amount of MCPA leached from the frozen soil columns in comparison to the unfrozen soil columns after four irrigations ($p < 0.05$).
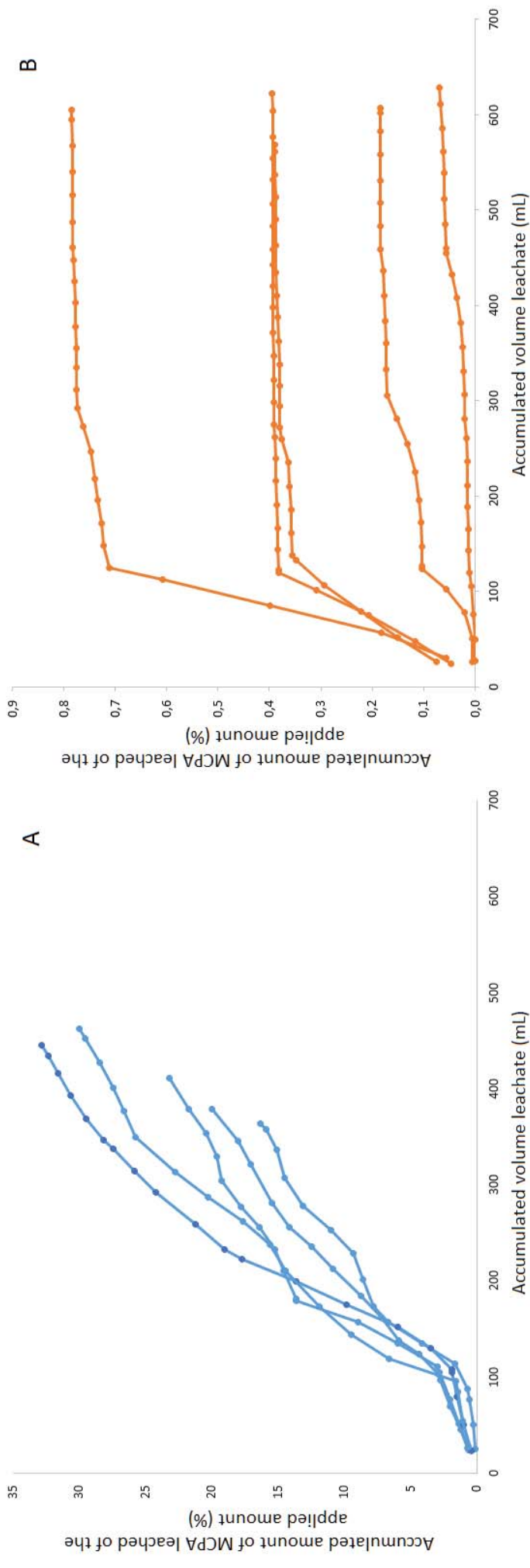
Fig. 13. Total accumulated amount of MCPA leached as percentage of the applied amount from five frozen (A) and five unfrozen (B) soil columns during a leaching study consisting of four irrigations. There is a different scale on the y – axis due to the higher amounts of MCPA leached from frozen soil columns.

40

Fig. 14 present the time dependent changes in accumulated amount of MCPA after the start of irrigation during each of the four irrigations. There was a small amount of MCPA leached overall in the 1st irrigation (maximum ~ 2.9 %). Higher amounts of MCPA was detected in the leachate from frozen soil columns in comparison to unfrozen soil columns (Fig. 14 A). There was a statistically significant difference between the amount of MCPA leached from frozen soil columns in comparison to the unfrozen soil columns during the 1st irrigation at ($p < 0.05$).

By comparing the 1st and 2nd irrigation for the frozen soil columns, increased amounts of MCPA leached during the 2nd irrigation. MCPA leached from frozen soil columns varied between 3 and 17 % (Fig. 14 B). However, less MCPA leached from unfrozen soil columns in the 2nd irrigation compared to the 1st irrigation. There was a statistically significant higher MCPA leached from frozen in comparison to unfrozen soil columns ($p < 0.05$).

 MCPA leached from frozen soil columns in the 3rd irrigation varied between 5 and 12 % of the applied amount (Fig. 14 C). The amount of MCPA detected in the leachate from unfrozen soil columns was similar to MCPA leached from the 2nd irrigation (~ 0 %). There was a statistically significant higher MCPA leached from frozen soil columns in comparison to unfrozen soil columns ($p < 0.05$).

The amount of MCPA leached from frozen soil columns in the 4th irrigation varied between 1 and 6 % (Fig. 14 D). There was a rapid increase in the amount of MCPA leached from frozen soil columns, especially from two soil columns, indicated by the steep slopes. This could also be observed in the 2nd and 3rd irrigation (Fig. 14 B and C). Amount of MCPA leached from unfrozen soil columns in the 4th irrigation was comparable to the 3rd irrigation. There was a statistically significant higher amount of MCPA detected in the leachate from frozen soil columns in comparison to unfrozen soil columns ($p < 0.05$). In contrast, the maximum amount of MCPA leached from unfrozen soil columns was however detected in the 1st irrigation (maximum 0.7 %).
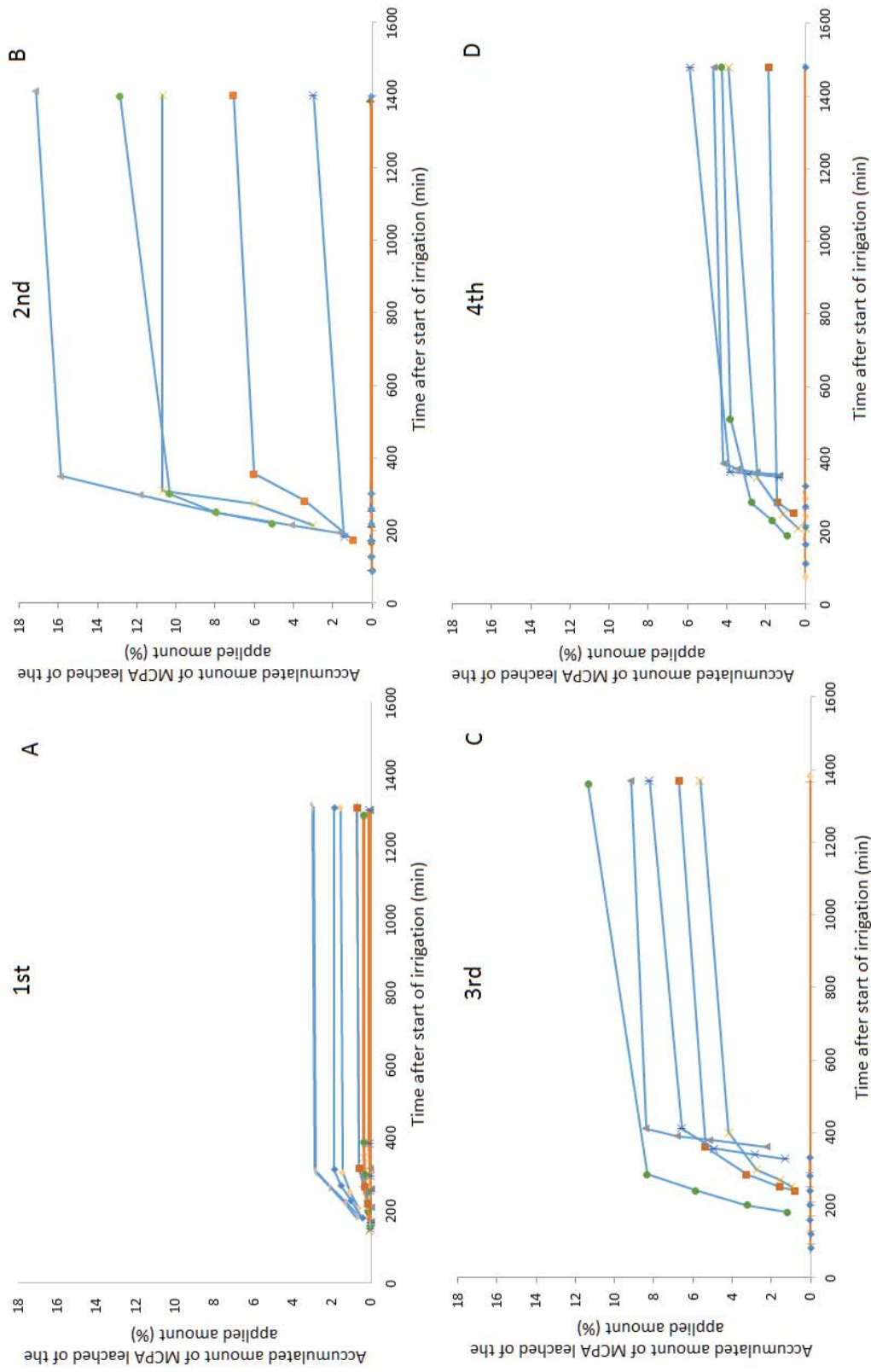
41

Fig 14. Accumulated amount of MCPA leached as percentage of the applied amount from each of the five frozen (blue) and five unfrozen (brown) soil column during the 1ˢᵗ (A), 2ⁿᵈ (B), 3ʳᵈ (C) and 4ᵗʰ (D) irrigation.

42

4.1.4 Breakthrough curves for bromide and MCPA

BTCs from two frozen soil columns are used in presenting the results from frozen soil columns. Two different trends are represented by Fig. 15 (column no. 5 and 12) and Fig. 16 A (column no. 2, 6 and 27). The peak relative bromide concentration for two soil columns was reached in the 1st irrigation, while the peak relative MCPA concentration was reached in the 2nd irrigation (Fig. 15).

Fig. 15. Breakthrough curve (BTC) for bromide and MCPA from one frozen soil column, representative for two out of five frozen soil columns from a leaching study consisting of four irrigations. Black point marks the start of each irrigation.

The peak relative MCPA concentration and peak relative bromide concentration was reached in the 2nd irrigation for three soil columns (Fig. 16 A). MCPA followed the tracer well after the 1st irrigation. This pattern was typical for all breakthrough curves for frozen soil columns. Results from the study of the leaching experiment representative for unfrozen soil columns presented as breakthrough curves from all irrigations are displayed in Fig. 16 B, which represents the typical breakthrough curve for unfrozen soil columns in the experiment. Peak relative bromide concentration was reached in the 3rd irrigation for four out of five soil columns (Appendix 3; column no. 4, 8, 9 and 26). Peak relative MCPA concentration was reached during the 1st irrigation for four out of five unfrozen soil columns (Appendix 3; column no. 4, 8, 9 and 26). Peak relative MCPA concentration for the remaining unfrozen soil column was reached in the 3rd irrigation (Appendix 3; column no. 15).
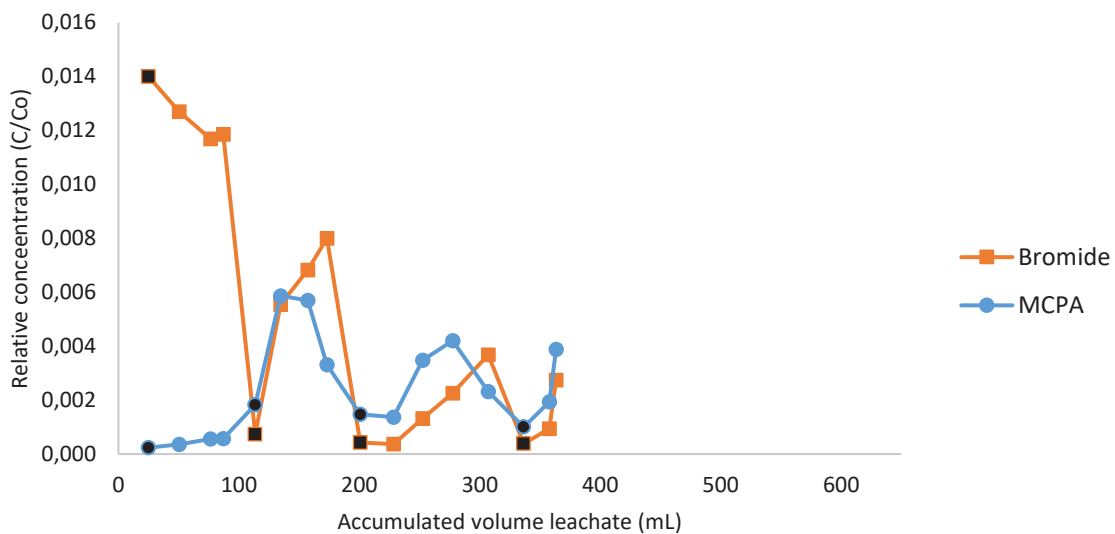
43
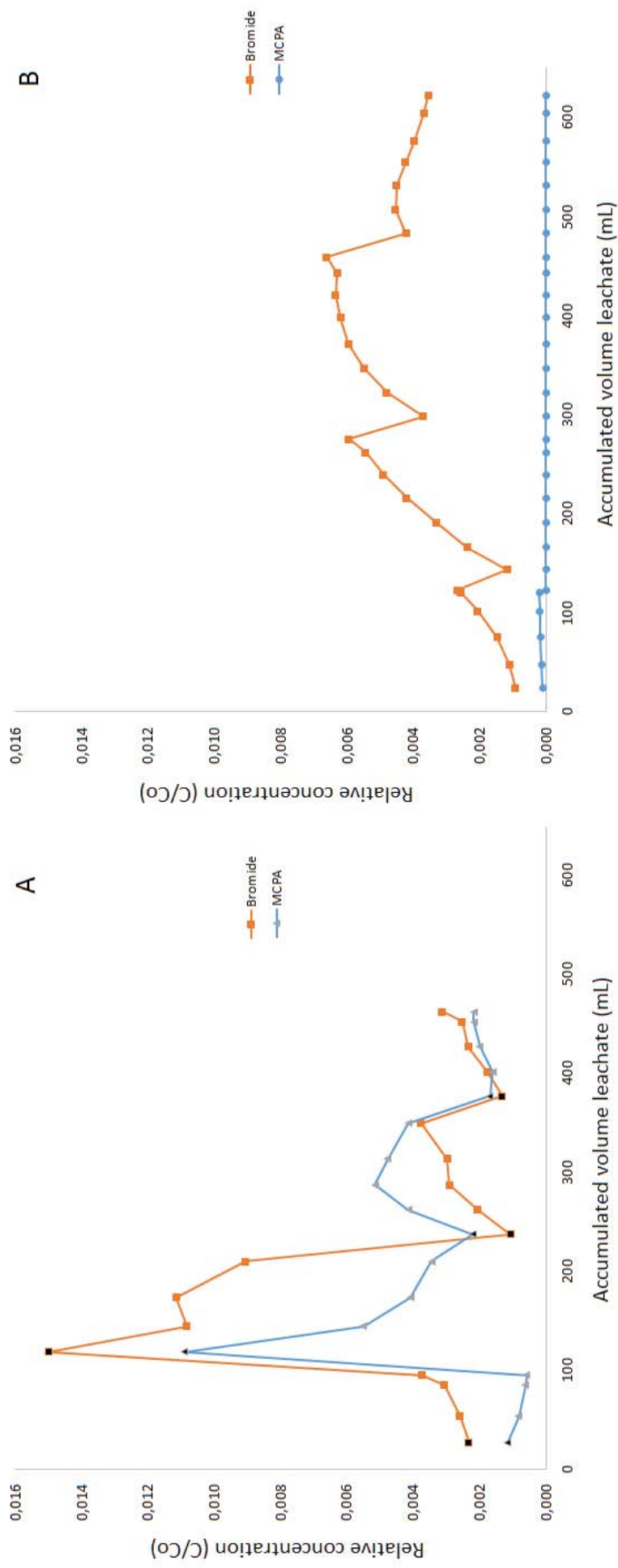
Fig. 16. Breakthrough curves (BTCs) for bromide and MCPA from; a frozen soil column (no. 2), representative for three out of five frozen soil columns (A) and an unfrozen soil column (no. 8), representative for unfrozen soil columns (B) during a leaching study consisting of four irrigations. Black point marks the start of each irrigation.

44

Leaches of MCPA from unfrozen soil columns are represented by an unfrozen soil column separately in Fig. 17 (due to the small amount leached). The peak relative MCPA concentration was reached in the 1st irrigation, except of one column (Appendix 3; column no. 15). Peak relative MCPA concentration of the soil column that behaved differently was reached in the 3rd irrigation, while peak relative bromide concentration was reached in the 2nd irrigation (Appendix 3; column no. 15).
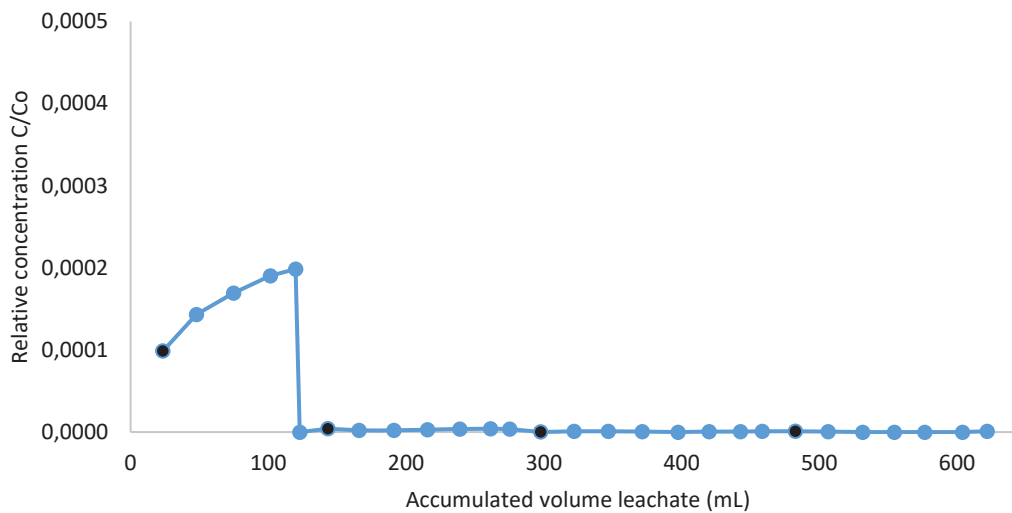
Fig. 17. Breakthrough curve for MCPA from an unfrozen soil column (no. 8), representative for four unfrozen soil columns from a leaching study consisting of four irrigations. Black point marks the start of each irrigation.

The maximum MCPA concentration measured in frozen and unfrozen soil columns during the leaching experiment are given in table 10. The measured concentration decreased in the following order.

- Frozen soil: 2nd irrigation > 3rd irrigation > 4th irrigation > 1st irrigation.
- Unfrozen soil: 1st irrigation > 2nd irrigation > 3rd irrigation > 4th irrigation.

Table 10. Maximum concentration (µg/l) measured in leachate of MCPA for each irrigation in frozen and unfrozen soil columns from Kroer topsoil (0-20 cm).

Treatment	Column no.	Maximum concentration (µg/L) MCPA leached during irrigations			
		1st	2nd	3rd	4th
Frozen (-3°C)	2	324.9	3081.7	1459.2	616.9
	5	464.5	1579.5	1116.6	856.1
	6	402.2	2897.3	935.4	714.8
	12	161.4	1658.6	1187.6	1096.4
	27	299.2	2530.5	1254.6	837.8
Unfrozen (+2-4°C)	4	111.3	8.4	1.2	0.6
	8	56.1	1.2	0.3	0.3
	9	30.9	11.2	2.5	0.3
	15	2.7	2.3	7.7	2.3
	26	41.0	7.4	1.6	0.2

4.2 Evaluation of potential effects of environmental concentrations of MCPA on aquatic organisms

Use of pesticides has been monitored throughout the growing season in the cereal dominated Skuterud catchment since 1995 (Fig. 18). The use of MCPA varies between years due to differences in weather conditions and agricultural practice such as tillage (Bechmann, 2014). Number of water samples analysed each year varied from 7 to 21. The JOVA monitoring data show in 2013, more than 120 kg of MCPA was applied to 75 % of the agricultural area. Only 11 % of the area lay in stubble through the winter this year. In addition, high amount of precipitation and little snow cover characterized the season (Hauken et al., 2015). Moreover, MCPA was in this year detected in six out of ten environmental water samples. The average concentration of MCPA was higher in 2010 in comparison to the following years due to a high detection of MCPA in a water sample from May at 3.3 µg/l (table 11), exceeding the concentration level which indicates potential negative effects to aquatic organisms (MF-value) at 1.4 µg/L. Detections of MCPA in percentage of analysed water samples are given in Appendix 10 (same trend).
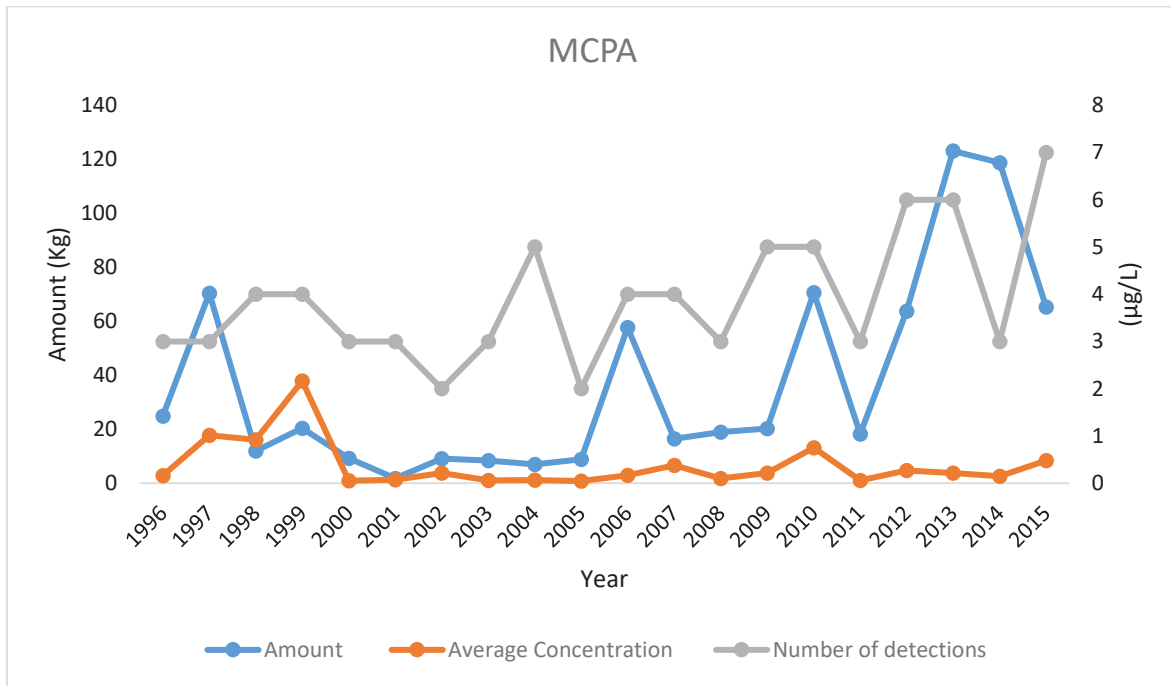
Fig. 18. Total applied amount per year (Kg), average concentration (µg/l) and number of detections of MCPA during the growing season at the Skuterud catchment.

By comparing the maximum measured environmental concentration (MEC) for each year in the period between 1995 to 2015 with the MF-value, results showed an exceedance in 1999, 2010 and 2015 (table 11). Assessing the different aquatic organism used for toxicity tests: aquatic plants, invertebrate and fish, all the exceedances indicated a potential risk towards aquatic plants, with extremely high risk in 1999, high risk in 2010 and a moderate risk in 2015.

Table 11. Worst-case samples (µg/L) of MCPA detected in environmental water samples in agricultural streams in the Skuterud catchment and resulting colour grading exceedance above MF-value and calculated Toxic Units (TUs) at three trophic levels represented by the aquatic organisms; aquatic plants, invertebrates and fish at the Skuterud Catchment throughout the growing season in the period 1995 to 2015.

	MCPA			
Year	**Maximum measured environmental concentration (µg/L)**	**Toxic unit (TU)**		
		TU$_{aquatic\ plant}$	**TU$_{invertebrate}$**	**TU$_{fish}$**
1995	0.51	0.04	0.00	0.00
1996	0.15	0.01	0.00	0.00
1997	0.11	0.01	0.00	0.00
1998	0.88	0.06	0.00	0.00
1999	7.60	0.54	0.00	0.00
2000	0.06	0.00	0.00	0.00
2001	0.08	0.01	0.00	0.00
2002	0.31	0.02	0.00	0.00
2003	0.09	0.01	0.00	0.00
2004	0.11	0.01	0.00	0.00
2005	0.05	0.00	0.00	0.00
2006	0.25	0.02	0.00	0.00
2007	0.68	0.05	0.00	0.00
2008	0.17	0.01	0.00	0.00
2009	0.67	0.05	0.00	0.00
2010	3.30	0.24	0.00	0.00
2011	0.09	0.01	0.00	0.00
2012	0.89	0.06	0.00	0.00
2013	1.06	0.08	0.00	0.00
2014	0.25	0.02	0.00	0.00
2015	1.60	0.11	0.00	0.00

5. Discussion

5.1 Soil freezing and bromide leaching: implications for water transport through partially frozen soil

The conservative tracer bromide is assumed to be very close connected to the flow of water and degradation and sorption can be considered close to negligible (Levy & Chambers, 1987). However, the non-linear relationship between the amount of bromide leached and accumulated volume of leachate for unfrozen soil columns (Fig. 11 B), indicates a small delay in the amount of bromide leached in comparison to the water flow. In contrast, bromide was transported more rapidly through the frozen soil columns, indicated by the more linear relationship between the amount of bromide leached in comparison to the accumulated leachate, especially in the beginning prior to the accumulation of 200 mL leachate (Fig. 11 A). The increased amount of bromide leached from frozen soil columns compared to unfrozen soil columns during the 1st irrigation (Fig. 12 A, for three out of five frozen soil columns) could be comparable to the study by van der Kamp et al. (2003) who reports that snowmelt infiltrates through air-filled macropores. The equilibration of the soil columns (see section 3.2) resulted in withdrawal of water and subsequent opening of the largest pores. Only capillary forces prevented water from the smaller pores to drain. Subsequently, air-filled macropores that were present and conserved could contribute to a rapid transport of water and bromide through the partially frozen soil upon the 1st irrigation.

Transport of water and solutes through non-uniform soils with preferential flow are usually characterized by an early arrival of solutes and a late tail time (Levy & Berkowitz, 2003; Larsbo et al., 2014). Early arrival relative peak concentration characterizes the BTCs for frozen soil columns (Fig. 15 and 16 A) in comparison to unfrozen soil columns (Fig. 16 B), indicating a faster transport of bromide through frozen soil columns. As mentioned, macropores present may have functioned as preferential flow pathways and transported the tracer bromide predominately through the soil at a high rate. This phenomenon is also described by Stadler et al. (2000). Water present in smaller pores prior to the 1st irrigation, froze when the temperature was kept below zero for an extended period. Thus, possibly may have contributed to a more dominating role of the air-filled macropores for vertical solute transport.

During the 3rd and 4th irrigation, the amount of bromide leached from the unfrozen soil columns exceeded the amount from frozen soil columns (Fig. 12 C and D). The breakthrough curves of the unfrozen soil columns (see example in Fig. 16 B) are smoother than for the frozen soil, indicating that the water moved more slowly through the unfrozen soil columns. This vertical transport of water and solutes could be explained by the matrix flow dominated by convection, dispersion and diffusion. The matrix flow may have during the 3rd and 4th irrigation pushed the bromide and water front through the soil's profile which resulted in an increased volume of accumulated leachate and thus an increased amount of bromide (table 5). More water leached from the soil should result in more bromide being washed-out. Estimated pore volume for the Kroer soil is ~ 600 mL (see Appendix 2 for calculations), thus indicating the estimated pore volume for unfrozen soil columns could have been reached after four irrigations (Fig. 11 B). Additionally, the decreasing amount of bromide leached from frozen soil columns in the 3rd and 4th irrigation in comparison to the 1st irrigation, could be explained by the fixed amount of bromide applied had been washed out during the 1st irrigation. In general, the soil columns did not reach the asymptote (Fig. 11), indicating that additional irrigations may have resulted in more bromide being washed out.

5.2 Soil freezing and MCPA leaching: implications for pesticide transport through partially frozen soil

Pesticides dissipates in the environment through chemical, photochemical or biological degradation and transport processes including sorption (Thorstensen & Lode, 2001a). MCPA degrades rapidly through aqueous photolysis at pH 5, where the main degradation product is 2-MCP. Consequently, photolysis might affect the collection of samples prior to freezing. The half-life of MCPA in soil is between 7 to 41 days at 20 °C (European Comission, 2008). Studies shows that pesticides degrade slowly in soil temperatures below 5 °C (Stenberg et al., 1998; Stenrød et al., 2008). Hence, degradation of MCPA while storing leachate samples and soil columns prior and during the experiment is considered negliglible, but can not be excluded. MCPA is stable to hydrolytic degradation. Further, MCPA's key metabolite 2-MCP, associates well with the aqueous phase (water solubility 4000 mg/l) (European Commision, 2008). However, 2-MCP was not detected in any of the analysed leachate samples from the unfrozen soil columns.

A comparison of the breakthrough curves for MCPA and bromide from frozen soil columns (see example in Fig. 15 and 16 A), shows that the mobile MCPA follows roughly the same pattern as bromide. However, MCPA does not show the initial rapid transport as bromide and does not appear to be preferentially transported through the assumed initially open macropores for two soil columns in the 1st irrigation (Fig. 15). This could be explained by the higher sorption affinities to soil for MCPA than bromide, possibly making the pesticide unavailable for initial macropore flow. Hillel et al. (2004) argues that diffusion processes in the soil plays an important role for transport of solutes, especially at low water flow rates. Hence, different diffusion rates between bromide and MCPA might explain the early arrival of bromide, especially during the 1st irrigation (Fig. 15), when the water flow was governed by the irrigation rate (no accumulated water on top of the soil). Increased diffusion of the lighter bromide molecule in comparison to the heavier MCPA molecule downward with the pore water could potentially enhance the transport of bromide.

Two frozen soil columns behaved differently in the initial transport during the 1st irrigation, where neither MCPA or bromide were transmitted at high amounts (Fig. 16 A). The temperature at the start of the 1st irrigation was at 0 °C (Fig. 10 A), which indicated that the soil had started to thaw. Consequently, the low degree of ice-blockage in the soil matrix due to the presumed low initial soil water content may had enabled MCPA to enter the soil matrix and become immobilized through sorption. This is supported by the study reported by Shipitalo et al. (1989) who shows that atrazine and bromide is transported into the soil matrix, limiting macropore flow at first rainfall after application. However, Shipitalo et al. (1989) invastigates unfrozen soil that may not be fully comparable to our results. Thawing of porewater after freezing happens at different temperatures depending on the pore size (Ireson et al., 2013), thus both ice and water might have been present during the 1st irrigation. The observed phenomenon by Shipitalo et al. (1989) could also be a contributing factor for the generally low amount of MCPA leached from all frozen soil columns during the 1st irrigation compared to later irrigations (Fig. 14 A and e.g. B). Furthermore, the chemical active microsites inside the macropores may have contributed to increased sorption of MCPA due to the applied irrigation rate was too low for effectively bypassing the matrix.

Preferential flow pathways must be activated through wetting of the adjacent matrix to transport water and solutes rapidly through the soil (Beven & Germann, 1980). Further, initial and boundary conditions, such as initial water content, amount of irrigation and intensity realize macropore flow (Jarvis, 2007). Assuming this applies to frozen soil upon thawing, suggests a need for hydrological activation of the macropore network. The less prominent role of macropore flow during the 1st irrigation (Fig. 16 A), indicated that the soil was either too dry and or the irrigation rate too low for efficient preferential flow of bromide and MCPA. This is supported by Klaus et al. (2014), who identified a threshold for triggering preferential flow of herbicides through the macroporous unfrozen soil dependent on irrigation and soil water content. Furthermore, the expected spatial variability of the soil, and thus, the non-uniform distribution of macropores, may suggests that a macropore network could be present further down the soil profile, not open to the soil surface, thus not available for preferential flow until reaching the macropore network. As previously discussed, initial transport of bromide through the 1st irrigation (Fig. 15), however, implies that macropores were activated and indicated preferential flow for two of the five frozen soil columns.

The degree of ice-filled pores controls the infiltration capacity of the soil in cold climate conditions (Ireson et al., 2013). Subsequently, the hydraulic conductivity decreases as the pore water freeze. This could be observed as the first leachate samples were collected at an earlier time-point from unfrozen soil columns than frozen soil columns. Significant ponding events is a function of soil moisture prior to freezing and frost depth (Kitterød, 2008). The 2nd to 4th irrigation was characterized by ponding events and a delayed release of frost, indicated by the late retrieval of leachate samples, indicating prolonged ice blockage through higher soil water content. This suggest that a frozen soil layer of <20 cm impeded infiltration of the irrigation water. This is somewhat in accordance with the study by Iwata et al. (2011) who shows that snowmelt infiltrates through a thin (<14 cm) frozen soil layer, while a thicker (<37 cm) frozen soil layer impedes the infiltration. Consequently, increased volume of water accumulated on top of the temporarily-impermeable frozen soil, was available for transport upon thawing. On soil thawing, the flux of water was high, especially during the 2rd to 4th irrigation, which was observed during the leaching experiment and illustrated by the steep slopes in the curves of the frozen soil columns in e.g. Fig. 14 B and C, causing MCPA which initially might be weakly adsorbed to soil particles and/or active microsites during the 1st irrigation, remobilized and transported through macropores thus bypassing sorption sites in

the soil. This event could be compared to the snowmelt event described by Kitterød (2008), who argues that the effect of surface ponding in combination with the structure of the soil may result in a rapid flow locally. The remobilization of MCPA during the 2nd irrigation is supported by the field scale experiments on unfrozen soils by McGrath et al. (2010) and Klaus et al. (2014), where pre-event soil water remobilizes the pesticides when entering the macropore flow pathways. This suggests a similar behaviour in the partially frozen soil columns.

MCPA leached from frozen soil columns varied between 16 % and 33 % of the applied amount after four irrigations (table 6). Applying the VKM grading system (VKM, 2012), resulted in a mobility grading to *very high* after all four irrigations (see Appendix 11 for details). Spatial variability at the soil sampling site and of the macropores was reflected by the variances of recovery of MCPA and bromide in the leachate. This could especially be observed by the collection of the leachate samples from the 2nd irrigation (Fig. 14 B). The results showed that the amounts of MCPA leached from frozen soil columns were still increasing after four irrigation (Fig. 13 A), indicating that additional irrigations could have resulted in higher total amounts of pesticides leached.

The small amount of MCPA transported through the unfrozen soil columns, indicated that the silty clay loam at estimated 30 % initial water content (Appendix 4), was susceptible for initial preferential flow through the open macropore network during the 1st irrigation (Fig. 17). However, the total amount of MCPA leached from unfrozen columns was less than 1 % of the applied amount after all four irrigations. Similar proportions leached has been observed in earlier studies for dimethoate ($K_f = 0.36$, University of Hertfordshire, 2017) (Uhlen et al., 1994). However, the study reported by Thorstensen & Lode (2001a) shows that less than 0.1 % MCPA remains in a loam soil from Kroer after 84 days. This may be explained by the degradation of MCPA in soil is ~ 25 days at 25 °C and degradation of MCPA in this study was less important.

There was a negative correlation between bromide and MCPA leached from unfrozen soil columns (Fig. 16 B). The amount of MCPA leached from unfrozen soil columns was higher

during the 1st irrigation in comparison to later irrigations (Fig. 14 A compared to e.g. 14 B). For three out of five soil columns, more than 90 % of the total accumulated amount of MCPA after four irrigations leached through the soil during the 1st irrigation (see Appendix 5), while the maximum accumulated amount of bromide was reached during the 3rd irrigation (Fig. 12 C). Bromide seemed to be mainly transported through uniform flow (matrix flow). The different flow pattern between MCPA and bromide may be explained by macropore flow, the higher affinity to soil particles for MCPA, different diffusion rates between MCPA and bromide and to a lesser extent degradation. Solutes diffuse from high to low concentration gradient and are dominating at low water rates (Hillel, 2004). The characteristic "saw tooth" shape of the BTCs for unfrozen soil columns (Fig. 16 B), indicated that bromide diffusion occurred at the start of each irrigation and gradually diminished as the pressure gradient (e.g. convection) became more dominant. Moreover, macropore flow appeared to be less prominent than in the frozen soil columns, indicated by the smoother shape of the BTCs (Fig. 15 B). This could be due to a diminishing role of the macropores with increasing soil water content (Hillel, 2004). This is supported by Larsbo et al. (2014) showing that the diffusive flux between macropores and the soil matrix increases and subsequently decreases preferential flow at near-saturated conditions. It should be mentioned that the soil water content was not accurately measured, but it can be assumed that the water content increased by the irrigation.

Important factors affecting sorption of MCPA in soil are water solubility, polarity and acid-base constant. The mobility of MCPA in clay soil within normal soil pH (4-5), are in general enhanced by the net repellence of negatively charged clay molecules due to the anionic form of MCPA (Hiller et al., 2006). However, the mobility, solubility and volatility of MCPA is sensitive to pH changes when the acid-basic character (pK_a) is close to the pH value of the soil (Wauchope et al., 2002). Consequently, MCPA (pK_a =3.07) could be present both in the ionized and unionized form (X^-/XH) at low pH, thus decreasing the net repellence and subsequent the mobility. A change in pH unit will change the ionized/unionized-ratio by a tenfold (Wauchope et al., 2002). The pH of the soil was approximately 5.5 (table 1), while the pH of the irrigation water applied was approximately 5. Introducing the artificial rainwater to the silty clay loam may have developed a certain anion-exchange capacity. Further, iron minerals could be present in the soil (see Fig. 7). Thus, positive charged Fe hydroxides in its protonated form present at low pH, could contribute to additional adsorption of MCPA in the soil. Thus, decreased the mobility of MCPA in the soil. Furthermore, unrelated to surface

charge, if iron hydroxide was present (neither protonated or deprotonated), covalent bonds could form between the unionized MCPA and the hydrate iron oxide retaining MCPA in the soil. Soil extraction would determine the residual amount of MCPA and 2-MCP in the soil. Applying the mobility grading system of VKM, resulted in *moderate* mobility for the leaching of MCPA from unfrozen soil after all four irrigations.

To sum up, the small amounts of MCPA leached from unfrozen soil columns was mainly transported through the initial macropore flow while bromide were transported by the matrix flow. The difference seen could be explain by the different diffusion rates of MCPA and bromide and the higher sorption affinities to soil for MCPA. Conversely, MCPA leached from frozen soil columns were higher during the 2nd and 3rd irrigation than the 1st irrigation, indicating a well-developed activated macropore network functional as efficient preferential pathways when high amounts of available ponding water caused higher degree of preferential transport in later irrigations upon thawing. However, the temperature at the start of irrigation may have affected the outcome during the 1st irrigation. Increased amounts of MCPA was recovered from frozen soil columns (16-33 %) in comparison to unfrozen soil columns (<1 %). MCPA in frozen soil seemed to follow the pattern for bromide well, except of the initial part of the leaching study. The leaching pattern for MCPA and bromide in the unfrozen soil does however show large discrepancies. It appears that freezing and thawing alters the flow pattern for MCPA, indicating a larger role of the macropores and thus, an increased amount of MCPA transported through the frozen topsoil.

5.3 Pesticide use and leaching under cold climate conditions: implications for potential effects in the aquatic environment

The nature of the pesticide, soil properties, climate and agricultural management affects the transport, sorption and degradation of pesticides in soils affecting the risk of exposure in the environment. In this thesis, mainly the transport was investigated by the contributing factor of climate (Fig.1). Accordingly, the reported leaching study indicates important effects of soil freezing and thawing during winter on the risk of leaching of the mobile herbicide MCPA in the widespread soil type Albeluvisol. These implications however are restricted to the transport of mobile pesticides that would behave similar to MCPA such as pesticides with similar sorption properties and, hence relatively high mobility in soil. Furthermore, as

mentioned in the introduction, pesticides applied to winter cereal is of relevance when addressing the influence of cold climate conditions to the leaching of pesticides.

Pesticide risk assessments in the environment are based on the toxicity of the pesticide and the pesticide exposure (Fig. 1). As described in section 2.4.2, monitoring data and concentration levels indicating potential environmental effects (MF-values), are used in the evaluation. However, monitoring data during the winter has not been prioritized in JOVA. Hence, there is a lack of field data to assess the potential effects of winter conditions on the leaching of pesticides at the field scale. However, monitoring data during the growing season could provide a basis for comparison to assess the risk of exposure during winter.

The small catchment Skuterud, which represents agricultural areas in eastern Norway, has been monitored for pesticide use in cereal production since 1995. The catchment area is mainly dominated by the same soil type as used in the leaching experiment; Albeluvisol (Appendix 6). Reported use, number of detections and average concentrations of MCPA in stream water in the period 1996-2015 (Fig. 18), shows a correlation between increase in reported use of pesticides and number of detections of MCPA in water samples from 2005 (see Appendix 10 for detections in percentage of analysed water samples). Evaluating the MF-value (1,4µg/l) for the maximum concentration measured in 14-day composite water samples through the growing season of each year, resulted in exceedance of the limit value in 1999, 2010 and 2015 (table 11), posing a risk of effect in aquatic organisms. Furthermore, TUs calculations for three aquatic organisms (table 11), indicated that the risk of exposure was directed towards aquatic plants. The sampling methodology at Skuterud may however, cause an underestimation of the maximum concentrations (Stenrød et al., 2014), and hence the potential effect concentrations in two ways: MCPA degrades rapidly through aqueous photolysis and may degrade within the analysis is carried out, and dilution of the samples will affect the sampling (Petersen et al., 2013) as the sampling method gives a mean concentration for the sampling period. Thus, indicating that measured concentrations may be even higher than measured and the actually occurring peak concentrations that might have acute toxic effect on aquatic organism is not captured. The sample methodology can be assumed to propose a challenge when measuring environmental concentrations during the winter and early spring as well.

56

Based on the behaviour of MCPA in the reported leaching experiment, unstable temperature conditions during winter with repeated freezing and thawing of the soil could severely increase the environmental concentrations in surface waters during the winter and early spring. Although an extrapolation of the results from the leaching experiment to the field scale is theoretical and with a high degree of uncertainty, the maximum measured concentration in the leaching experiment (table 10), resulted in more than a ten-fold higher maximum measured concentration in leachate from frozen soil as compared to unfrozen soil. This implies that winters with unstable temperature conditions causing intermittent freezing and thawing of the soil as well as rainfall events, may cause increased leaching of mobile pesticides in comparison to more stable winters. Further, cold winters with below zero soil temperatures may lead to leaching of higher pesticides concentrations during snowmelt in spring. Thus, pesticides applied in winter cereal can increase its persistence through low degradation rates during the autumn with low soil temperatures and negligible degradation when the frost sets in. Subsequently moderate rainfall can accumulate on top of a frozen layer of approximately 20 cm or infiltrate through air-filled macropores, causing preferential flow and leaching.

Although the study was limited to the top soil (0-20 cm), further leaching of pesticides through the more fertile top soil down to depths with lower microbiological activity and lower organic carbon content increases the risk of leaching to tile drains and ground waters. Hence, the environmental concentration for mobile pesticides during the summertime, represented by MCPA at Skuterud (see table 11), could potentially be increased during winter with below zero soil temperatures through the dominating processes of decreased degradation rates and increased preferential flow of mobile pesticides to tile drains and subsequent to stream waters after rainfall and thawing. Consequently, the use of mobile autumn applied pesticides in the Skuterud catchment would increase the risk of exposure, increasing the risk of effects in aquatic organisms if transported to tile drains. However, leaching of mobile pesticides to tile drains and streams would cause the pesticides to dilute, thus decreasing the exposure. Pesticide leaching through snowmelt would also dilute due to the increase in water flow. Furthermore, site factors such as hillslopes complicates matters additionally, making it difficult for a realistic assessment at the field scale. Runoff would occur if the impermeable frozen soil layer restricted infiltration in sloped hillsides.

Stenrød et al. (2008) underlines the importance of including freezing and thawing processes in risk assessments in cold climate conditions. However, environmental pesticide concentration levels during cold climate conditions are limited. Thus, modelling tools in compliance with knowledge on the effect of cold climate condition on leaching could be equally important in assessing the risk towards the environment. Still, monitoring programs in the northern countries and other areas with seasonally frozen soils should include pesticide monitoring during the late autumn, winter and early spring.

Statistics Norway (SSB) has monitored the use of pesticides in Norway through several postal sample surveys the last years. Herbicides are the most common types of pesticide sprayed in winter wheat, being applied to more than 95 % of the area sprayed in the period 2003 to 2014. The single most sprayed herbicide in winter wheat in 2014, was the sulfonylurea herbicide Hussar OD (a.i., iodosulfuron) which was applied to 7600 ha (Aarstad & Bjørlo, 2016). Express SX (a.i., tribenuron – methyl) was also widely used this year. As mentioned above, mobile pesticides applied to winter cereals with comparable mobility to MCPA, could potentially behave similar as the reported leaching study and subsequently pose a risk towards aquatic organisms. The mentioned sulfonylureas are considered mobile (Almvik et al., 2008), thus could be comparable to the behaviour of MCPA. Hence, tribenuron-methyl could be transported rapidly through macropores to tile drains upon thawing posing a risk towards the aquatic environment. Algae and aquatic plants are especially at risk due to the high toxicity. Other relevant herbicides is prosulfocarb, which is another of the few herbicides approved for autumn spraying in winter wheat, with a substantial increase in sales statistics over the last few years (Mattilsynet, 2015). However, prosulfocarb is only considered as slightly mobile (University of Hertfordshire, 2017), thus might not behave similarly as the pesticide selected in the study. However, in a study reported by Klaus et al. (2014), shows that particle bound transport could consist of as much as 20 % of the leaching through macropores, indicating that prosulfocarb too, could possibly leach to the tile drains. This could also apply to the slighly mobile glyphosate (University of Hertfordshire, 2017), which is commonly sprayed during autumn before tillage or sowing of winter wheat.

5.4 Climate change and future challenges on the use of pesticides in winter cereal

Northern countries are expected to experience a positive sum effect on agricultural production due to climate change (Olesen & Bindi, 2002). This is mainly due to better conditions for crop production and a prolonged growing season. Consequently, winter cereal production and pesticide usage is expected to increase in the autumn (Seehusen et al., 2016). Hence, the need for knowledge regarding the effect of winter conditions on the leaching of pesticides are important. Future climate challenges could potentially effect winter cereal production and the transport of pesticides through several factors.

Climate change may cause an increase in precipitation in Norway. Subsequently, high amount of precipitation could be damaging to tillage practices and soil erosion etc. affecting cereal production (Greipsland & Stenrød, 2016). In a median emission scenario (RCP 4.5) it is expected that days with heavy rainfall will increase by 48 % in eastern Norway in the period 2071-2100 in comparison to 1971-2000. Looking at the winter season (Des. - Feb.) and early spring (March-May), days with heavy rainfall will increase by 108 % and 76 %, respectively (Hanssen-Bauer et al., 2015). A coincide of heavy rainfall on a thick frozen soil layer will result in surface runoff. The surface runoff could be even more enhanced if the rainfall coincides with snow melting. Heavy rainfall on thin frozen soil layer on agricultural land however might infiltrate and be transported to tile drains and eventually to surface waters.

According to Hanssen-Bauer et al. (2015), temperature will increase during the winter in contrast to the summer. Climate change might also impact the freezing and thawing episodes through increased frequency and magnitude (Mellander et al., 2007). In addition, snow melt during spring may arrive earlier. Herbicides applied to winter cereal has been the primary focus in this thesis. However, in a moister climate, increased use of fungicide could be necessary to control fungal plant diseases in winter cereal. Increased use of fungicides has been a trend in cereal production in Norway in the period of 2001-2014 (Aarstad & Bjørlo, 2016). Prothioconazole (Proline) is fungicide that was legalized for cereal application in 2008. Although prothioconazole is non-persistent and only slightly mobile (University of Hertfordshire, 2017), its key metabolite prothioconazole – destio are frequently detected in

surface waters (Bechmann et al., 2014), indicating that it could also be present in agricultural streams during winter and early spring as well.

To widen the scope even further, autumn spraying of fungicides to control potato late blight could be relevant to leaching during cold climate conditions. Ridomil Gold MZ Peptide with the slightly mobile pesticide active ingredient mancozeb and the very mobile metabolite ETU (University of Hertfordshire, 2017), was used on 47 % of the treated area against fungal diseases (Aarstad & Bjørlo, 2016). The pesticide turnover sales number, shows an increasing trend in the period 2010-2014 (9116, 10720, 13825, 14511, 12684 kg a.i, per year) (Mattilsynet, 2015), supporting the presumed effect of global warming upon the development of fungal diseases.

Thus, in a changing climate with increased temperatures and more moist conditions, fungicides used in winter cereals and to control potato late blight might pose a future challenge on the risk of leaching to surface waters and potentially harm aquatic organisms under cold climate conditions with below zero soil temperatures.

5.5 Uncertainties and limitations

To investigate the transport of pesticides through soils with macropore structures in a small-scale laboratory, it is crucial to limit the disturbance of the soil structure when sampling. However, due to the dry condition of the soil at the time of soil sampling, some disturbances might have occurred, hence undisturbed soil columns were difficult to obtain. Furthermore, the expected spatial variability across the agricultural field along with the relatively low number of soil column replicates collected (five frozen and five unfrozen) were limiting to the extent of the evaluation of the leaching results. The experimental set-up however, was considered the largest attribute to uncertainty, where the main problematic part was the irrigation regime. The rate of the irrigation varied among the tubes and it was observed increased deviance as the temperature in the room was lowered. The thermistor-holes and temperature wires also resulted in some water leakage along the wires, which might had resulted in some bromide and pesticide losses. Furthermore, the evaluation and comparison of the results to existing studies were difficult since few studies have been conducted on the topic, hence the reported leaching study were for the most compared to macropore flow in unfrozen soil.

6. Future research

The significant difference in the amount of MCPA leached from frozen soil columns in comparison to unfrozen soil columns indicates further research is needed on the dissipation of pesticides through sorption/desorption processes and the degree of preferential transport of pesticides realized by rainfall in soil affected by freezing and thawing processes. Future work should include a larger field-scale study to verify the result from the reported leaching study. In addition, x-ray tomography would be helpful to quantify the macropores. Furthermore, there is a lack of monitoring data during winter and early spring of agricultural affected streams. Thus, there is a need for collection of monitoring data at the field scale to further address the effects of cold climate conditions on leaching of pesticides and to conduct risk assessments to evaluate the exposure and effect towards the environment. Modelling tools could be an important asset in conducting risk assessment.

7. Conclusion

A leaching experiment with undisturbed soil columns subjected to freezing and subsequent thawing during rainfall events, demonstrated increased amounts of MCPA leached from frozen soil columns (15-33 %) in comparison to unfrozen soil columns (<1 %), indicating higher degree of macropore flow in frozen compared to unfrozen soil columns. The increased leaching of MCPA was realized through the available accumulated water on top of the temporarily-impermeable frozen soil which caused a wash-out through bypassing the sorption sites through the well-developed macropore network upon thawing. Future challenges such as an increase in the frequency of freezing and thawing of soil and increasing use of pesticides in winter cereals in cold climate areas, may increase the potential risk of leaches of pesticides and exposure to aquatic environment near agricultural fields. Hence, with the future challenges underlining the importance of addressing the effect of cold climate conditions on the leaching of pesticides, pesticide monitoring of agricultural affected streams in Nordic areas with winter cereal productions should be carried during late autumn, winter and early spring.

Referring to the study objective and on the basis on these concluding remarks:

- The hypothesis proposed in section 1.1 is rejected.

8. References

Aarstad, P. A. & Bjørlo, B. (2016). Bruk av plantevernmidler i jordbruket 2014. Reports 2016/02. Statistisk Sentralbyrå.

Almvik, M., Riise, G., Bolli, R., Christiansen, A., Odenmarck, S.R., Børresen, T. & Waage Tveit, C. (2008). Transport of sulfonylurea herbicides from a barley field in Norway: field and laboratory studies. Bioforsk Report 3 (105)

Andersson, M. & Kreuger, J. (2011). Preliminära riktvärden för växtskyddsmedel i ytvatten Beräkning av riktvärden för 64 växtskyddsmedel som saknar svenska riktvärden, Teknisk Report 144 Uppsala: Swedish University of Agricultural Science.

Bechmann, M., Stenrød, M., Pengerud, A., Grønsten, H. A., Deelstra, J. & Eggestad, H. O. (2014). Erosjon og tap av næringsstoffer og plantevernmidler fra jordbruksdominerte nedbørfelt – Sammendragsrapport fra Program for jord- og vannovervåking i landbruket (JOVA) for 1992-2013. Bioforsk Report 9 (84)

Bechmann, M. & Eggestad, H. O. (2016). Temperaturendringer, plantevekst og avrenning. NIBIO POP 2(2).

Beven, K. & Germann, P. (1980). The role of macropores in the hydrology of field soils. Institute of Hydrology. Report no. 69

Beven, K. & Germann, P. (2013). Macropore and Water Flow in Soil Revisited. Water Resources Research 49 (6): 3071-3092.

Commision Regulation (EU) (2011). Commision Regulation (EU) No 546/2011 of 10 June 2011 implementing Regulation (EC) No 1107/2009 of the European Parliament and of the Council as regards uniform principles for evaluation and authorisation of plant protection products.

Deelstra, J., Kværnø, S. H., Skjevdal, R., Vandsemb, S., Eggestad, H. O. & Ludvigsen, G. H. (2005). A general description of the Skuterud catchment. Jordforsk report 61 (5).

Derby, N. E. & Knighton, R. E. (2001). Field-scale Preferential Transport of Water and Chloride Tracer by Depression – Focused Recharge. Journal of Environmental Quality 30(1): 194-199.

European Commision (2000). Directive 2000/60/EC of the European Parliament and of the Council establishing a framework for the Community action in the field of water policy.

European Commission (2003). Technical Guidance Document on Risk Assessment – Part II, Italy: European Chemicals Bureau, Institute for Health and Consumer Protection.

European Commision (2008). Review report for the active substance MCPA. SANCO/4062/2001-final. European Commision, Health and Consumer Protection Directorate – General Directorate D

European Commission (2009). Directive 2009/128/EC of the European Parliament and of the Council of 21 October 2009 establishing a framework for Community action to achieve the sustainable use of pesticides.

European Food Safety Auhority (2013). Guidance on tiered risk assessment for plant protection products for aquatic organisms in edge-of field surface waters. EFSA Journal 11 (7): 3290.

Eklo, O. M., Aspmo, R. & Lode, O. (1994). Runoff and leaching experiments of dichlorprop, MCPA, propiconazole, dimethoate and chlorsulfuron in outdoor lysimeter and field catchemnt aareas. I: Norwegian Journal of Agricultural Sciences: Contamination of pesticides from agricultural and industrial areas to soil and water. Agricultural University of Norway: Supp. 13:53-78.

Flury, M. (1996). Experimental Evidence of Transport of Pesticides through Field Soils—A Review. Journal of Environmental Quality 25(1): 25-45.

Food and Agriculture Organization of the United Nations, Update 2015. (2015). World reference base for soil resources 2014: International soil classification system for naming soils and creating legends for soil maps, Rome: World Soil Resources Reports 106.

Forskrift om plantevernmidler. (2015). FOR-2015-05-06. LMD 455, 2015.

Greipsland, I. & Stenrød, M. (2016). Nedbørsendringer og virkninger på jordbruk. NIBIO POP 2(4).

Greve M.H., Helweg A., Yli-Halla M., Eklo O.M., Nyborg A.A., Solbakken E., Öborn I., Stenström J. & Tiberg E. (1998) Nordic Reference Soils. 1. Characterisation and classification of 13 typical Nordic soil. 2. Sorption of 2, 4-D and glyphosate, Tema Nord Environment 537: 1-106.

Hanssen - Bauer, I., Førland, E. J., Haddeland, I., Hisdal, H., Mayer, S., Nesje, A., Nilsen, J . E. Ø., Sandven, S., Sandø, A. B., Sorteberg, A., Ådlandsvik, B. (Red.). (2015). Klima i Norge 2100: Kunnskapasgrunnlag for klimatilpasning oppdatert i 2015. Miljødirektoratet report 2/2015.

Hauken, M. & Stenrød, M. (2011). Work Methods in JOVA. [Online] Bioforsk. Available at: http://www.bioforsk.no/ikbViewer/page/prosjekt/tema/artikkel?p_dimension_id=18844&p_menu_id=18851&p_sub_id=18845&p_dim2=19697&p_document_id=91511 [Accessed 3. May 2017].

Hauken, M. (Red.) (2015). Jord- og Vannovervåking i landbruket (JOVA). Feltrapporter fra programmet i 2013. Bioforsk report 10 (73).

Hayashi, M., van der Kamp, G. & Schmidt, R. (2003). Focused infiltration of snowmelt water in partially frozen soil under small depressions. Journal of Hydrology 2003(270): 214-229.

Hendrickx, J. M. & Flury, M. (2001). Uniform and Preferential Flow Mechanisms in the Vadose Zone. The National Academic Press.

Hillel, D. (2004). Introduction to Environmental Soil Physics: USA: Academic Press.

Hiller, E., Khun, M., Zemanová, L., Jurkovic. & Bartal, M. (2006). Laboratory study of retention and release of weak acid herbicide MCPA by soils and sediments and leaching potential of MCPA. Plant soil and Environment 52(12): 550-558.

Ireson, A. M., van der Kamp, G., Ferguson, G., Nachshon, U. & Wheather, H. S. (2013). Hydrogeological Processes in Seasonally Frozen Northern Latitudes: Understanding, Gaps and Challenges. Hydrogeology Journal 21 (1): 53-66.

Iwata, Y., Nemoto, M., Hasegawa, S., Yanai, Y., Kuwao, K. & Hirota, T. (2011). Influence of rain, air temperature, and snow cover on subsequent spring-snowmelt infiltration into thin frozen soil layer in nothern Japan. Journal of Hydrology 401 (2011): 165-176.

Jarvis, N. (2007). A review of non-equilibrium water flow and solute transport in soil macropores: principles, controlling factors and consequences for water quality. European Journal of Soil Science 58(3): 523-546.

Jarvis, N., Etana, E. & Stagnitti, F. (2008). Water repellency, near-saturated infiltration and preferential solute transport in a macroporous clay soil. Geoderma 143(3-4): 223-230.

Kitterød, N. O. (2008). Focused flow in the unsaturated zone after surface ponding of snowmelt. Cold Regions Science and Technology 53 (1): 42-55.

Kladivko, E. J., Scoyoc, G. E. van., Monke, E. J., Oates, K . M. & Pask, W. (1991). Pesticide and Nutrient movementinto subsurface tile drains on a silt loam in Indiana. Journal of Environmental Quality, 20 (1): 264-270.

Klaus, J., Zehe, Erwin., Elsner, M., Palm, J., Schneider, D., Schröder, B., Steinbeiss, S., Schaik, L. V. & West, S. (2014). Controls of event-based pesticide leaching in natural soils: A systematic study based on replicated field scale irrigation experiments. Journal of Hydrology 512: 528-539.

Koestel, J. (2016). SoilJ – An ImageJ plugin for semi-automatized image-processing of 3-D X-ray images of soil column. Geophysical Research 18.

Kvernø, S. H. (2011). Variability and uncertainty in soil physical properties: effects of data source on functional criteria. PhD. Norwegian University of Life Science

Kværner, J., Sveistrup, J., Eklo, O. M., Almvik, M., Bolli, R., Stenrød, M. & Haugland, E. (2005). Jordvariasjon, avrenningsmønster, plantevernmidler. Grønn Kunnskap 9(2): 165-171.

Larsbo, M. (2005). An improved Dual-Permeability Model of solute Transport in Structured Soils: Model Development and Parameter Identification in Structured Soils. PhD. Swedish University of Agricultural Sciences.

Larsbo, M., Koestel, J. & Jarvis, N. (2014). Relations Between Macropore Network Characteristics and the Degree of Preferential Solute Transport. Hydrology and Earth System Sciences 18: 5255–5269.

Levy, M. & Berkowitz, B. (2003) Measurement and analysis of non-Fickian dispersion in heterogeneous porous media. Journal of Contaminant Hydrology 64(3-4): 203-226.

Levy, B. S. & Chambers, R. M. (1987). Bromide as a conservative tracer for soil-water studies. Hydrological Processes 1 (4): 385-389.

Mattilsynet (2015). Omsetningsstatistikk for plantevernmidler 2010 – 2014. Mattilsynet, avdeling nasjonale godkjenninger.

Mellander, P. E., Löfvenius, M. O. & Laudon, H. (2007). Climate change impact on snow and soil temperature in boreal Scots pine stands. Climate change 85(1): 79-193.

National Meterological Library and Archive. (2011). Fact Sheet No. 3 - Water in the atmosphere. [Online] Met Office. Availble at: http://www.metoffice.gov.uk/ [Accessed: 2. Feb 2017]

Olesen, J. E. & Bindi, M. (2002). Consequences of climate change for European agricultural productivity, land use and polic - Review. European Journal of Agronomy 16 (4): 239–262.

Pedersen, R. V. & Günther, M., 2017. Søkespekter. [Online]
Available at:
http://www.bioforsk.no/ikbViewer/page/fagomrader/fagomrade/omrade/tema/artikkel?p_document_id=45590&p_dimension_id=15111 [Accessed 3. May 2017].

Petersen, K., Stenrød, M. & Tollefsen, K. E. (2013). Initial environmental risk assessment of combined effects of plant protection products in six different areas in Norway. NIVA Report 6588

Russo, D. & Dagan, G., 1993. Water Flow and Solute Transport in Soils: Development and Applications. Berlin: Springer.

Seehusen, T. et al., 2016. Landbruket i møte med klimaendringene. Effekter av endret klima og behov for tilpasninger - Fagnotater som underlag for arbeidersgruppends hovudrapport [Online] Availble at: www.regjeringen.no [Accessed: 8. May 2017]

Shipitalo, M. J., Edwards, W. M., Owens, L. B. & Dick, W. a. (1989). Initial Storm Effects on Macropore Transport of Surface-Applied Chemicals in No-Till Soil. Soil Science Society of America Journal 54(6): 1530-1536.

Simunek, J., Jarvis, N. J., van Genutchen, M. T. & Gardenas, A. (2003). Review and comparison of models for describing non-equilibrium and preferential flow and transport in the vadose zone. Journal of Hydrolog, 272(10): 14-35.

Solbakken, E., Nyborg, Å., Sperstad, R., Fadnes, K., & Klakegg, O. (2006). Jordsmonnatlas For Norge: Beskrivelse av jordsmonn på dyrka mark i Vestfold. Skog og Landskap 2006 (1).

Spaans, E. J. A. & Baker, J. M. (1996). The Soil Freezing Characteristics: Its Measurement and Similarity to Soil Moisture Characteristic. Soil Science Society American Journal 60 (1): 13-19.

Sperstad, R. & Nyborg, Å. (2008). Beskrivelse av Jordsmonngrupper og Jordsmonnenheter på Dyrka Mark i Norge Karakteristikk, Egenskaper og utbredelse, Ås: Skog og Landskap.

Stadler, D., Stähli, M., Aeby, P. & Flühler, H. (2000). Dye Tracing and Image Analysis for Quantifying Water Infiltration into Frozen Soils. Soil Science Society of America Journal 64(2): 505–516.

Stenberg, B., Johansson M., Pell, M., Sjödahl – Svensson, K., Stenström, J. & Torstensson, L. (1998). Microbial biomass and activities in soil as affected by frozen and cold storage. Soil Biology and Biochemistry 30(3): 393-402.

Stenrød, M., Ludvigesen, G. H., Riise, G., Lundekvam, H., Almvik, M., Tørresen, K. S. & Øygarden, L. (2007). Redusert jordarbeiding og glyfosat En sammenstilling av norske og internasjonale forsknings- og overvåkingsresultater, samt en småskala feltstudie av avrenning av glyfosat ved ulik jordarbeiding. Bioforsk Report 2(105).

Stenrød, M. et al. (2008). Cold climate conditions: Effects on bioavailability and leaching of the mobile pesticide metribuzin in a silt loam soil in Norway. Cold Regions Science and Technology 53(1): 4-15.

Stenrød, M., Lode, O. & Holen, B. (2014). Plantevernmidler i vann - miljørisiko. BIOFORSK TEMA, 9(3).

Stenrød, M. (2015). Long-term trends of pesticides in Norwegian agricultural streams and potential future challenges in northern climate. Acta Agriculturae Scandinavica, Section B - Soil & Plant Science 65 Suppl. 2: 199-216.

Stenrød, M., Almvik, M., Eklo, O. M., Gimsing, A. N., Holten, R., Künnis – Beres, K., Larsbo, M., Putelis, L., Siimes, K., Turka, I. & Uusi – Kämppä. (2016). Pesticide regulatory risk assessment, monitoring, and fate studies in the northern zone: recommendations from a Nordic - Baltic workshop. Environmental Science and Pollution Resarch International 23: 15779-15788.

Stähli, M. & Stadler, D. (1997). Measurement of water and solute dynamics in freezing soil columns with time domain reflectometry. Journal of hydrology 195(1-4): 352-369.

Tarradellas, J., Bitton, G. & Rossel, D. (1997). Soil Ecotoxicology. Boca Raton, Florida: Lewis Publishers.

The European Comission (2009). Regulation (EC) no 1107/2009 of the european parliament and of the council of 21 October 2009 concerning the placing of plant protection products on the market and repealing Council Directives 79/117/EEC and 91/414/EEC, EU: Official Journal of the European Union.

Thorstensen, C. W. & Lode, O. (2001a). Laboratory Degradation Studies of Bentazone, Dichlorprop, MCPA, and Propiconazole in Norwegian Soils. Journal of Environmental Quality 30(3): 947-953.

Uhlen, G., Eklo, O. M. & Lode , O., 1994. Leaching and surface transportation of pesticides in lysimeter experiments and runoff plots. I: Norwegian Journal of Agricultural Sciences: Contamination of pesticides from agricultural and industrial areas to soil and water. Supp. 13

University of Hertfordshire (2017). The Pesticide Properties DataBase (PPDB) developed by the Agriculture & Environment Research Unit (AERU). [Online] University of Hertfordshire. Available at: http://sitem.herts.ac.uk/aeru/ppdb/en/index.htm

Van der Kamp, G., Hayashi, M. & Gallén, D., 2003. Comparing the hydrology of grassed and cultivated catchments in the semi-arid Canadian prairies. Hydrological Processes 17(3): 559-575.

van Genutchen, M. T. & Jury, W. (1987). Progress in unsaturated flow and transport modeling. Reviews of Geophysics – An AGU Journal 25 (2):135-140.

vanLoon, G. W. & Duffy, S. J. (2011). Environmental Chemistry a global perspective. 3 ed. Oxford, Oxford University Press.

VKM (2012). Metodedokument Faggruppe 2 – plantevernmidler. Norwegian Scientific Commitee for Food Safety [Online] 2012. Oslo, Norway. Available at: http://www.vkm.no/dav/4952950786.pdf

VKM (2015). Degradation and mobility of pesticides in Norwegian Soils: Opinion of the Panel on (or the Scientific Committee) of the Norwegian Scientific Committee for Food Safety [Online] VKM Report 2015: 34. Oslo, Norway. Available at: www.vkm.no

VKM (2017). About VKM [Online]. Oslo: Norwegian Scientific Committee for Food Safety. Available at: www.vkm.no [Accessed: 2. Feb. 2017]

Walker, C. H., Sibly, R. M., Hopkin, S. P. & Peakall, D. B. (2012). Principles of Ecotoxicology. United States of America: CRC Press.

Wauchope, R. D., Don, R., Simon, Yeh., Linders, J. B. H. J., Kloskowski, R., Tanaka, K., Rubin, B., Katayama, A., Kordel, W., Gerstl, Z., Lane, M. & Unsworth, J. B. (2002). Review Pesticide soil sorption parameters: theory, measurement, uses, limitations and reliability. Pest Management Science 2002(58): 419-445.

Wilson, L. G., Lorne, G. E. & Cullen , S. J. (1995). Handbook of Vadose Zone Characterization & Monitoring. Boca Raton, Florida: Lewis Publisher.

Øygarden, L., Grønlund, A., Skøien, S., Refsgaard K., Krokann, K., Nordskog, K. & Bechmann, M. (2012). Evaluering av Regional Miljøprogram (RMP) 2011. Bioforsk Report 7(21).

Appendix

Summary of appendixes

No.	Subject
1	Accumulated volume of leachate (mL) during irrigations
2	Breakthrough curves frozen soil columns
3	Breakthrough curves unfrozen soil columns
4	Calculations from method and material and estimated soil moisture
5	Example of calculations from results
6	Soil type (WRB group 2004 version) at the Skuterud catchment
7	Calibration of the ion selective electrode
8	Example of calibration of the peristaltic pumps
9	Pesticide analysis procedure LC – MS/MS
10	Skuterud catchment – use and detections of MCPA
11	Mobility grading of leaching study – adaption from VKM

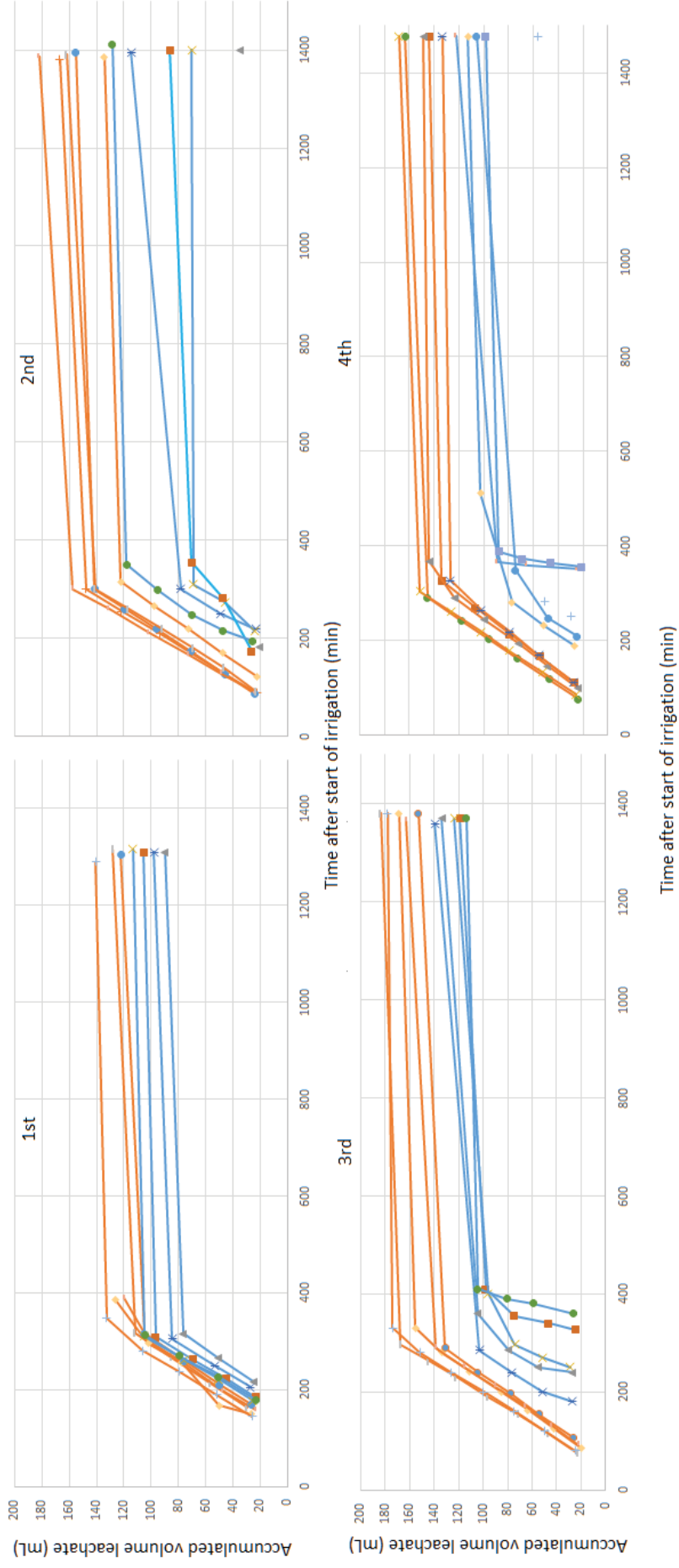APPENDIX 1: Accumulated volume of leachate (mL) during irrigations

Fig. 19. Accumulated volume of leachate (mL) from frozen (blue) and unfrozen (brown) soil columns during four irrigations in a leaching study on a silty clay loam subjected to freeze – thaw cycles.

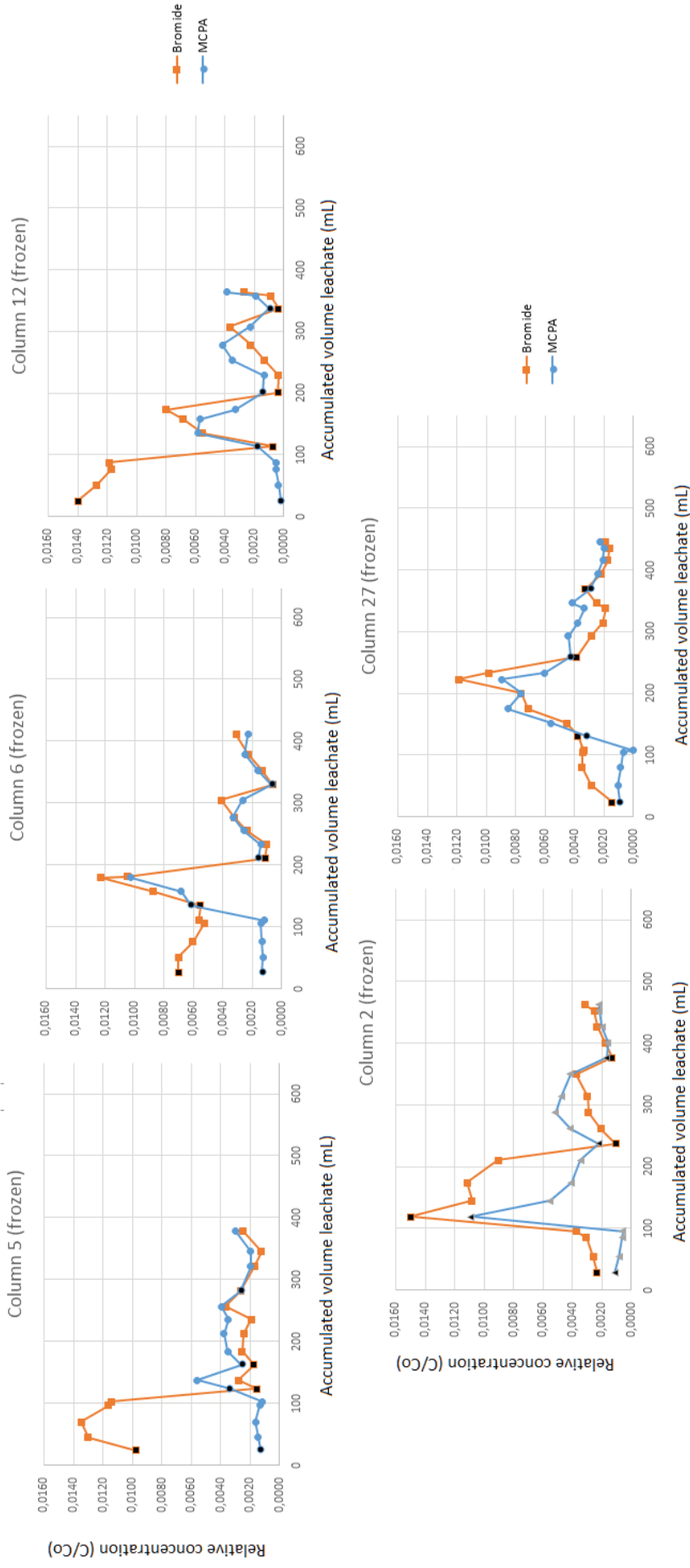# APPENDIX 2: Breakthrough curves frozen soil columns

Fig. 20. Breakthrough curves (BTCs) for bromide and MCPA from frozen soil column, from a leaching study consisting of four irrigations. Black point marks the start of each irrigation.

71

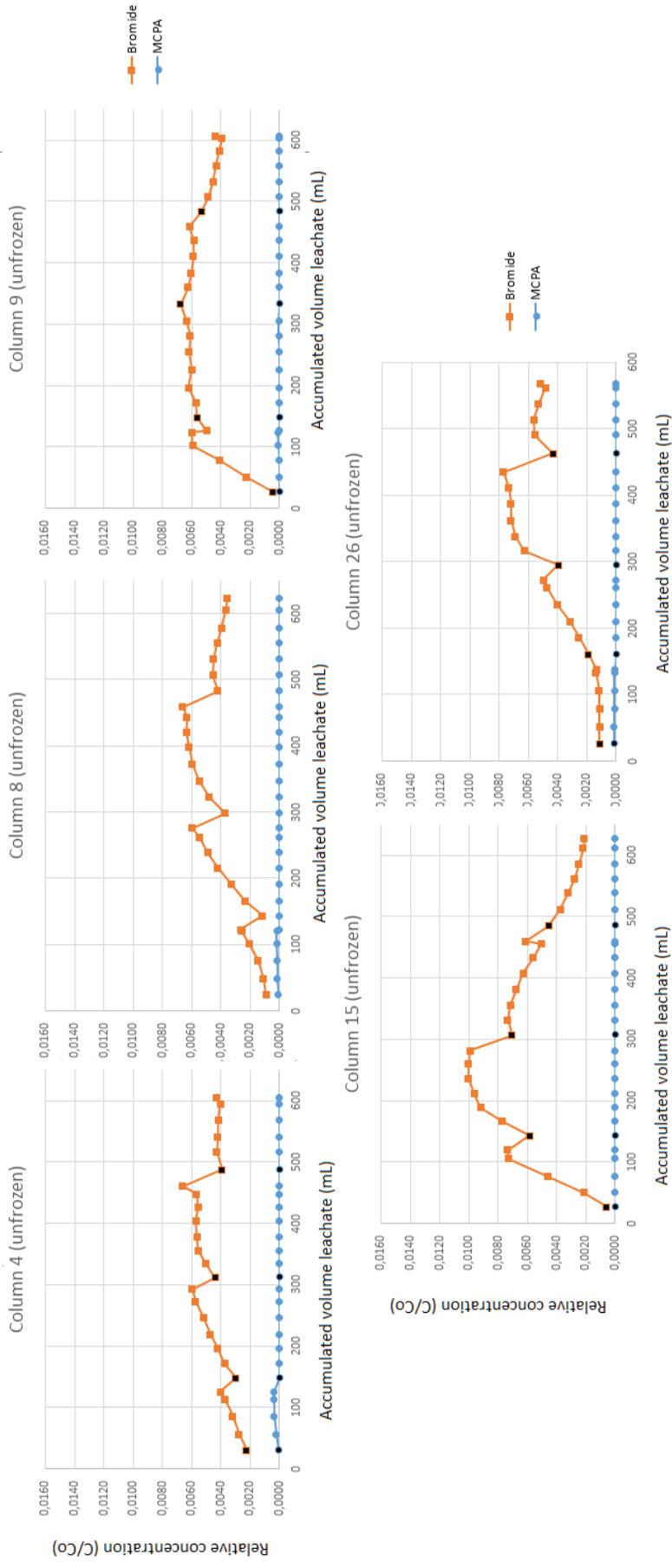# APPENDIX 3: Breakthrough curves unfrozen soil columns

Fig. 21. Breakthrough curves (BTCs) for bromide and MCPA from unfrozen soil columns, from a leaching study consisting of four irrigations. Black point marks the start of each irrigation.
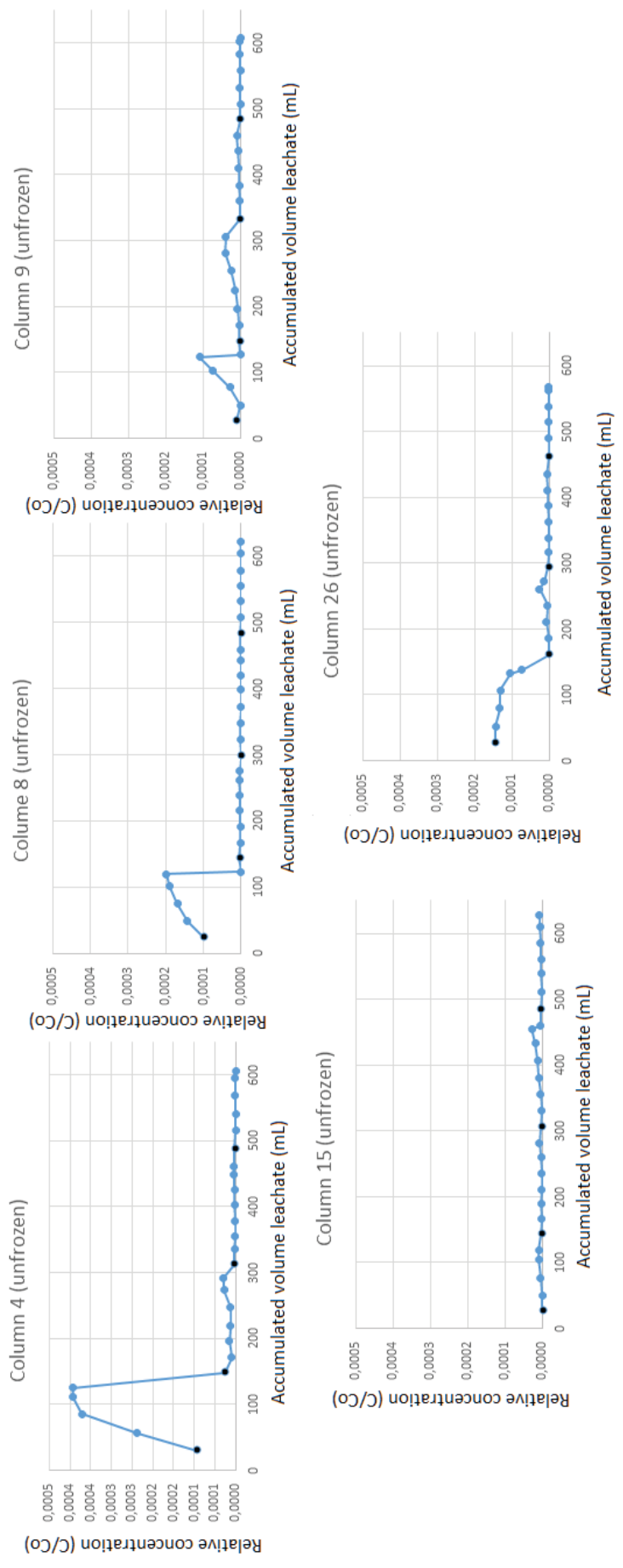
Fig. 22. Breakthrough curves (BTCs) for MCPA from unfrozen soil columns, from a leaching study consisting of four irrigations. Black point marks the start of each irrigation.

APPENDIX 4: Calculations from methods and materials

<u>Infiltration area used in the experiment:</u>

$Area = \pi \times r^2 = \pi \times 5^2$ cm^2 = 78,5 cm^2 = 0,00785 m^2 = 7,85x10^{-7} ha

<u>Actual infiltration area (inner diameter):</u>

$Area = \pi \times r^2 = \pi \times 4,6^2$ cm^2 = 66,5 cm^2 = 0,00665 m^2 = 6,65 x 10^{-7} ha

<u>Amount potassium bromide (KBr) used per column:</u>

Dosage = 74.47 kg/ha

Amount KBr per column used dissolved in 5 mL deionized water =

*Dosage * Area$_{Column}$* = 74.47 kg/ha * 7,85x10^{-7} ha = <u>58,45 mg</u>

Amount per column: 74.47 kg/ha * 6,65 x 10^{-7} ha = 49,52 mg

<u>Concentration (C) KBr:</u>

$C = \dfrac{m}{V} = \dfrac{58,45\ mg}{0,005\ L} = 11690$ mg/L

m = mass (mg)

V = volume (L)

<u>Amount bromide (Br) used per column as a tracer:</u>

Dosage = 50.00 kg/ha

Amount Br per column used dissolved in 5 mL deionized water =

*Dosage * Area$_{Column}$* = 50.00 kg/ha * 7,85x10^{-7} ha = 0,03925 g = <u>39,25 mg</u>

Amount per column: 50.00 kg/ha * 6,65 x 10^{-7} ha = 0,03325 g = 33,25 mg

<u>Concentration (C) Br:</u>

$C = \dfrac{m}{V} = \dfrac{39,25\ mg}{0,005\ L} = 7850$ mg/L

m = mass (mg)

V = volume (L)

<u>Stock Solution MCPA</u>

Due to the high amount of MCPA needed stock solution was firstly made from solid phase:

100 mg of MCPA diluted in 100 mL acetonitrile (1 mg/ml)

Amount MCPA used per column:

Application rate = 1.80 kg/ha

Amount per column used dissolved in 5 mL acetone:

Application rate $*$ Area$_{Column}$ = 1.80 kg/ha $*$ 7,85x10^{-7} ha = 1.413 x 10^{-6} kg = <u>1,413 mg</u>

Concentration (C) MCPA:

$$C = \frac{m}{V} = \frac{1,413 \, mg}{0,005 \, L} = 282,6 \text{ mg/L}$$

m = mass (mg)

V = volume (L)

Pesticides were available from stock solutions at NIBIO with concentration:

1 mg/ml = 1000 mg/L.

Volume pesticide solution needed in the column experiment:

44 columns x 5 mL/column = 220 mL

Decided to make 250 mL pesticide solution.

Dilution:

$$C_1 x V_1 = C_2 x V_2$$

C_1 = Initial Concentration

V_1 = Initial volume

C_2 = Final Concentration

V_2 = Final Volume

$$V_1 = \frac{C_2 x V_2}{C_1} = \frac{282,6 \, mg/L \, x \, 0,250 \, L}{1000 mg/L} = 0,07065 \, L = 70,65 \, mL$$

The calculated pesticide volume were extracted from the stock solution, mixed and diluted

with 250 mL acetone.

(1) Volume column/cylinder: π x 20 x 5^2 = 1570,80 cm^3

(2) Volume column/cylinder: π x 18 x 4.6^2 = 1196,57 cm^3 (real)

Estimated porevolume:

(1) 0,50 * 1570,79 cm^3 = 785,0 cm^3 = 785 mL

(2) 0,496^2 * 1196,57 cm^3 = 593,5 cm^3 = 593,5 mL

Irrigation rate (Theoretical):

Moderate rain: 5 mm/hour

Moderate rain for 5 hours: 25 mm

1 mm = 1 liter per m^2

Volume per hour = 5 liter/m^2 x 0,00785 m^2 = 0,03925 L = 39,25 mL

Total volume per irrigation per column = 39,25 mL x 5 hours = 196,25 mL

Volume water columns received in total (4 irrigations) = 4 x 196,25 mL = 785 mL

Indication of soil moisture in the soil columns

Table 12. Measurement of moisture a soil column using a HH2 Moisture Meter delivered by Delta – T. Column not used in the experiment due to the risk of having altered the pore structure.

Time of measurement	Water content (%)
Prior to being placed in water	25,1
After absorbing water and pre-equilibration	30,6
After equilibration (-30 cm)	29,9

---

[2] Porvolum RK8, Rustad

APPENDIX 5: Example of calculations from results

Example of calculations 1st irrigation: Column no. 2

Initial concentration bromide 7850 mg/L

Amount of bromide added to each column: 39.25 mg

Table 13. Example of calculation for soil column no. 2 first irrigation.

			Column no. 2 1st irrigation				
Sample no.	Sample collected: Time after start irrigation (min)	Measured bromide concentration (mg/L)	Total volume leachate sample (mL)	Amount of bromide leached (mg)	Accumulated amount of bromide leached (mg)	Accumulated amount of bromide leached in % of the applied amount	Relative concentration bromide (C/Co)
1	205	18.33	27.19	0.50	0.50	1.27	0.002
2	250	20.34	26.39	0.54	1.04	2.64	0.002
3	305	24.06	31.1	0.75	1.78	4.54	0.003
4	1310	29.32	10.28	0.30	2.08	5.31	0.003
Total				2.08			

Amount of bromide leached (mg):

$$Measured\ bromide\ concentration \left(\frac{mg}{L}\right) * \frac{Total\ volume\ of\ leachate\ sample\ (mL)}{1000} = 18.33 * \left(\frac{27.19}{1000}\right) \approx$$

0.50

Accumulated amount of bromide leached in column no. 2:

1. 0.50

2. 0.50 + 0.54

3. 0.50 + 0.54 + 0.75

4. 0.50 + 0.54 + 0.75 + 0.30

Total amount of bromide leached: 0.50 + 0.54 + 0.75 + 0.30 + 2.08

Accumulated volume leachate (mL):

Same as for accumulated amount of bromide leached only with respect to total volume leachate sample

Amount of bromide leached in column no. 2 in the 1st irrigation in percentage of the applied amount:

$$\frac{Total\ amount\ of\ bromide\ leached\ (mg)}{Amount\ of\ bromide\ added\ to\ column\ (mg)} * 100 = \frac{2.08}{39.25} * 100 = 5.3\ \%\ \text{(see Fig. 11 A; lowest frozen soil}$$

column

Relative concentration bromide (C/Co):

$$\frac{\textit{Measured bromide concentration } (\frac{mg}{L})}{\textit{Initial concentration bromide } (\frac{mg}{l})} = \frac{18.33}{7850} = 0.002$$

Table 14. Accumulated amount of bromide (mg) leached from frozen and unfrozen columns in the experiment for all irrigations and total accumulated amount of bromide leached (mg) after four irrigations. Percentage accumulated amount of bromide leached at each irrigation of the total amount detected in the percolate are given in bracket for irrigations were recovery of bromide was highest.

Column: frozen, unfrozen	1. Irrigation		2. Irrigation		3. Irrigation		4. Irrigation		Total	
	frozen columns (mg)	unfrozen columns (mg)	frozen columns (mg)	unfrozen columns (mg)	frozen columns (mg)	unfrozen columns (mg)	frozen columns (mg)	unfrozen columns (mg)	frozen columns (mg)	unfrozen columns (mg)
2, 4	2.08(12.3)	3.03(14.4)	10.10(59.7)	6.11(28.9)	2.88(17.0)	7.27(34.4)	1.85(10.9)	4.70(22.3)	16.92	21.12
5, 8	9.60(66.7)	1.49(7.6)	0,57(4)	4.61(23.6)	2.27(15.8)	8.14(41.7)	1.95(13.6)	5.28(27.0)	14.39	19.53
6, 9	5.43(38.2)	3.55(14.3)	4.88(34.3)	8.64(34.8)	2.31(16.2)	7.40(29.8)	1.61(11.3)	5.27(21.2)	14.22	24.86
12, 15	8.66(61.7)	3.94(13.9)	3.29(23.4)	11.34(39.6)	1.72(12.3)	9.04(31.9)	0.37(2.6)	4.07(14.3)	14.04	28.38
27, 26	2.36(17.1)	1.31(6.9)	7.27(52.8)	3.63(19.2)	2.43(17.7)	8.56(45.4)	1.70(12.4)	5.36(28.4)	13.76	18.86

Table 15. Accumulated amount of MCPA (µg) leached from frozen and unfrozen columns in the experiment for all irrigations and total accumulated amount of MCPA leached (µg) after four irrigations. Percentage accumulated amount of MCPA leached at each irrigation of the total amount detected in the percolate are given in bracket for irrigations. (MCPA leached of the applied amount (1413 µg) is presented in 4.1.2 and 4.1.3 for bromide and MCPA, respectively).

	MCPA leached									
	1. Irrigation		2. Irrigation		3. Irrigation		4. Irrigation		Total	
Column: frozen, unfrozen	frozen columns (µg)	unfrozen columns (µg)	frozen columns (µg)	unfrozen columns (µg)	frozen columns (µg)	unfrozen columns (µg)	frozen columns (µg)	unfrozen columns (µg)	frozen columns (µg)	unfrozen columns (µg)
2, 4	22.2(5)	10.0 (91)	181.6 (43)	0.9(8)	160.3(38)	0.1(1)	60.4(14)	0.0(0)	424.5	11.1
5, 8	41.6(14)	5.4 (97)	42.2 (15)	0.1(3)	116.2(41)	0.0(0)	83.0(29)	0.0(0)	283.0	5.6
6, 9	42.2(13)	1.4 (55)	150.9 (46)	0.9(37)	79.6(24)	0.2(6)	55.1(17)	0.0(1)	327.8	2.6
12, 15	10.1(4)	0.1 (15)	100.1 (43)	0.1(13)	95.0(41)	0.5(51)	26.2(11)	0.2(18)	231.3	1.0
27, 26	26.4(6)	5.0 (91)	242.6 (52)	0.3(6)	129.4(28)	0.1(2)	66.0(14)	0.0(0)	464.4	5.5

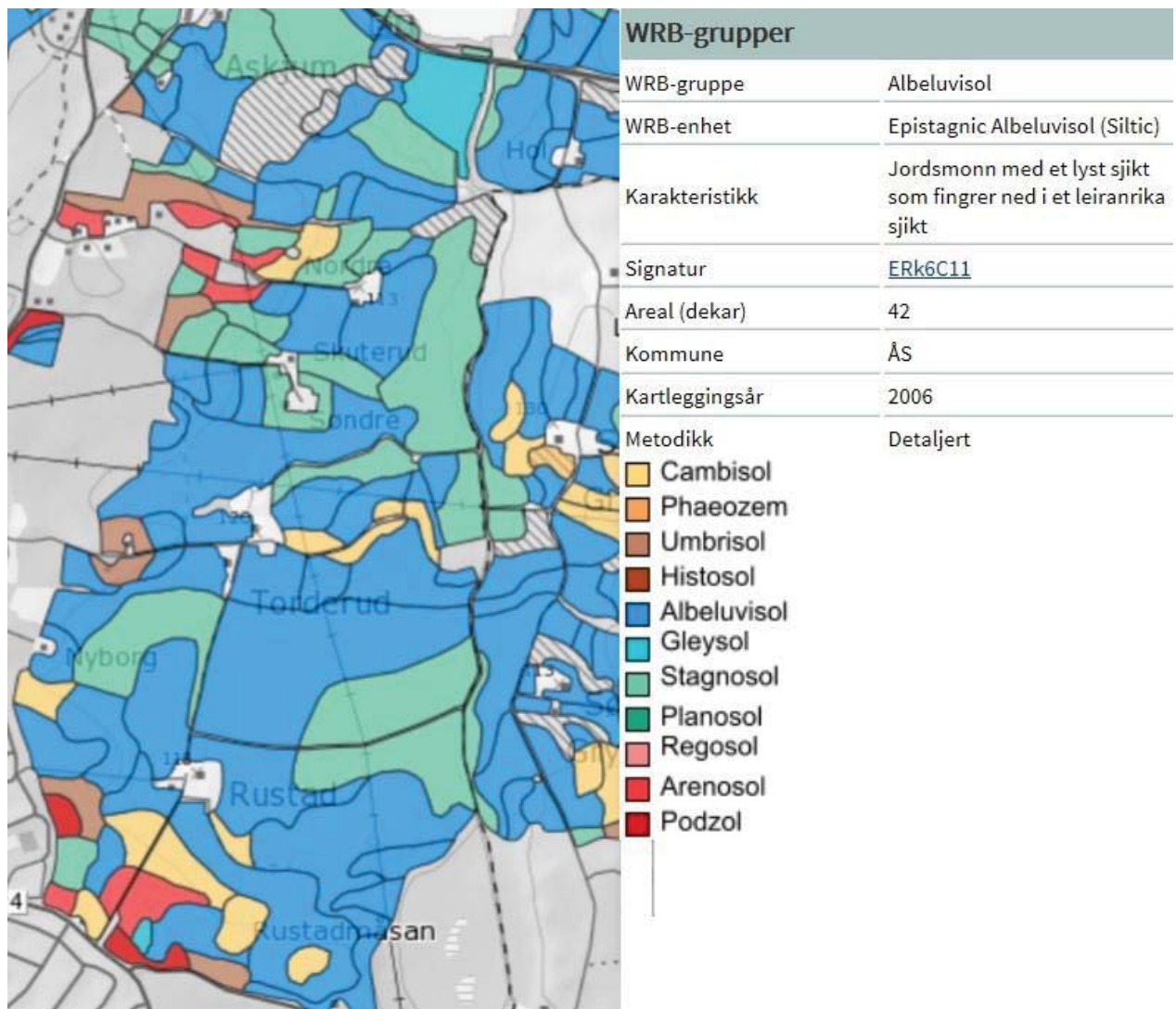APPENDIX 6: Soil type dominating the Skuterud catchment

Fig. 23. Dominating soil types in the Skuterud catchment. (Source: kilden.nibio.no, attrieved 09.02.2017)

APPENDIX 7: Calibration of the ion selective electrode

Procedure:

Two standards of 1000 ppm and 10 ppm was made. 1000 ppm bromide standard was made by diluting 12,52 mL bromide standard of 0,1 M; Orion cat. No. 943506 in 100 mL MiliQ water. The standard was stored in a plastic flask and valid for 1 week. 10 ppm Bromide standard was made by extracting 1 mL of the 1000 ppm bromide standard and 1 mL ISA diluted in 100 mL water. The table shows volume and concentration added to a 150 mL beaker containing 2 mL ISA and 100 mL water. The calibration curve was out of range if: slope < 85 % or >110 %, offset < - 60 mV or > + 60 mV and in the range between 54 – 60 mV/pX.

Table 16. Construction of a linear calibration curve from 1000 ppm and 10 ppm bromide standards.

Standard	Volume added	Concentration (ppm)
10 ppm	4.6 ml	0.43
10 ppm	30 ml	2.53
1000 ppm	2 ml	16.9
1000 ppm	4 ml	44.5
1000 ppm	10 mL	128.2

APPENDIX 8: Example of calibration of the peristaltic pumps

Table 17. Pump performance test at time one to control the irrigation rate

Irrigation rate (mL)					
Pump 1-10	1 hr	2 hr	3 hr	4 hr	5 hr
Slange 1	37	75	110	149	183
Slange 2	35	69	102	135	167
Slange 3	35	70	103	137	169
Slange 4	37	73	106	141	173
Slange 5	34	66	98	129	161
Slange 6	37	73	108	142	175
Slange 7	35	70	104	136	167
Slange 8	35	70	104	137	170
Slange 9	43	86	128	170	210
Slange 10	37	74	109	143	177

Table 18. Pump performance test at time two to control the irrigation rate

Irrigation rate (mL)					
Pump 1-10	1 hr	2 hr	3 hr	4 hr	5 hr
Slange 1	37	74	110	145	182
Slange 2	35	69	102	135	170
Slange 3	34	66	99	130	163
Slange 4	35	70	103	136	170
Slange 5	36	70	104	137	172
Slange 6	44	84	125	165	206
Slange 7	38	76	112	147	183
Slange 8	36	72	106	140	174
Slange 9	40	78	119	156	195
Slange 10	39	75	111	146	182

Pump performance: irrigation rate test of the autoclude pump at two different time one month apart: ($p > 0,05$)

APPENDIX 9: Pesticide analysis LC – MS/MS

Procedure:

Detection and quantification of MCPA and 2 – MCP were performed using Water Alliance 2695 LC – system coupled to Quattro Ultima Pt triple quadruple mass spectrometry by Micromass (Manchester, UK). 5 µl of sample volume was injected and the analytes were separated with 5mM formic acid (A) and methanol (B) as the mobile phase using Phenomenex Gemini C18 (100x2 mm, 3µm diameter). Initial mobile phase gradient was 30 % methanol which increased to 95 % after eleven minutes. Flow velocity was set to 0.3 ml/min and the column temperature to 30 °C. Total runtime per sample was 16 min. The analytes were ionized to anions then to gas phase at 3.0 kV, ion source temperature 120 °C and desolvation temperature at 35° C. using an electrospray ion source. The first MS was adjusted to filter MCPA (m/z 198.90 and 200.02) and 2 – MCP (m/z 140.95), the analytes were thereafter fragmented. Second MS were set to (mz 198.90> 140.88 and 200.02 > 141.90) and 2-MCP (pseudotransfer mz 140.95 > 140.95). The quantification was based on peak area of the fragment 140.88 (retention 7.4 min) and 140.95 (retention 6.2 min). Calibration curve of 5 – points with MCPA and 2 – MCP dissolved in MiliQ in the range 0.5 – 500 ng/ml were used to calculate the concentration of the samples. 2,4 – D, internal standard (IS) were added to all samples and calibration standards to adjust for variance during the analysis and was detected at transition mz 218.82 > 160.73.
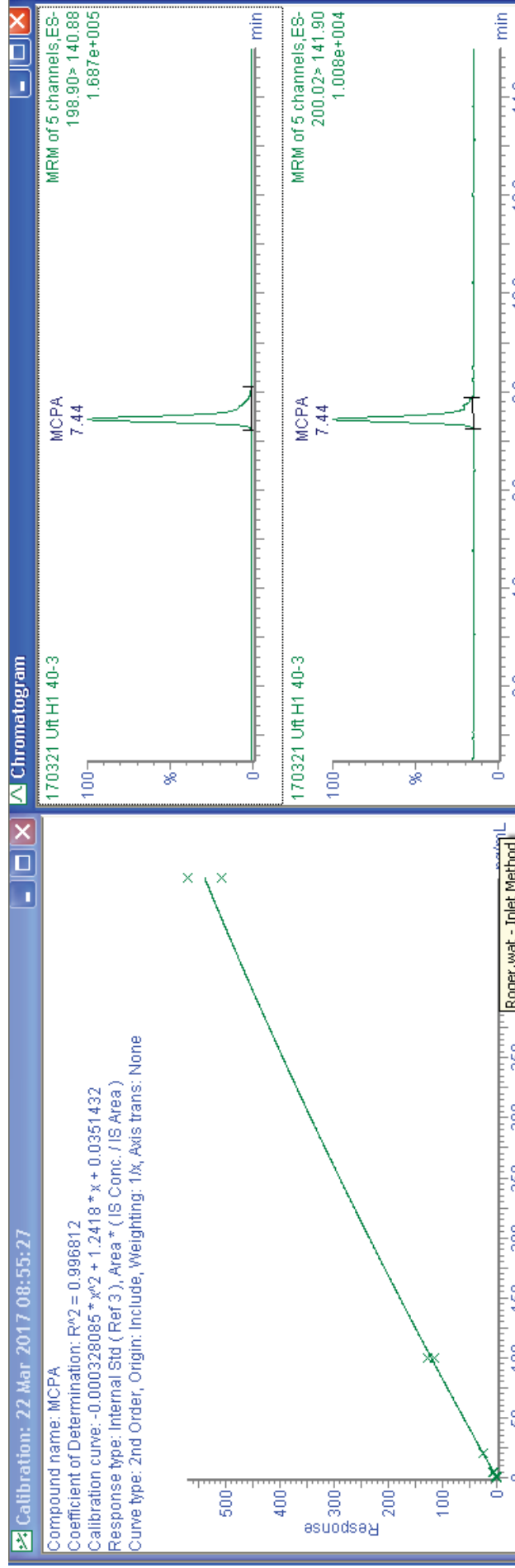
Fig. 24 Chromatogram of the standard (5ng/ml) and sample (32 ng/ml)

APPENDIX 10: Skuterud catchment - Use and detections of MCPA
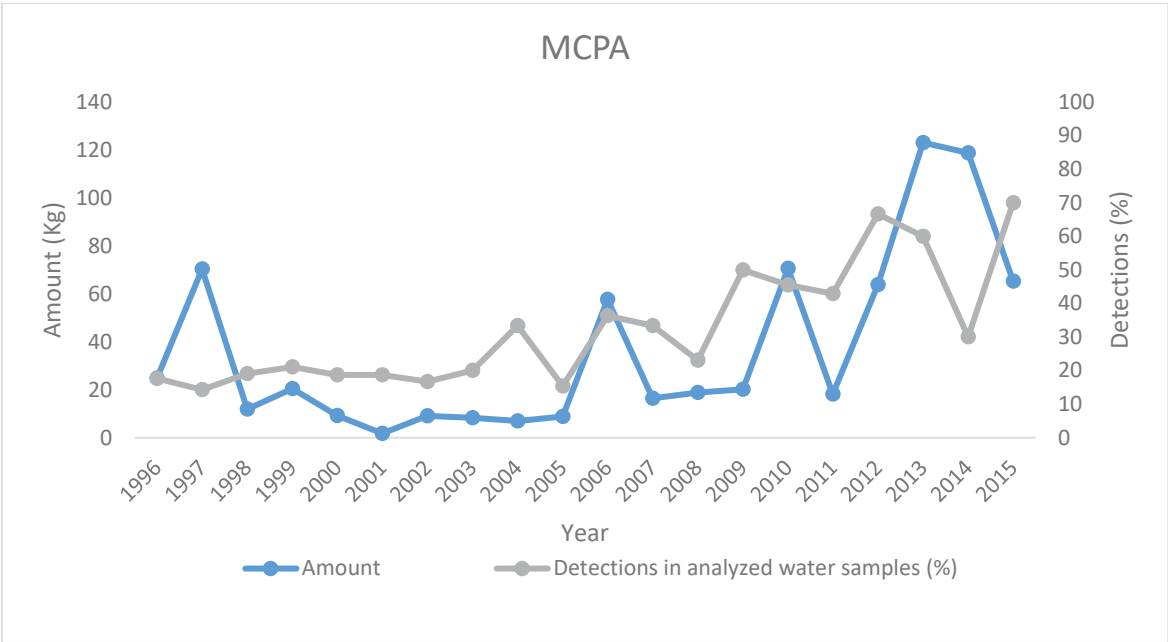

Fig. 25. Total applied amount per year (Kg), detections of MCPA in percent of analysed water samples through the growing season at the Skuterud catchment.

APPENDIX 11: VKM grading of leaching of pesticides in lysimeter experiments and field experiments

Table 19. Mobility grading of leaching of pesticides from soil columns experiments

Soil Column Experiment	
% leaching of the applied amount	Mobility grading
> 5	Very high
2-5	High
1-2	Middle
0.5-1	Moderate
< 0.5	Low

Adaption from VKM (2012).