Joint ICE, IEMA, CIWEM and National Telford Institute Event

WETLAND SYSTEMS
Storm Water Management Control

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Sustainable Flood Retention Basins

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Introduction

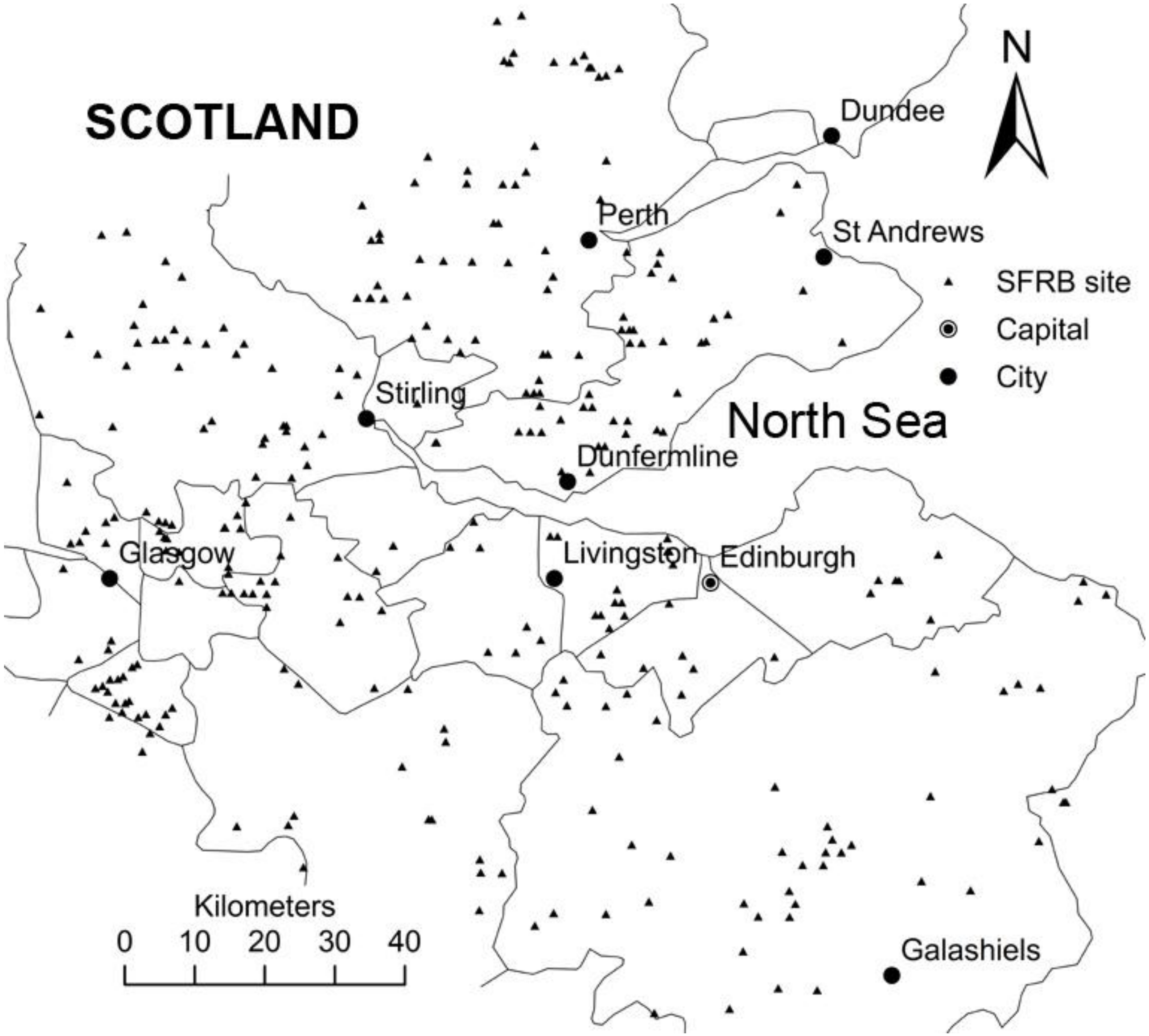
- The **EU Flood Directive 2007** requires member states to undertake **assessments** of artificial and natural **water bodies** including wetlands for their potential to contribute to **sustainable flood risk management** by 2013.
- A logical **concept** to **locate**, **classify** and **assess wetlands**, retention structures and flood defence structures would be highly valuable for **sustainable flood risk management planning**.

Introduction

“A **Sustainable Flood Retention Basin** is defined as an impoundment or integrated **wetland**, which has a pre-defined or potential role in **flood and diffuse pollution control** that can be accomplished cost effectively through best management practice, **achieving sustainable flood risk management** and enhancing sustainable drainage, pollution reduction, biodiversity, green space, and recreational opportunities for society.” (Scholz, 2006)

Introduction: Site Identification

Study area



Introduction: Variables and Data Structure

List of SFRB **variables**

ID	Variable and unit	ID	Variable and unit
1	Engineered (%)	21	Impermeable Soil Proportion (%)
2	Dam Height (m)	22	Seasonal Influence (%)
3	Dam Length (m)	**23**	Site Elevation (m)
4	Outlet Arrangement and Operation (%)	**24**	Vegetation Cover (%)
5	Aquatic Animal Passage (%)	25	Algal Cover in Summer (%)
6	Land Animal Passage (%)	26	Relative Total Pollution (%)
7	Floodplain Elevation (m)	**27**	Mean Sediment Depth (cm)
8	Basin and Channel Connectivity (m)	28	Organic Sediment Proportion (%)
9	Wetness (%)	**29**	Flotsam Cover (%)
10	Proportion of Flow within Channel (%)	**30**	Catchment Size (km^2)
11	Mean Flooding Depth (m)	**31**	Urban Catchment Proportion (%)
12	Typical Wetness Duration ($d\ a^{-1}$)	32	Arable Catchment Proportion (%)
13	Estimated Flood Duration ($d\ a^{-1}$)	**33**	Pasture Catchment Proportion (%)
14	Basin Bed Gradient (%)	**34**	Forest Catchment Proportion (%)
15	Mean Basin Flood Velocity ($cm\ s^{-1}$)	35	Natural Catchment Proportion (%)
16	Wetted Perimeter (m)	36	Groundwater Infiltration (%)
17	Maximum Flood Water Volume (m^3)	**37**	Mean Depth of the Basin (m)
18	Flood Water Surface Area (m^2)	**38**	Length of Basin (m)
19	Mean Annual Rainfall (mm)	**39**	Width of Basin (m)
20	Drainage ($cm\ d^{-1}$)	40	Viniculture Catchment Proportion (%)

Introduction: SFRB Types

- Hydraulic Flood Retention Basin (SFRB Type 1)
- Traditional Flood Retention Basin (SFRB Type 2)
- Sustainable Flood Retention Wetland (SFRB Type 3)
- Aesthetic Flood Treatment Wetland (SFRB Type 4)
- Integrated Flood Retention Wetland (SFRB Type 5)
- Natural Flood Retention Wetland (SFRB Type 6)

Introduction: SFRB Types

- Drinking water and hydropower reservoirs (Type 1).
- Former drinking water reservoirs and traditional retention basins (Type 2).
- Sustainable drainage systems (Type 3).
- Constructed wetlands (Type 4).
- Artificial water bodies within parks (Type 5).
- Natural wetlands (Type 6).

Introduction: Overall Aim

- To outline the SFRB concept and methodology for **using cost-effective** and reliable **variables** to **predict time-consuming**, **costly** and dependant **variables** with the aid of a **self-organizing map model**; and
- To outline the methodology on how to **predict the SFRB types** with the self-organizing map model.

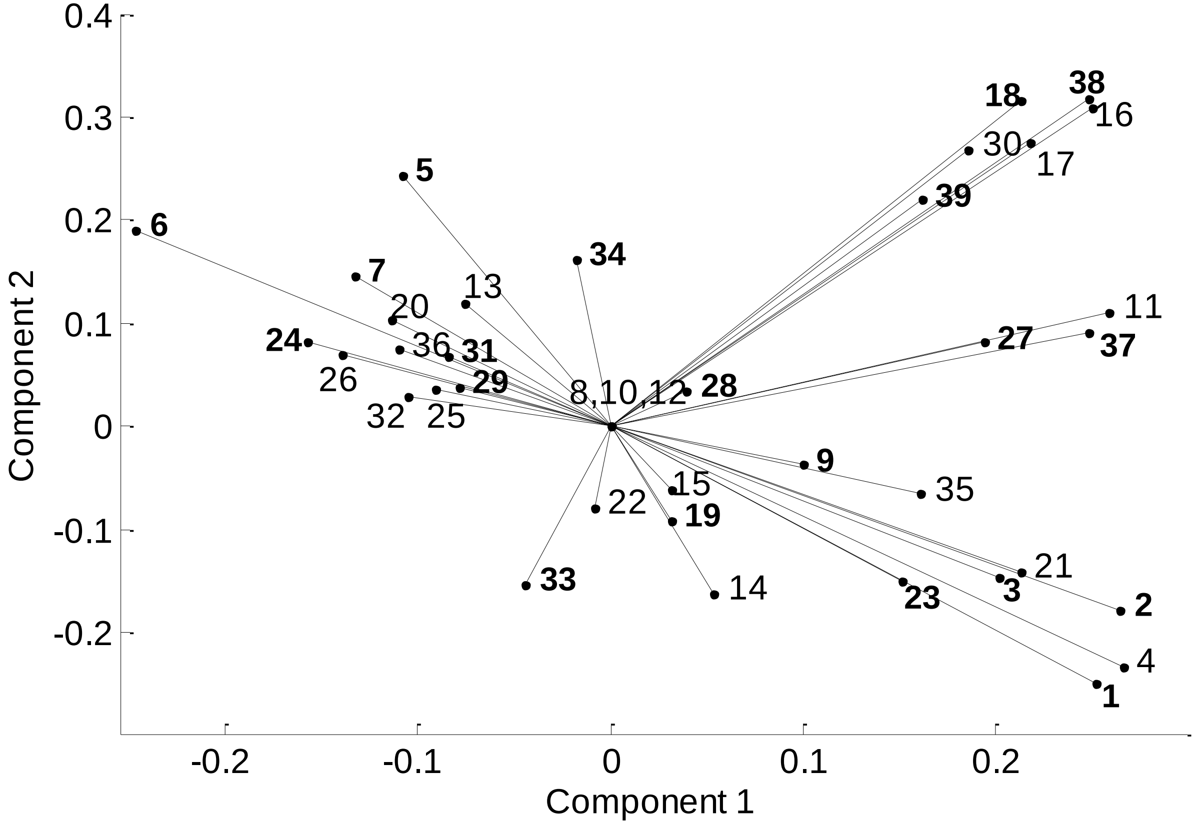
Methodology

- Apply a **principal component analysis** to **identify relationships** between variables.
- Use the **self-organizing map** to **visualize** the **links** among variables.
- Apply the **self-organizing map** to **identify** the important and difficult to obtain variables and other easier-to-determine and inexpensive **variables**.

Methodology

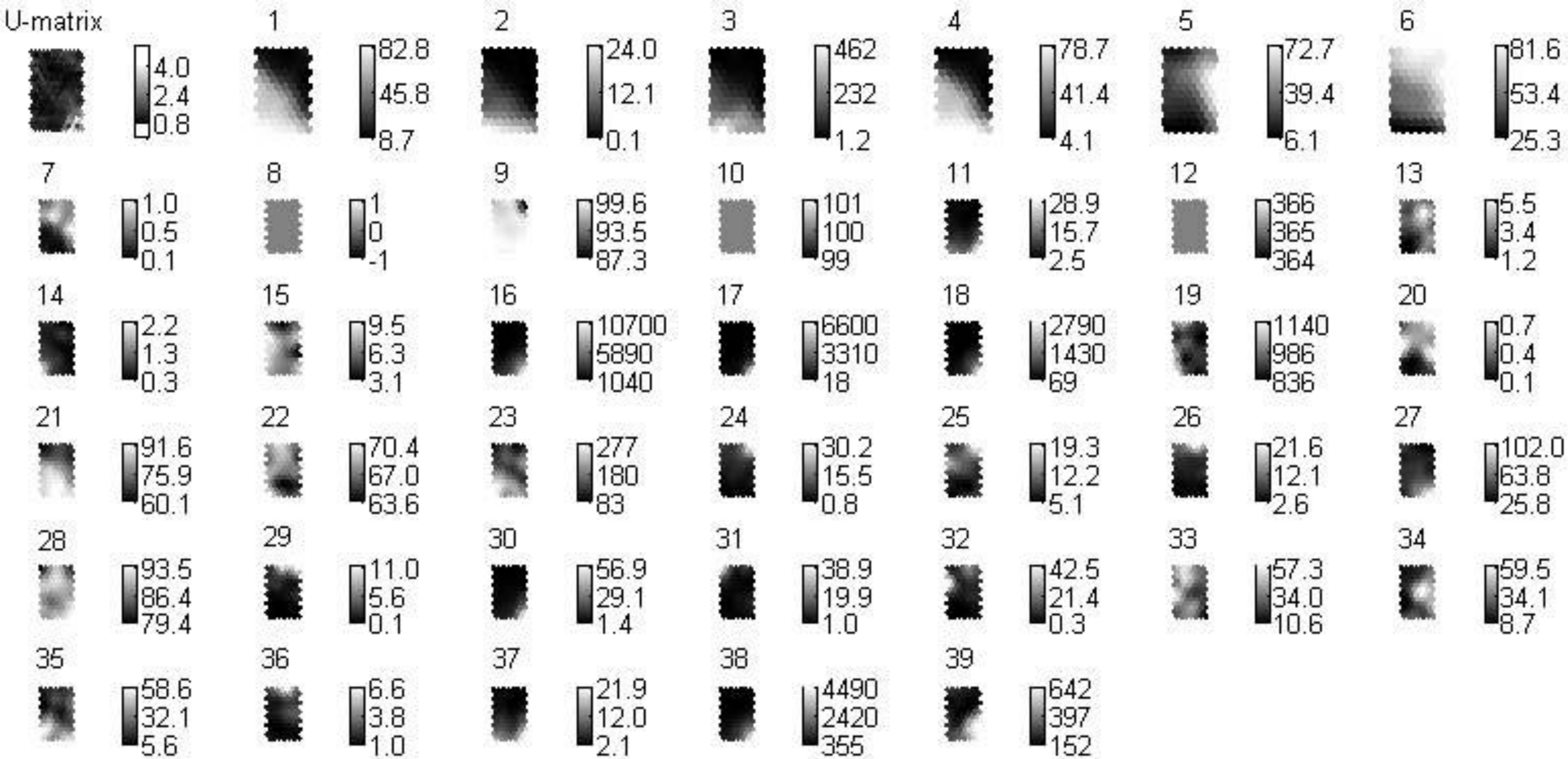
- Apply the self-organizing map model to **predict** difficult and expensive **variables** and **SFRB types**.
- Software package: **Matlab 7.1** for statistical analysis and modeling.

Methodology: Principal Component Analysis



Loading plot of the **principal component analysis** identifying the most important classification variables and their similarities with each other

Methodology:
Self-organizing Map Model



Self-organizing map: Similar and corresponding patterns between variables indicate a close relationship; therefore, some variables can be eliminated from classification

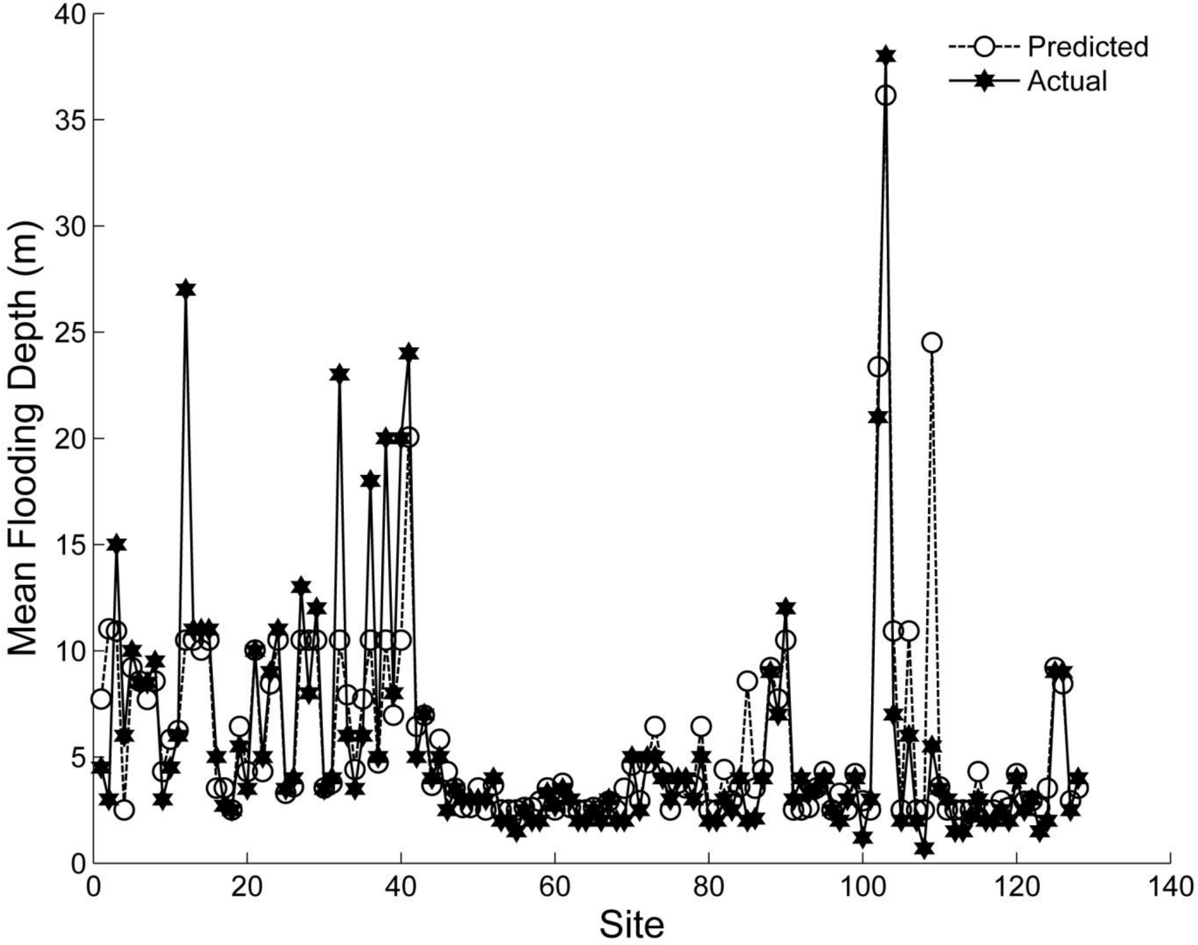
Results and Discussion

- With the help of a **principal component analysis**, **20 groups of variables** (with similar contribution to classification) were **identified**.
- Therefore, **20 key variables** representing one group each were **identified**.
- The **self-organizing model** was successfully applied to **visualize** and subsequently **verify** the **relationships** identified by the principal component analysis.

Results and Discussion

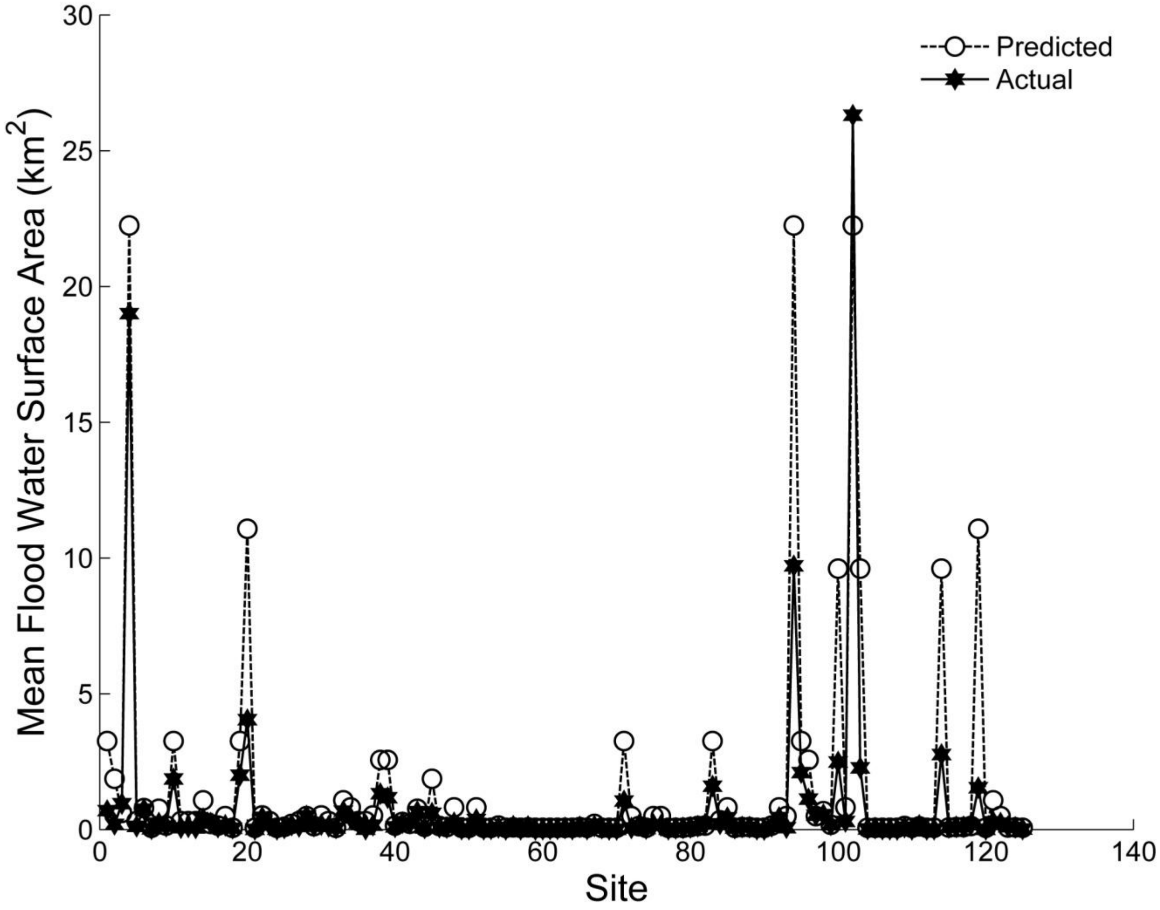
- For example, **low** data values for the variable ***Land Animal Passage*** are difficult to determine, but are **linked** to **high** values for the variables ***Engineered*** and ***Outlet Arrangement and Operation***.
- This finding **verifies** their **negative correlation** identified using principal component analysis.

Results and Discussion



Using *Wetted Perimeter*, *Flood Water Surface Area*, *Catchment Size*, *Mean Depth of the Basin*, *Length of Basin*, and *Width of Basin* to predict ***Mean Flooding Depth***

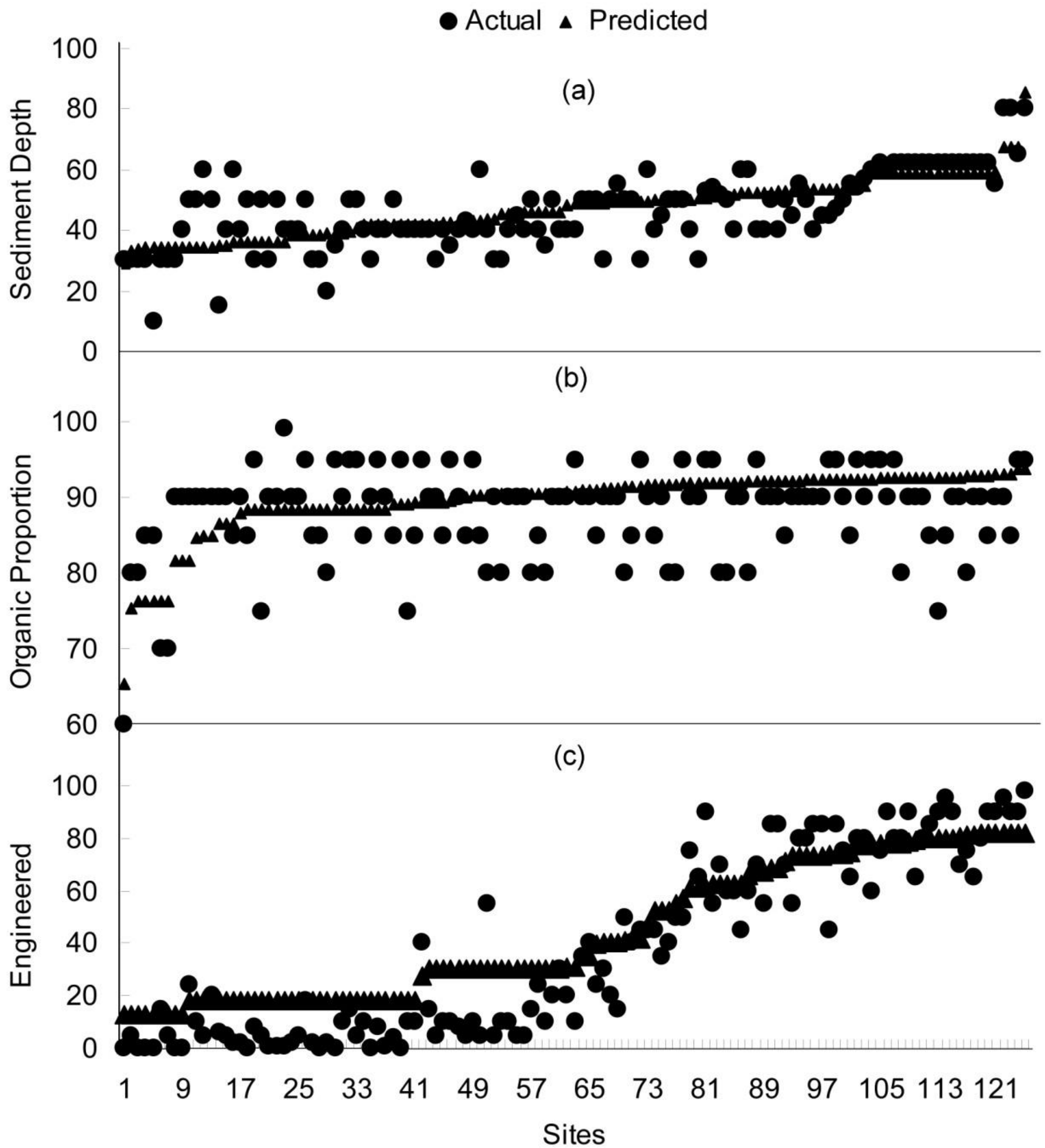
Results and Discussion



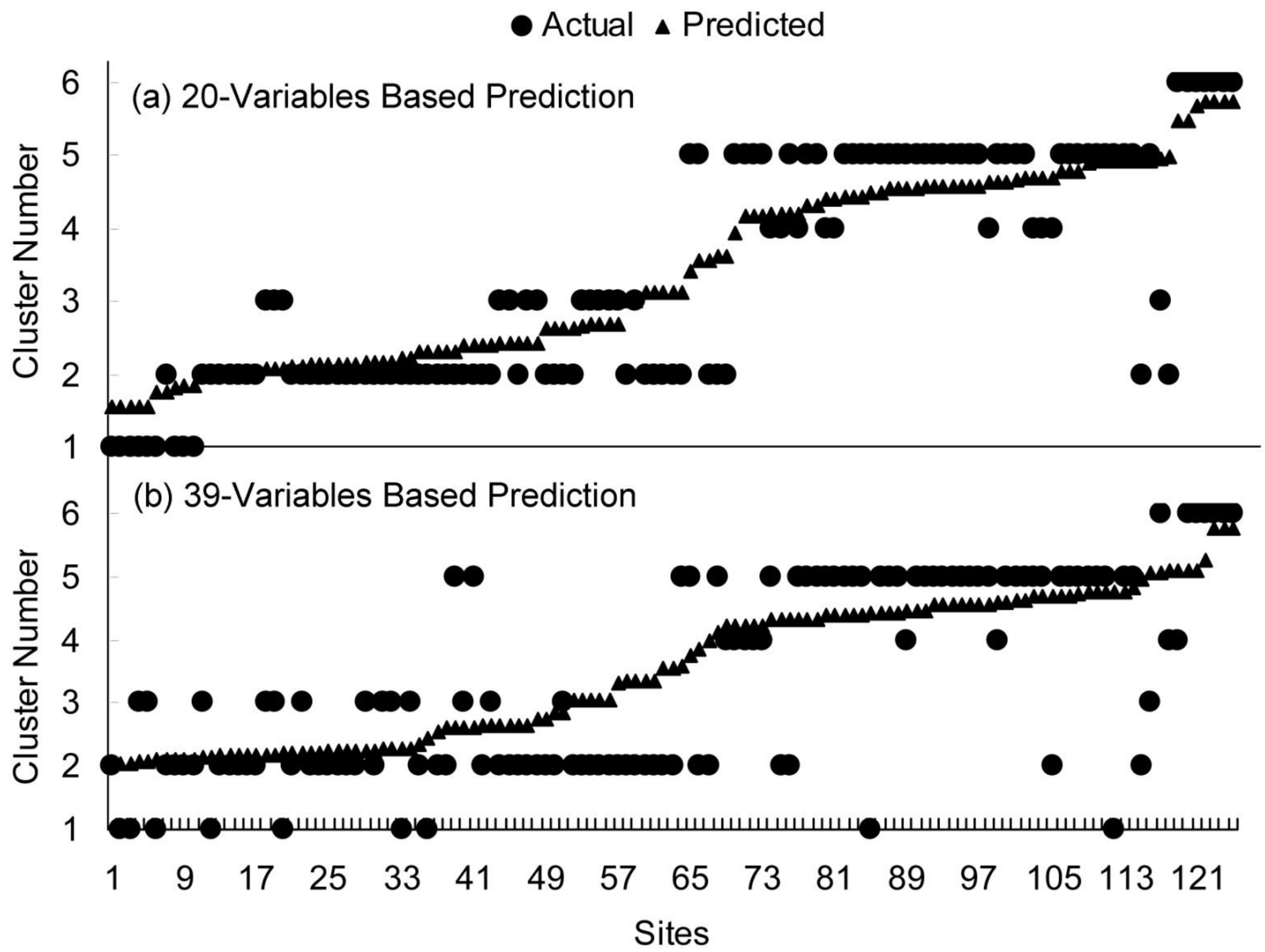
Using *Catchment Size*, *Mean Depth of the Basin*, *Length of Basin* and *Width of Basin* to estimate ***Flood Water Surface Area***

Results and Discussion

Prediction results for the variables (a) ***Sediment Depth***, (b) ***Organic Proportion*** and (c) ***Engineered***



Results and Discussion



Prediction of SFRB types with the self-organizing map model based on (a) 20 and (b) 39 variables

Conclusions

- The **20 most important variables** were identified to **classify water bodies** including wetlands and SFRB.
- The visualization of the relationships between variables displayed by the **self-organizing map** was **verified** the results of the **principal component analysis**.

Conclusions

- Using highly correlated variables, which are easy to determine, to **predict costly** and difficult to obtain **variables** makes the SFRB assessment more **rapid** and **cost efficiently**.
- The self-organizing map model presents a **rapid and reliable** method of **predicting** different **types of water bodies** (i.e. SFRB types 1 to 6).

Conclusions

- The **prediction model** can help to **avoid misunderstandings** concerning the water body status between different stakeholders and the scientific community.
- Learning the function and the status of water bodies will **help decision-makers** in **managing land use** in a sustainable manner and developing **flood risk management plans**.

Integrated Constructed Wetlands

- Background
- Objectives
- Materials and Methods
- Results and Discussion
    - *Treatment Performance*
    - *Community Analysis*
    - *Bacterial Community Character*
    - *Microbial Community Assembly*
- Conclusions

Background

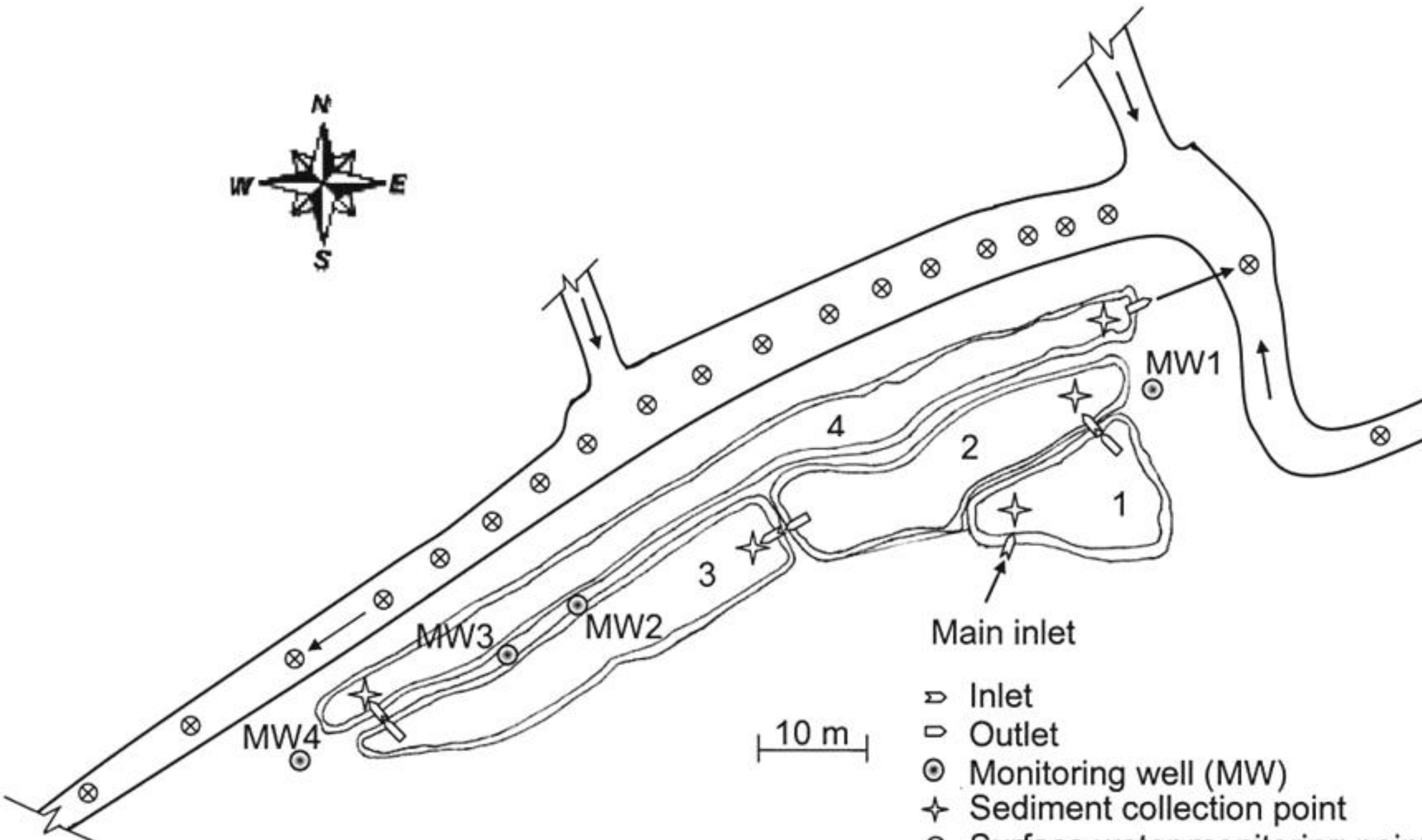
- **Non-point** sources of **water pollution** from anthropogenic sources such as those arising from intensive **farming** systems are a major environmental concern worldwide.
- Wastewaters from agricultural activities have high concentrations of **nutrients** and organic matter.
- **Constructed wetland** systems are low cost and an effective treatment technology to control the diffuse nature of agricultural wastewater pollution sources.

Objectives

- To compare the **community composition** and **diversity of microorganisms** involved in nitrogen transformation from the **litter** and **sediments**.
- To assess a full-scale representative **ICW** system treating **ammonia-rich** wastewater.
- To **relate** differences in **diversity** to nitrogen **removal efficiencies**.
- To study differences in community composition have been analysed in the context of an ecological theory of **community assembly**.

Materials and Methods

Study Site



Materials and Methods

Water Treatment

- **Grab samples** for each wetland cell **inlet** and **outlet** were taken at an approximately **fortnightly** basis.
- Samples were analysed for pH, temperature, dissolved oxygen, biochemical oxygen demand, chemical oxygen demand, **ammonia-nitrogen**, **nitrate-nitrogen**, molybdate reactive phosphorus (soluble reactive phosphorus and *Escherichia coli*.

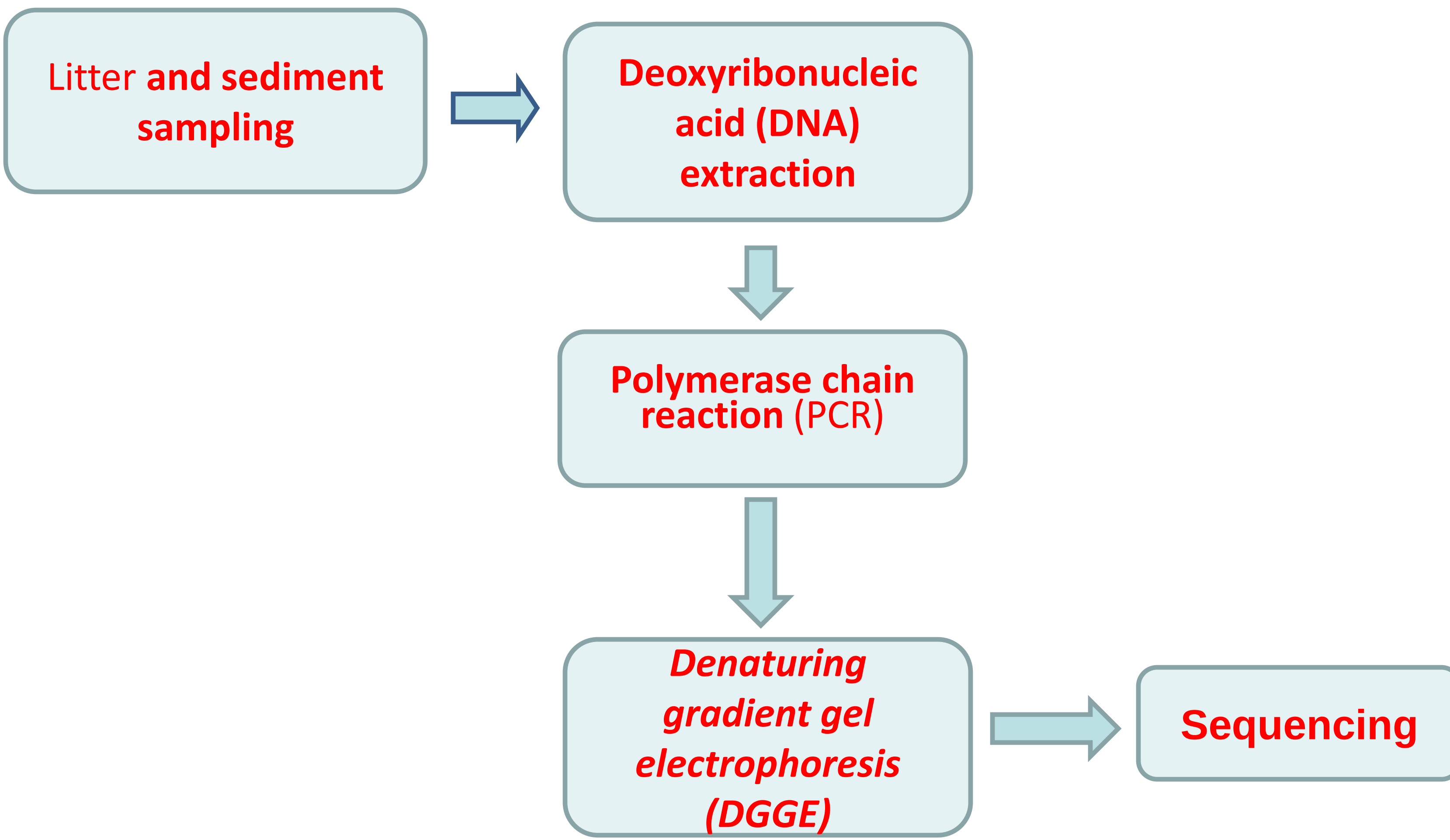
Materials and Methods

Molecular Toolbox

- Molecular methods were employed to study **ammonia-oxidisers** and putative **denitrifiers** in the wetland environment.
- **Polymerase chain reaction** (PCR) based methods were used for the nitrogen removing bacteria community analysis.

Materials and Methods

Molecular Toolbox



Materials and Methods

Sample Collection

- **Duplicate** field litter and sediment **samples** were collected from each wetland cell of the ICW system.
- For each sampling location, all **buried litter** in an area of **0.2 m^2** was collected.
- Sediment samples were collected from the same area with a **sediment sampler** (**diameter** of **4 cm**).
- The **upper 3 cm** of sediment located below the sediment-water interface were used for **analysis**.

Materials and Methods

Sample Collection

- The **samples** were collected near the **influent** point of each cell with an additional sample at the **outlet** of the last cell.
- All samples were **frozen immediately** after collection.
- Transported to the University of Newcastle for subsequent molecular microbiological analysis.

Materials and Methods

DNA Extraction

- The duplicate sediment and litter samples were subjected to deoxyribonucleic acid (**DNA**) **extraction**.

- Application of the **FastDNA® SPIN kit for Soil** (MP Biomedical Inc., USA) according to the manufacturer's protocol.

Materials and Methods

PCR and Agarose Gel Electrophoresis

- **Polymerase chain reaction** is a method to **multiply DNA segments** by repeating cycles of high and low temperature to separate DNA strands and to synthesize new strands.
- **Agarose gel electrophoresis** is a method to **separate DNA molecules by size**.

Materials and Methods

PCR and Agarose Gel Electrophoresis

- The **ammonia-oxidising bacterial** community was assessed using **primers** (Kowalchuk et al.1997).
- The **denitrifying bacterial** community was assessed using **functional gene primers** (Throback et al., 2004).

Materials and Methods

Denaturing Gradient Gel Electrophoresis (DGGE)

- **DGGE** is a **molecular fingerprinting** method that **separates PCR-generated DNA** products.
- DGGE analyses were employed for the **separation of double-stranded DNA** fragments that are **identical in length**, but **differ in sequence**.
- **Polyacrylamide gels** (120×120×1 mm) were prepared with a denaturing gradient.

Materials and Methods

Denaturing Gradient Gel Electrophoresis (DGGE)

- The composition of **100% denaturant** was defined as 7M urea and 40% (vol/vol) formamide (Muyzer et al., 1993).
- The **gels were polymerised** with 15 µL of TEMED and 150 µL of ammonium persulphate.

Materials and Methods

Sequencing

- The **cleaned PCR products** were then **sequenced**.
- The **sequences** were then **BLAST analysed**.
- The internationally recognized NCBI BLAST (www.ncbi.nih.gov) was used to **find closely related sequences** available in the public databases.

Materials and Methods

Analysis of Denaturing Gradient Gel Electrophoresis

- The **presence and intensity of bands** in the DGGE gels were analysed using **Bionumerics**.
- Comparisons of the **similarities** in community **composition** between different environmental sources (**leaves** or **sediments**) and **wetland cells** were conducted.
- Use of **non-metric multidimensional scaling** in combination with **cluster analysis** and **analysis of similarities**.
- Application of **Primer 6 for Windows**.

Materials and Methods

- The **Bray-Curtis algorithm** was used to calculate pairwise **similarities** between samples using **presence-absence data**.
- The **Shannon-Weaver diversity index** *H* (applied on taxa abundance data; i.e. band intensity), **band richness** *S* and **Pielou's evenness** *J'* were calculated.
- These Parameters were used to **compare the diversity** of the predominant communities detected by DGGE.

Materials and Methods

- The **nature of the assembly of the bacterial communities** within the ICW was **evaluated** using the Simberloff method, which is incorporated in to the **Raup and Crick similarity index** of the PAlaeontological Statistics (PAST) program.

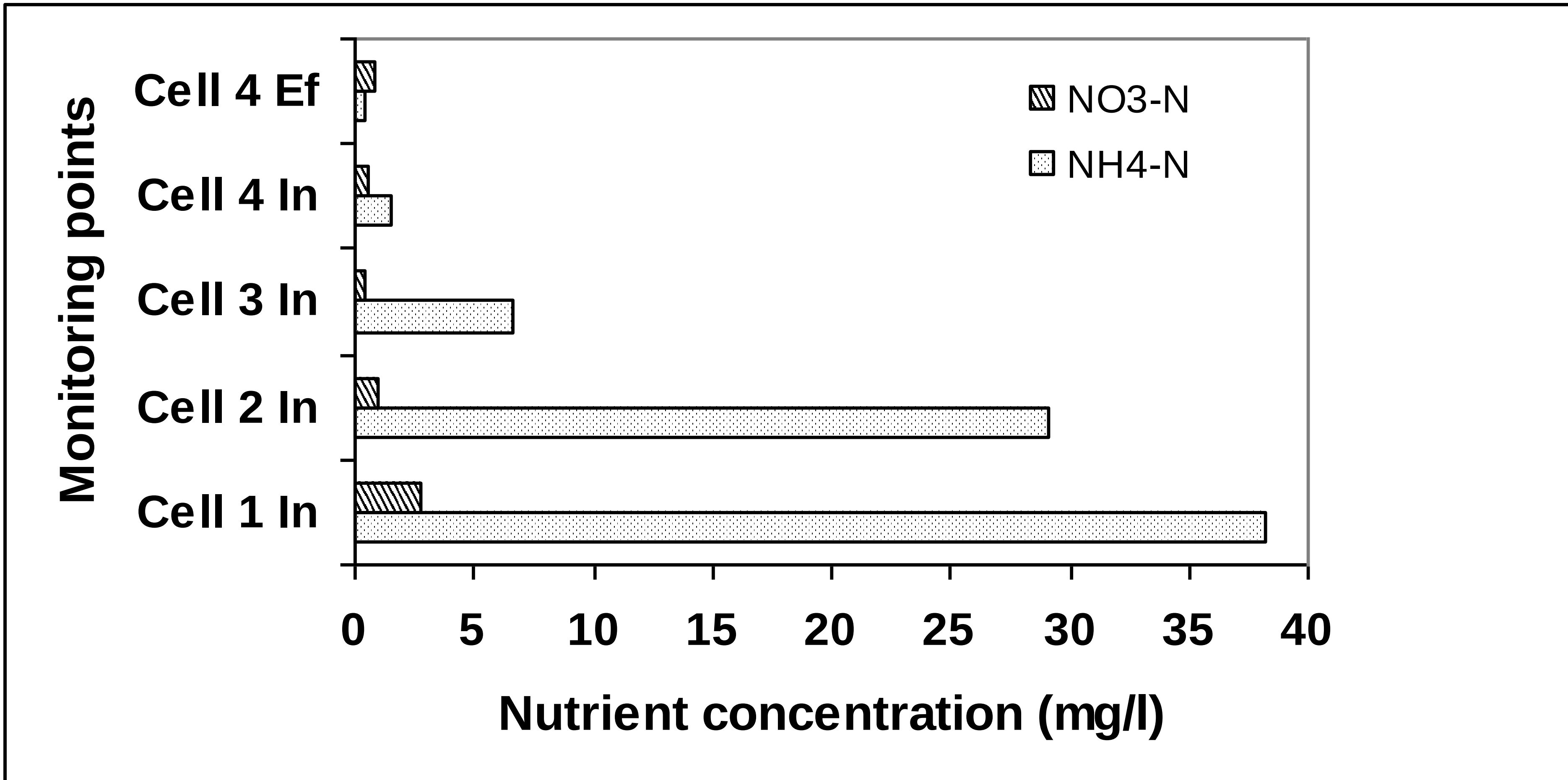
Results and Discussion

Treatment Performance

Parameter	ICW 11		
	Inlet	Outlet	RR %
Temperature (ºC)	13.8	14.9	-
pH	8.12	7.37	-
Electrical Cond. (µS)	1469	373	-
SS (mg/l)	78.4	15.3	80.5
BOD_5 (mg/l)	593.1	5.8	99.0
COD (mg/l)	1341.5	50.4	96.2
NH_4 –N (mg/l)	28.60	0.39	98.6
NO_3 –N (mg/l)	2.60	0.83	68.0
MRP (mg/l)	8.13	0.83	89.8

Results and Discussion

Nitrogen Removal Potential

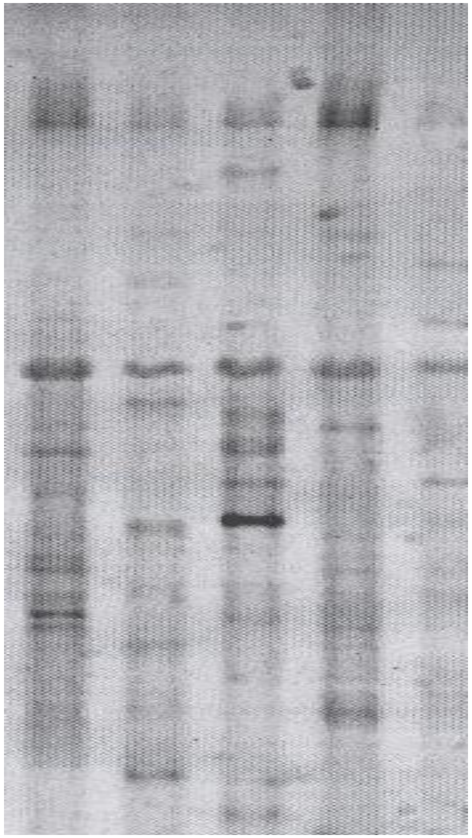


Nutrient Reductions in Selected ICW Cells
(In, influent; Ef, effluent)

Results and Discussion

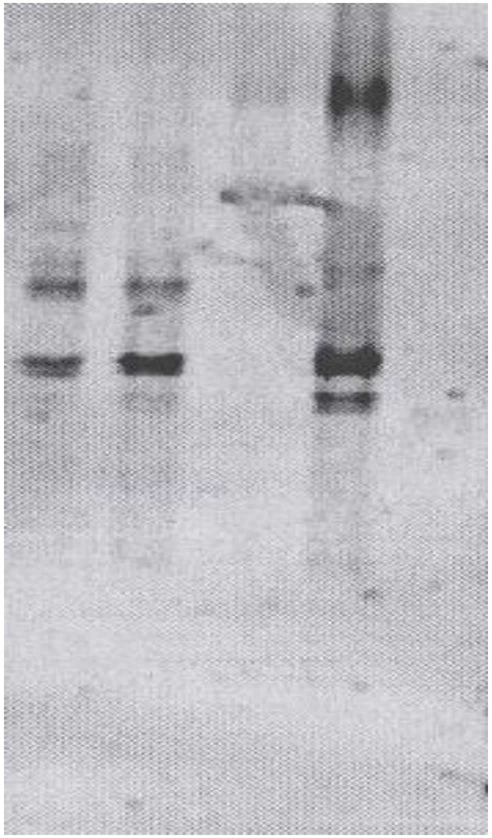
DGGE Profiles of PCR Products

Denitrifying bacteria



C1 C2 C2 C3 C4

Ammonia oxidising bacteria



CI C1 C2 C3 C4

Results and Discussion

Community Analysis

Sequence	ICW 11										Accession number	% Similarity	Strain
	C1 In		C2 In		C3 In		C4 In		C4 Ef				
	L	S	L	S	L	S	L	S	L	S			
C1			+		+						AY123811	97	Nitrosomonas sp. Nm59
C2	+	+			+			+			AY123801	99	Nitrosospira sp. Nsp12
C3	+				+						AY727031	100	Nitrosospira sp. En271
C4	+				+						AY792265	98	Uncultured beta proteobacterium clone

L= Litter S= Sediment

Results and Discussion

Community Analysis

Sequence	ICW 11										Accession number	% Similarity	Strain
	C1 In		C2 In		C3 In		C4 In		C4 Ef				
	L	S	L	S	L	S	L	S	L	S			
nirK													
C8	+		+								EU448024	81	Uncultured denitrifying bacterium clone T23_D5 nitrite reductase (nirK) gene
C9	+		+								EU448024	81	Uncultured denitrifying bacterium clone T23_D5 nitrite reductase (nirK) gene
C10	+		+								FM209186	86	Pseudomonas aeruginosa LESB58 complete genome sequence
C11			+								AY345247	78	Pseudomonas aeruginosa strain DN24 copper-dependent nitrite reductase
C12										+	EF623501	100	Uncultured bacterium clone LK22mK-28 nitrite reductase (nirK) gene
C13		+						+			AM230857	77	Paracoccus sp. R-26824 nirK gene for nitrite reductase
C14								+			DQ783326	96	Uncultured bacterium clone T1R2_0-7cm_038 NirK (nirK) gene
C16		+				+					AM419485	89	Uncultured organism partial nirK gene for putative copper containing dissimilatory nitrite reductase, clone Fin28
C18	+										EF615316	86	Uncultured bacterium clone P1m_nirK-33 nitrite reductase (nirK) gene
C19	+										DQ337794	87	Uncultured bacterium clone S12m_nirK-33 NirK (nirK) gene
C21	+		+		+				+		AM230832	82	Rhizobium sp. R-24663 nirK gene for nitrite reductase
C22			+								DQ337762	89	Uncultured bacterium clone P7m_nirK-25 NirK-lke (nirK) gene
C23	+		+		+						DQ304404	88	Uncultured bacterium clone Ag100-6 putative nitrite reductase (nirK) gene
nirS													
C24											AY078267	85	Thauera terpenica strain 21Mol putative dissimilatory nitrite reductase (nirS) gene,
C25	+		+		+		+				AM230919	90	Dechloromonas sp. R-28451 nirS gene for nitrite reductase
C26	+		+								AM230913	84	Dechloromonas sp. R-28400 nirS gene for nitrite reductase

Results and Discussion

Diversity

Diversity indices for the ammonia-oxidising and denitrifying bacterial communities in sediment and litter of the ICW system (mean ± SD)

Primer/ Genes	Component	Shannon's Index (H)
CTO (Ammonia-oxidisers)	Litter	0.68 ± 0.80
	Sediment	n.d.
*nir*K (Denitrifiers)	Litter	2.04 ± 0.29
	Sediment	0.89 ± 0.80
*nir*S (Denitrifiers)	Litter	2.31 ± 0.18
	Sediment	1.60 ± 0.68

n.d. no data

Results and Discussion

Bacterial Community Characterisation

- Putative **ammonia-oxidising bacteria** were **detected** by PCR in **all wetland cells except** for the **last cell** of the ICW, and at **more locations** in **leaf litter than** in **sediments**.
- **Denitrifiers** were **detected** in **all samples**.
- Analysis of putative **ammonia-oxidising bacteria** and **denitrifier** community fingerprints showed that there were **significant** (ANOSIM; $p < 0.05$) **dissimilarities between communities**.

Results and Discussion

Microbial Community Assembly

- A comparison of microbial communities between the wetland cells suggested that the **ammonia-oxidising bacteria** and **denitrifying bacterial populations** were **no more similar or dissimilar than if** they had been **assembled by chance**.
- Most of the **Raup and Crick indices** were between **0.05 and 0.95**.

Conclusions

- For **ammonia-oxidizing bacteria**, both ***Nitrosospira*** and ***Nitrosomonas*** were detected in the studied wetland system.
- Concerning **denitrifying bacteria**, ***Paracoccus***, ***Pseudomonas***, ***Rhizobium*** and ***Dechloromonas*** were identified.
- The **litter** component of the studied wetland system **supported more diverse nitrogen removing bacteria** (ammonia-oxidising and denitrifying) **than** the **sediments**.

Conclusions

- **Nitrogen removing bacteria** communities in the full-scale wetland system were **no more similar or dissimilar than** would be expected **by chance alone**.
- This suggests that stochastic assembly processes may be important in shaping these communities.

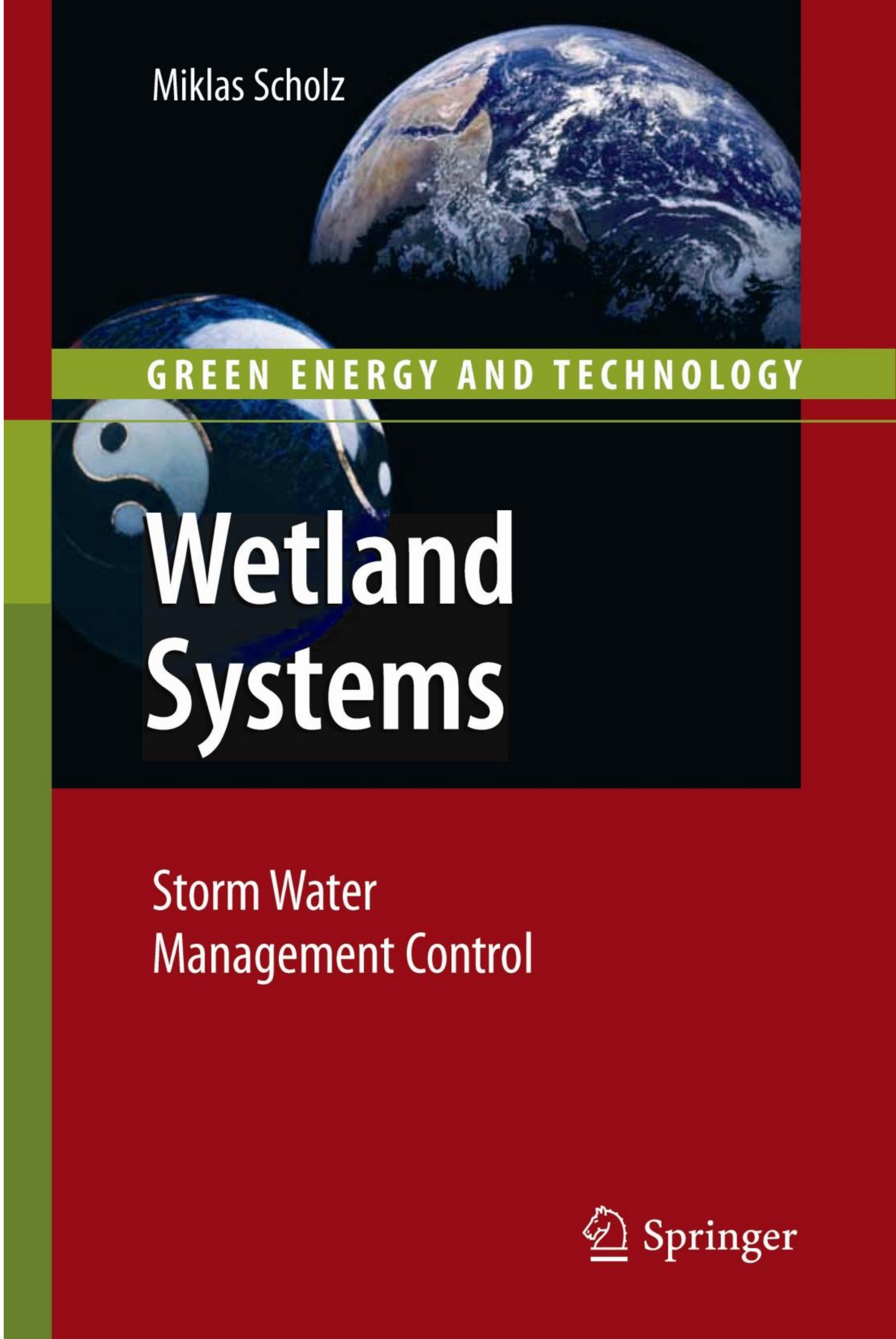
Acknowledgements

- The **European Regional Development Fund Interreg IVB 2007-2013** funded this research project.
- The **Chinese Scholarship Council** and The University of Edinburgh Joint Scholarship supports Ms. Qinli Yang's PhD study.
- **University of Munich**, Germany.
- **Co-workers**: Chris Sagar, Louise Blackhall, Mark Keane, Rodney Moody and Michelle Robinson.

Acknowledgements

- **Paul Carroll** and **Susan Cook**, Waterford County Council, Ireland.
- **Andy Gray** and **John Norman**, University of Edinburgh.
- Department of Environment, Heritage and Local Government, Ireland.
- The University of Edinburgh Development Trust.

Further Reading



Thank you for your attention!