Sorption of sulfamethoxazole, sulfachloropyridazine and sulfamethazine onto six New Zealand dairy farm soils

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Abstract
We have investigated the sorption potential of three sulfonamides (SAs) in six New Zealand dairy farming soils using a modified batch equilibrium method employing 0.005 M $CaCl_2$ as background solution. Both liquid and solid phases were extracted to analyse for the antibiotic concentrations in order to avoid underestimation that may arise a result of photolysis or biotic degradation. The experimental data were later used to construct Freundlich isotherms to determine the effective distribution coefficients. Low log K_{oc} value for all SAs suggests considerable leaching potential for SAs under conditions that are conducive for leaching. The sorption affinity for all soils followed the trend SCP>SMZ>SMO.

Key Words
Partitioning coefficient, sulfonamides, sorption

Introduction
Veterinary antibiotics (VAs) are used in large amounts for therapeutic, and prophylactic purposes in addition to their application as growth promoters. After administration, however, the majority of the antibiotics (up to 80%) are excreted unchanged or as their metabolites (Tolls, 2001). New Zealand has a rapidly expanding dairy industry, and well established beef, sheep, and pig and poultry production the livestock population excretes about 40 times more waste than the human population (Sarmah *et al.* 2006). With increases in the intensive use of antibiotics in NZ agriculture and direct land application of waste, there is concern that excreted compounds will migrate to the receiving environment with potential impact on surface and groundwater. Furthermore frequent use of antibiotics can also give rise to antibiotic resistant microbial populations, thereby creating a heightened concern among regulatory bodies and industries. Sulfonamides are a common class of antibiotic that are widely used in livestock operation, and because of their ionic nature they have the ability to leach through the soil and into the groundwater. Currently there are no fate data available for this group of antibiotics under varied pedo-climatic conditions in New Zealand. Therefore the aim of this study was to derive partitioning coefficients of three selected sulfonamides on six dairy farming soils with contrasting properties.

Methods
Soil
Six top soils (0-5 cm) representative of the dairy farming areas of New Zealand were collected fresh, air-dried, sieved (2mm), and physio-chemical properties determined (Table 1).

Table 1. Selected properties of soils used in the study.

Soils	pH 1:2.5 $CaCl_2$	OC (%)	CEC ($cmol_c$/kg)	Sand %	Silt %	Clay %	SSA (m^2g^{-1})
Matawhero silt loam	6.1	2.1	30	11	62	27	---
Te Kowhai silt loam	5.1	5	21.7	9	54	37	19.7
Hamilton clay loam	5.1	4	17.2	19	51	30	22.3
Horotiu silt loam	5.4	8.2	25	34	48	17	19.7
Manawatu sandy loam	5.4	3.3	9.7	87	11	2	13.6
Gibsons sandy loam	5.2	1.1	7.6	45	41	14	---

Chemicals

Sulfamethoxazole (SMO), Sulfachloropyridazine (SCP) and Sulfamethazine (SMZ) of >99% purity, and calcium chloride dihydrate ($CaCl_2 \cdot 2H_2O$ >99% purity) were obtained from Sigma Aldrich Chemical Company, St. Louis, MO. Acetonitrile, methanol (Chrom AR® HPLC) and dichloromethane (DCM) were obtained from Mallinckrodt.
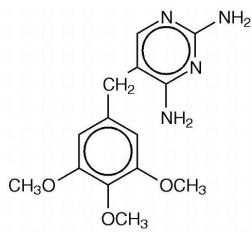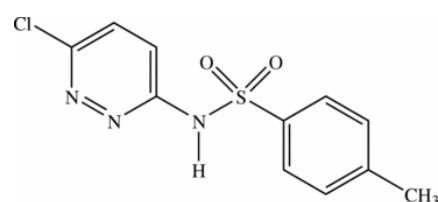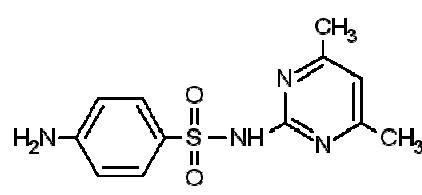
Sulfamethoxazole (SMO)　　　Sulfachloropyridazine (SCP)　　　Sulfamethazine (SMZ)

Figure 1. Molecular structures of antibiotics used in the study.

Table 2. Selected properties of the antibiotics used in the study.

Properties	Sulfamethoxazole	Sulfachloropyridazine	Sulfamethazine
Molecular formula	$C_{10}H_{11}N_3O_3S$	$C_{10}H_9N_4ClO_2S$	$C_{12}H_{14}N_4O_2S$
Molecular weight (g/mol)	253.28	284.72	278.3
Aqueous solubility (mg/l)	3942	8235	1500
Vapour pressure (Pa)	1.74×10^{-5}	1.77×10^{-7}	3.64×10^{-11}
log K_{ow}	0.48	0.31	0. 8
pK_a	6.1	5.7	7.45

Batch sorption experiment

Air-dried soil samples (2 g) were weighed into 35 mL glass centrifuge tubes. Aliquots (30 mL) of SMO, SCP and SMZ prepared separately at 6 initial concentrations (1.5, 3, 5, 7.5, 10 &15 mg/L in 0.005 mol/L) $CaCl_2$ were added to the tubes, which were wrapped in aluminium foil, and shaken (12 h) to equilibrate (based on a kinetic study shown in Figure 2) in the dark (23°C ± 2).
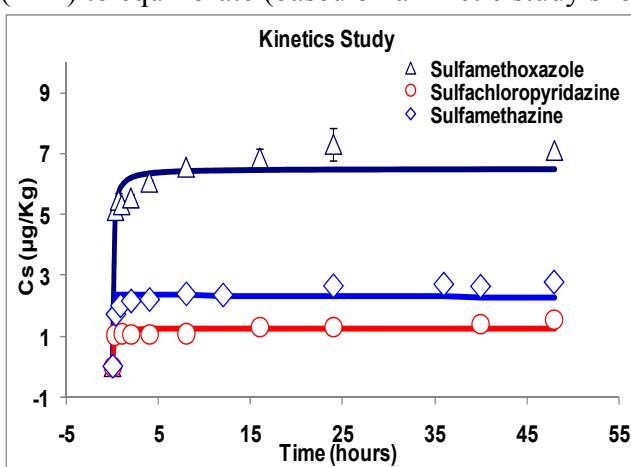

Figure 2. Sorption kinetics
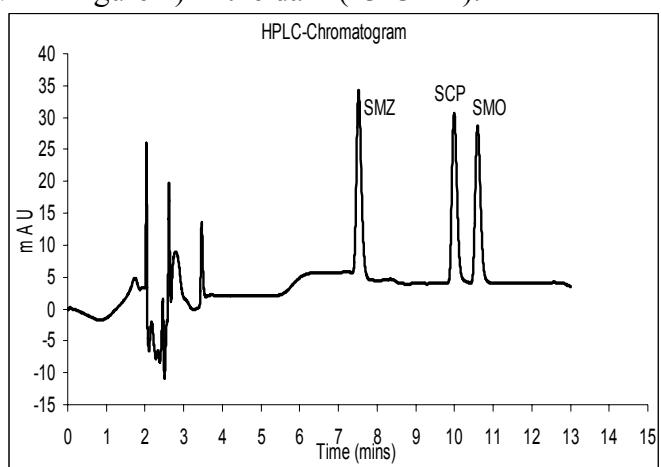
Fig 3. HPLC Chromatogram

Extraction and analysis

Following equilibration, tubes were centrifuged (1750g x 10 min), the supernatants were analysed directly and the residual soils were extracted with DCM (5 mL). An aliquot (1mL) of each solvent extract was evaporated to dryness under N_2, and the residue was reconstituted in methanol (0.5 mL) and analysed by High Performance Liquid Chromatography with detection at 275 nm. Separation was achieved using a C_{18} Luna column (Figure3) eluted isocratically with acetonitrile: trifluoroacetic acid: tetrahydrofuran (40:55:5 for SMO, 32:63:5 for SCP and 31:64:5 for SMZ) at1.0 mL min^{-1} and an injection volume of 20μL.

Results and discussion

The Freundlich model ($C_s = K_f C_w^N$, where C_s is the sorbed concentration in L/kg; C_w is the equilibrium solution concentration in mg/L; K_f is the Freundlich coefficient and N is an exponent which determines the degree of non-linearity) could describe the isotherms constructed by extraction scheme in all soils much better than the isotherms produced using data from a difference scheme, and hence parameters were derived using only data obtained from extraction technique and results are discussed only for the extraction scheme (Table 3). Extraction resulted in acceptable recoveries (90-95%) for all the 3 compounds in the 6 soils. The degree of isotherm linearity (*N*) for SCP and SMZ varied between 0.87–1.11 in the six soils. SMO showed a

highly non linear pattern (N=0.75) in just one soil (Manawatu). For SMO, Te Kowhai gave a linear isotherm ($N = 1$). K_d^{eff} values for the three antibiotics (at C_w = 1.5 mg/L) in the soils ranged from 0.37–4.6 L/kg. Matawhero soil (OC=2.1%) gave the smallest log K_{oc} for all the 3 compounds. In general, sorption of sulfonamides was influenced by hydrophobic interactions and sorption increased with increases in OC content, except for Gibsons sandy loam soil. Average log K_{oc} value for all sulfonamides between 1.21-2.38 log units suggested moderate to high leaching potential for SAs under conditions conducive for leaching.
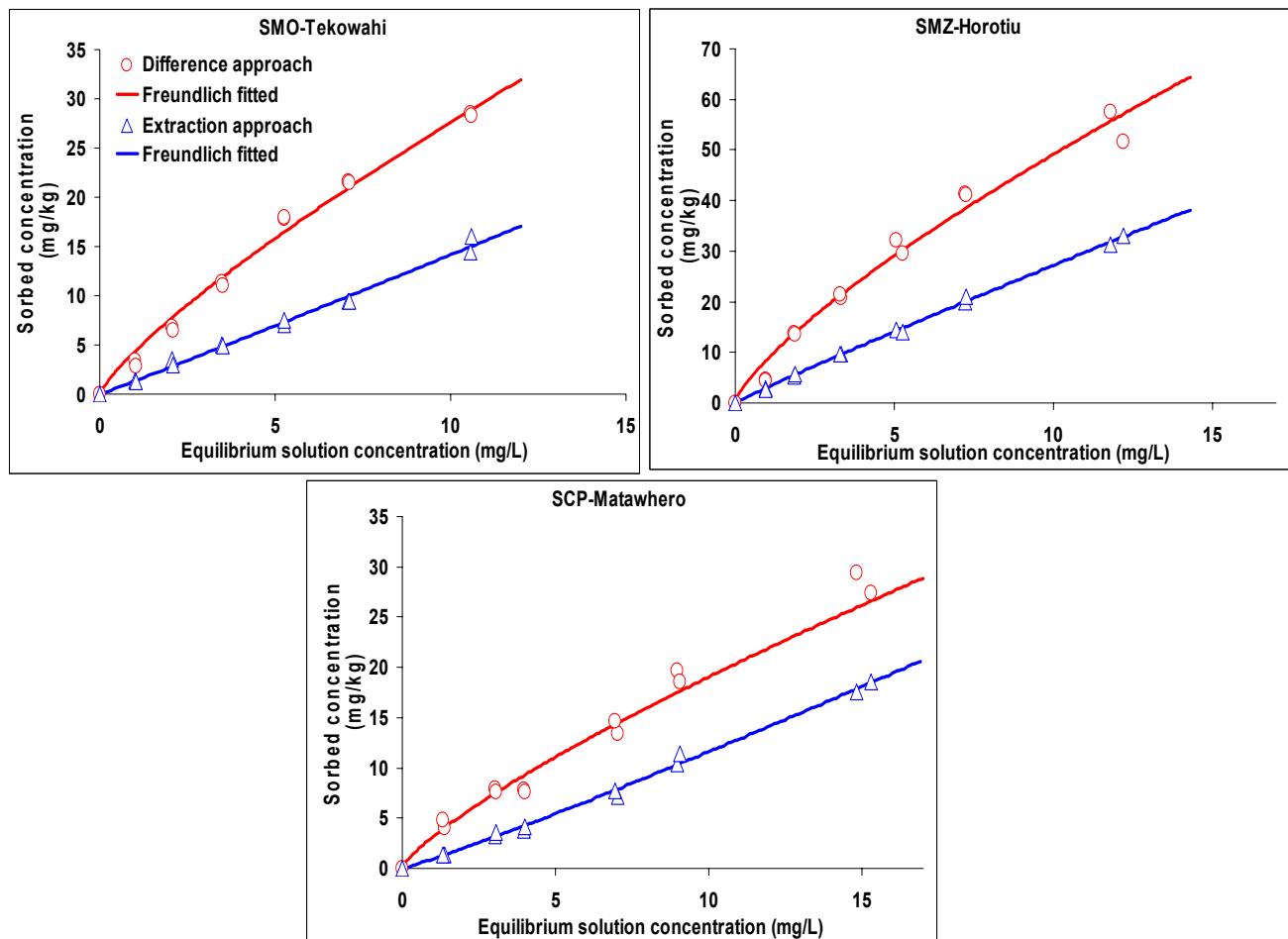

Figure 4. Example of multiple-concentration batch sorption isotherms for SMO, SCP and SMZ using difference and solvent extraction schemes. Symbols represent measured data, while solid lines represent Freundlich model fits.

Conclusion

We have demonstrated that the technique of employing DCM to extract the solid phase provides good recoveries, and values are more robust than those obtained through a difference scheme when used to estimate sorption parameters. Overall sorption affinity for all compounds in six soils followed an order: SCP > SMZ >SMO. Based on their partitioning coefficient values, it can be concluded that these compounds may potentially be a risk to the groundwater under conditions conducive for leaching such as high rainfall. Sorption of these compounds is also likely to be pH dependent, and the compounds can behave like weak acids. Therefore much work is warranted to understand their sorption behaviour under realistic field situation especially in soils amended with manure, as manure pH can be higher than normal field soils.

References

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Table 3. Summary of sorption parameters derived from the multiple-concentration isotherms constructed using solvent extraction scheme in selected soils.

Soils	Sulfamethoxazole					Sulfachloropyridazine					Sulfamethazine				
	K_d^{eff}	K_f	N	R^2	Log K_{oc}	K_d^{eff}	K_f	N	R^2	Log K_{oc}	K_d^{eff}	K_f	N	R^2	Log K_{oc}
Matawhero silt loam	0.37	0.34	1.21	0.99	1.21	1	0.97	1.08	0.99	1.66	0.82	0.82	0.98	0.97	1.59
Te Kowhai silt loam	1.36	1.35	1.02	0.99	1.43	4.6	4.54	1.03	0.99	1.96	1.14	1.13	1.01	0.96	1.35
Hamilton clay loam	1.9	2	0.88	0.99	1.7	4.57	4.74	0.91	1	2.07	2.88	2.91	0.98	1	1.86
Horotiu silt loam	2.4	2.51	0.9	1	1.49	2.63	2.52	1.11	0.99	1.49	2.12	2.19	0.91	0.99	1.43
Manawatu sandy	1.36	1.51	0.75	0.98	1.87	4.31	4.55	0.87	0.99	2.14	0.77	0.88	0.98	1.44	0.77
Gibsons sandy loam	0.77	0.81	0.86	0.98	1.66	2.62	2.61	1.01	1	2.38	0.27	1.27	1	1.35	0.27

K_d^{eff} is the concentration dependent effective sorption distribution coefficient ($K_d^{eff} = K_f C_w^{N-1}$) using lowest aqueous equilibrium solution concentrations of $C_w = 1.5$ mg/l; concentration K_{oc} is the OC-normalized sorption coefficient calculated using $K_{oc} = K_f C_w^{N-1} foc^{-1}$ at $C_w = 0.5$ mg/l; K_d^{eff} in l/kg and K_f in mg^{1-N} l^N/kg