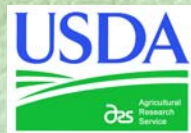


US EPA ARCHIVE DOCUMENT

Comparison of Batch and Flow Experimental Data on Retention of Manure-borne *Cryptosporidium parvum* oocysts in soils

Yakov Pachepsky and Daniel Shelton

**USDA-Agricultural Research Service
Environmental Microbial Safety Laboratory, Beltsville, MD**



LAYOUT

- 1. Background**
- 2. Batch experiments with two soils**
- 3. Column experiments**
with saturated and unsaturated soils
- 4. Parameter estimation and comparison**
- 5. Conclusions**

Background

- Dairy/beef animals are a major source of *C. parvum* oocysts
- Most transport/retention studies have been conducted with purified oocysts in distilled water or oocysts in calf diarrhea.
- Manure is a complex matrix and can affect oocyst attachment to soils in several different ways.
- Understanding of oocyst transport under field conditions requires information on oocyst-soil-manure interactions.

Batch experiments

1. Dairy manure was diluted to obtain 1% and 10% suspensions.
2. Manure suspensions were inoculated with purified *C. parvum* oocysts to have concentrations ca. 10^5 oocysts/mL.
3. Nine ml of sandy loam and clay loam soil 1% suspensions were mixed 1 ml with manure suspensions, vortexed thoroughly, and incubated for 2 h with a gentle shaking.
4. Suspensions were centrifuged for 10 min at 100 g, the top 9 mL of supernatant was removed using a glass pipette and 9 ml of distilled water added to tubes. The Step 3 above was repeated.
5. Oocysts were enumerated as previously described by Kuczynska and Shelton (1999).

Two replications were conducted independently with different bovine manures from Beltsville, MD.

Data inspection showed that the data for two manures are essentially identical.

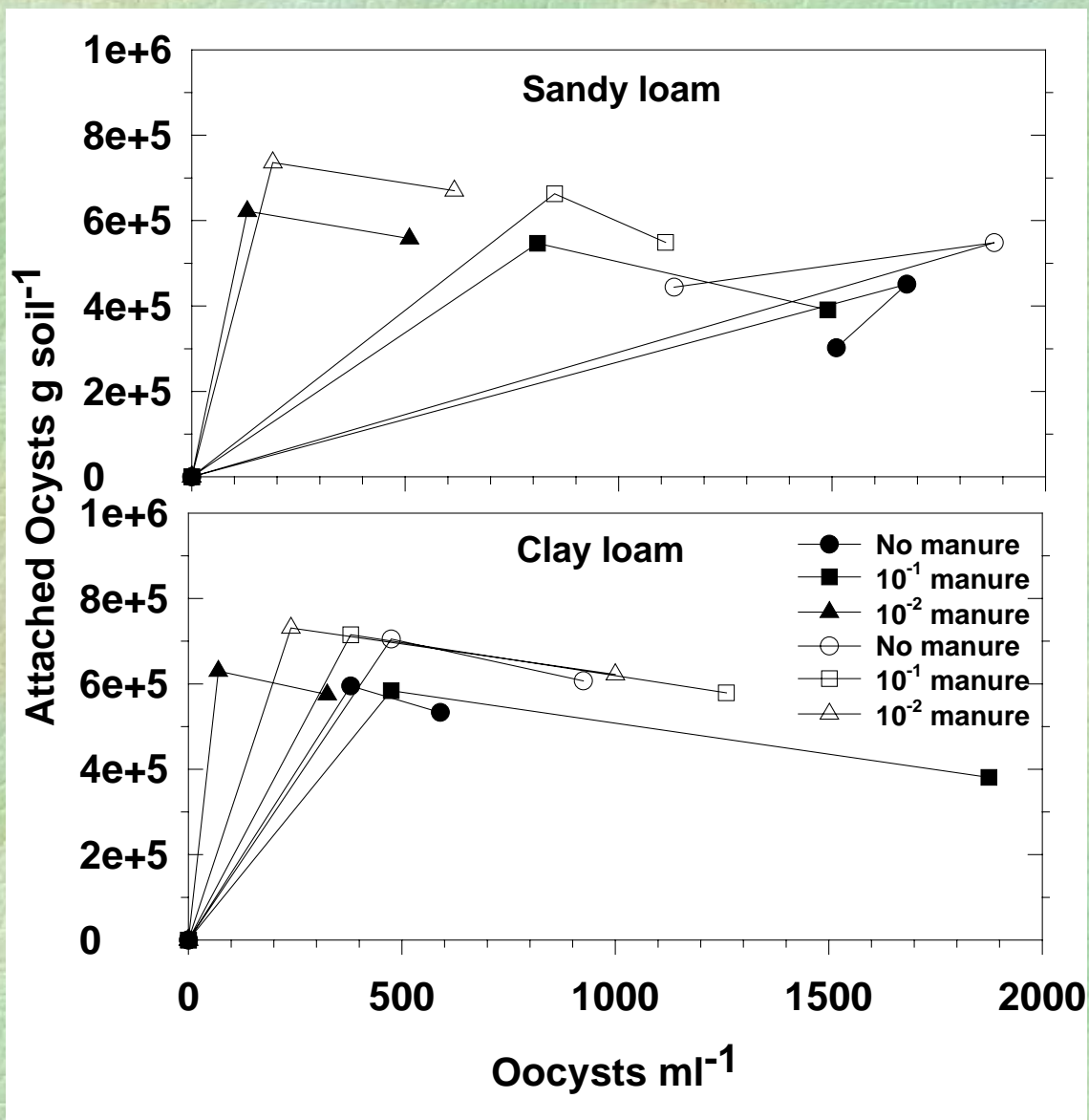
ANOVA supported this conclusion; the manure concentration and the soil texture were significant factors at $P < 0.001$

Percentage of oocysts attached to soil particles

<u>Treatment</u>	<u>Sandy Loam (K_d)</u>	<u>Clay Loam (K_d)</u>
No manure	72.0±3.6 ¹ (280)	93.1±2.9 (1520)
1.0% manure	86.7±2.2 (730)	93.1±3.8 (1360)
0.1% manure	97.4±1.8 (4300)	97.7±2.0 (6000)

¹Mean and S.D.

Oocyst attachment to soil particles in presence and in absence of manure



Column experiments

Columns 10 cm long and 11.5 cm in diameter were packed with air-dried and sieved clay loam and sandy loam soil to 50% porosity.

Columns were slowly saturated from the bottom.

50 grams of bovine manure, seeded with 7.1×10^6 oocysts, were applied to the surface of soil columns and covered with straw.

Rainfall was applied at ca. 1.6 cm/h for 8 hours.

Saturated flow and unsaturated flow at 5 kPa suction were created for each soil.

Leachate was collected at one-hour intervals.

At the end of the experiment, the manure on the surface of cores was collected and cores were sliced into five 2 cm sections.

Leachate and soil samples were processed as previously described (Kuczynska and Shelton, 1999).

Data analysis

$$\frac{\partial}{\partial t}(\theta_a c + \rho s) = \frac{\partial}{\partial x} \left(\theta_a D \frac{\partial c}{\partial x} - J_w c \right) - \mu_l \theta_a c - \rho \mu_s s$$

$$s = K_d c$$

$$R \frac{\partial c}{\partial t} = \frac{\theta_a}{\theta} D \frac{\partial^2 c}{\partial x^2} - v \frac{\partial c}{\partial x} - \mu c$$

$$R = \frac{\theta_a}{\theta} + \frac{\rho K_d}{\theta}$$

$$\mu = \mu_l \frac{\theta_a}{\theta} + \frac{\rho K_d \mu_s}{\theta}$$

$$c_m(t) = c_0 \exp(-\lambda t)$$

θ_a = the porosity available for oocyst transport

ρ = soil bulk density

s = the adsorbed amount of oocyst,

D = the dispersion coefficient

J_w = the volumetric water flux density

μ_l and μ_s = first-order removal rate constants

K_d = the distribution constant

v = the average pore-water velocity

R = the retardation factor

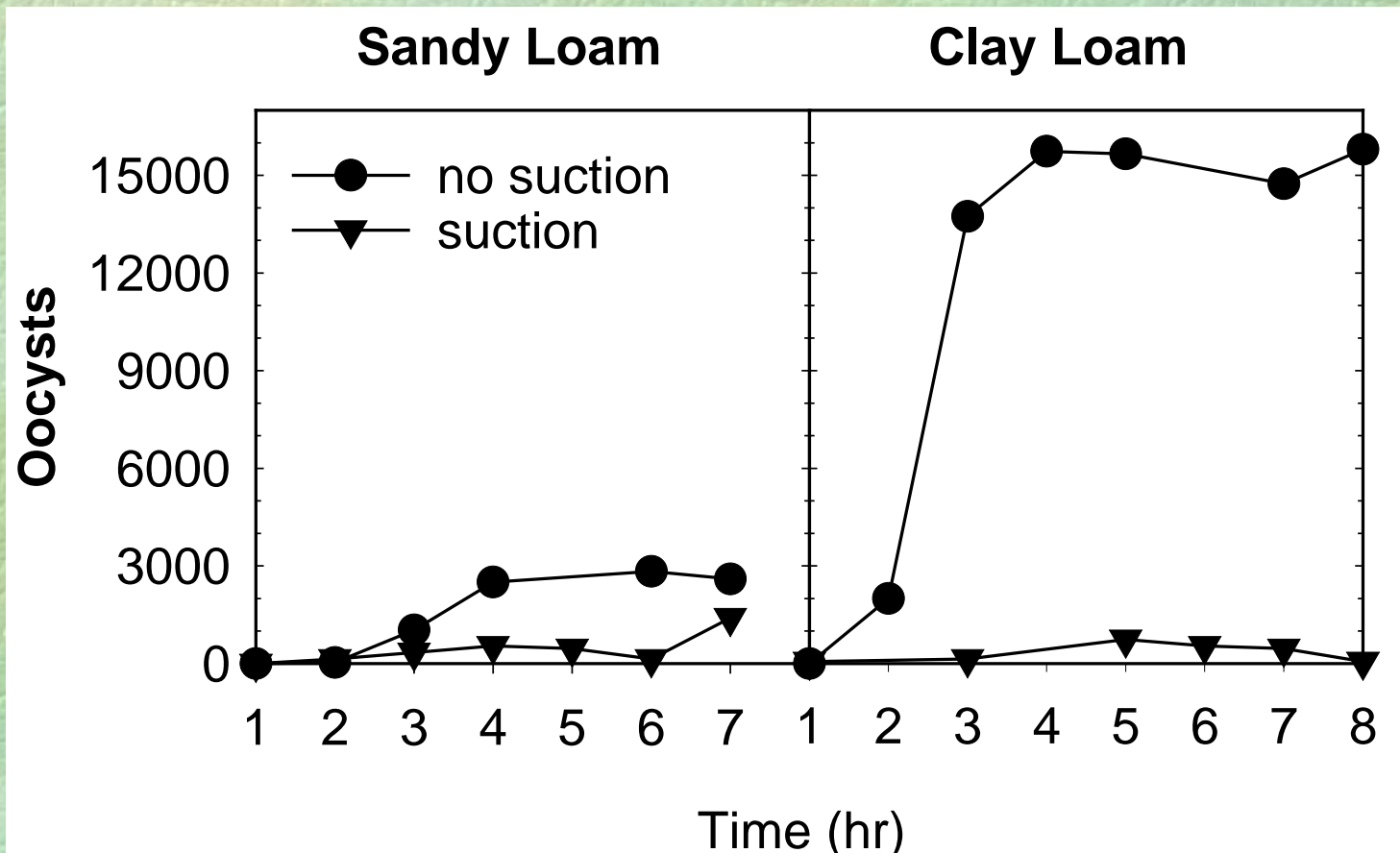
μ = the combined first-order removal rate constant

c_m = the concentration of oocyst released from manure

Parameter estimation

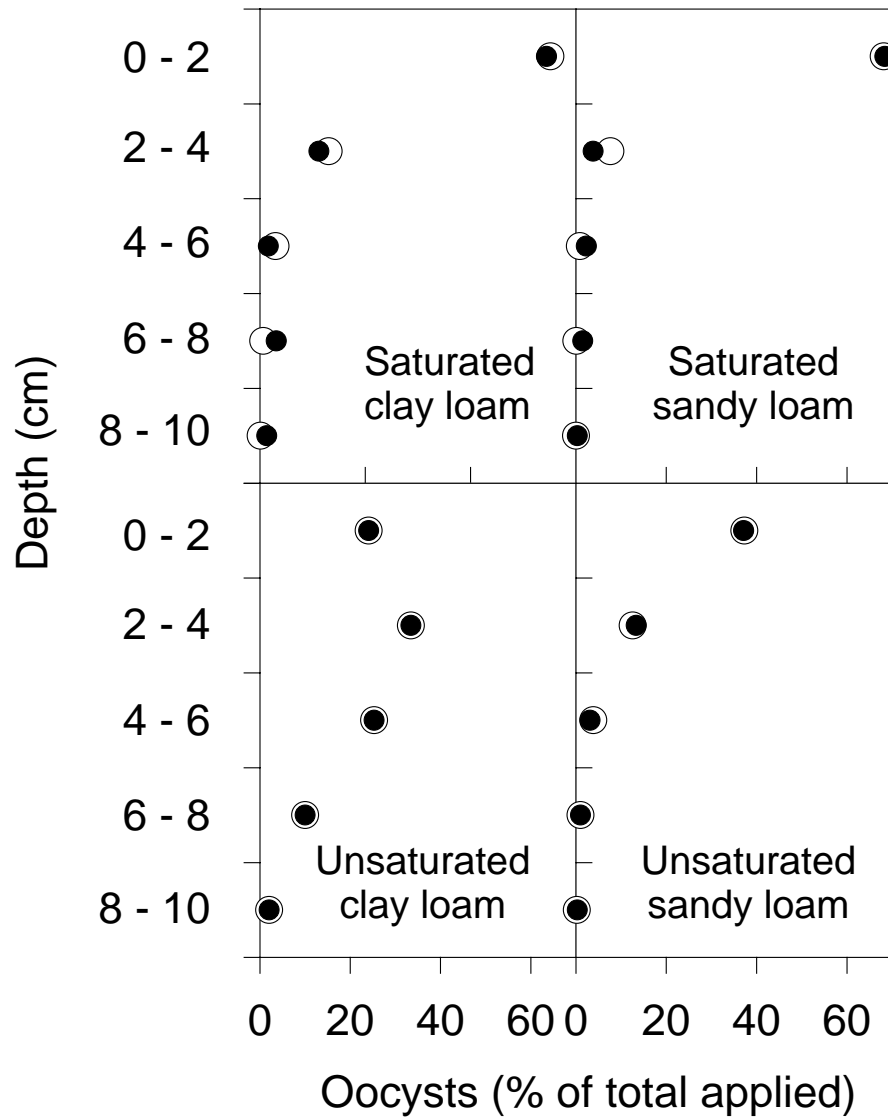
1. Values of θ and ν were measured.
2. Values of λ were computed from the percentage of oocysts left on the soil surface.
3. The value of D was computed as $D=lv$ with $l=1$ cm and $l=0.5$ cm in saturated and unsaturated samples, respectively.
4. The analytical solution (van Genuchten, 1985) was fitted to data on profile oocyst distributions and cumulative amount of oocysts in leachate to estimate R and μ

Oocyst leaching

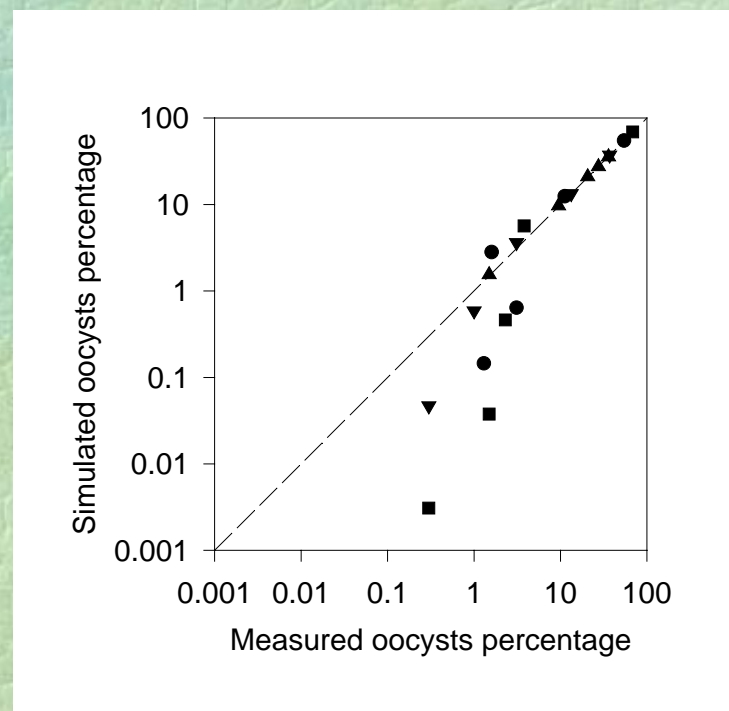


Cumulative leaching (%)

	Clay loam	Sandy loam
Saturated	1.3	0.4
Unsaturated	<0.1	<0.1



Measured (●) and simulated (○) oocyst profile distributions



Transport parameters

Parameter	Saturated columns		Unsaturated columns	
	Clay loam	Sandy loam	Clay loam	Sandy loam
ν (cm h ⁻¹)	2.7	2.6	3.3	1.5
D (cm ² h ⁻¹)	2.7	2.6	1.7	0.7
λ (h ⁻¹)	0.212	0.503	0.240	0.134
μ^{\dagger} (h ⁻¹)	2.77 - 4.13	5.28 - 9.96	0.00 - 0.16	0.62 - 0.81
R^{\dagger}	0.00 - 0.97	0.00 - 1.56	4.51 - 4.61	2.44 - 3.82

[†]Confidence intervals of the optimized values at the 5% significance level

Release of manure colloids: $\lambda=0.12-0.18$ h⁻¹ (Bradford and Schijven, 2002),
 $\lambda=0.32$ h⁻¹ (Shelton et al., 2002).

Oocyst removal rates: 2 to 30 h⁻¹ at $\nu=6 - 60$ cm h⁻¹ (Harter et al., 2000, sands),
 1500 to 3000 h⁻¹ at $\nu= 40,000$ cm h⁻¹ (Brush et al., 1999, glass beads)

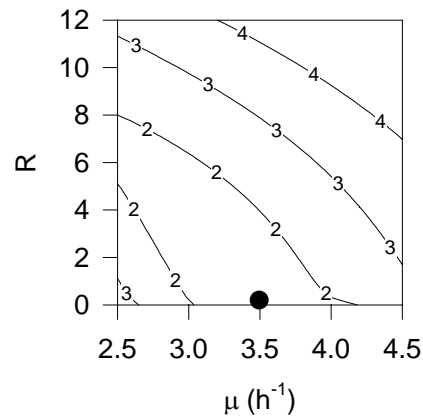
Removal rates and retardation coefficients are markedly different in saturated and unsaturated columns.

$K_d = 1.4 -7.7$ from column data, K_d between 100 and 700 in batch experiments

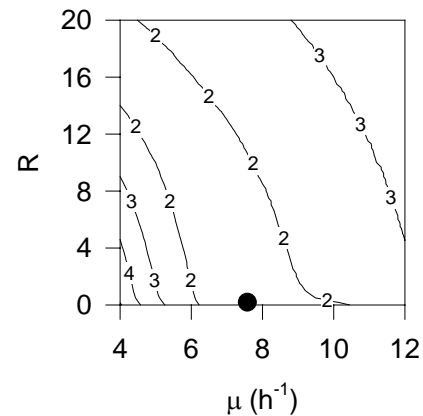
Challenge for parameter estimation

The same overall model error can be achieved with various combinations of R and μ .

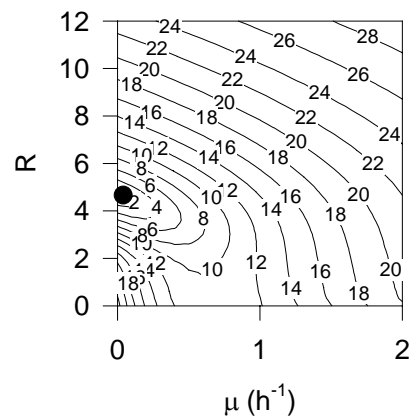
Saturated clay loam



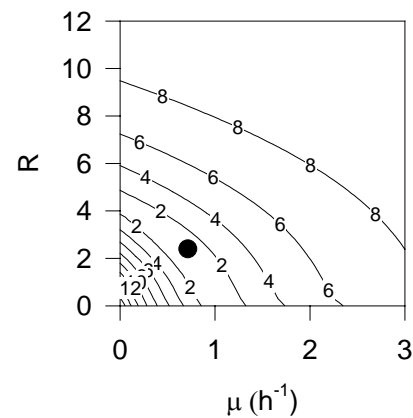
Saturated sandy loam



Unsaturated clay loam



Unsaturated sandy loam



Conclusions

Presence of manure markedly affects attachment of *C. parvum* oocysts to soil.

Soil texture is a substantial factor of oocyst attachment in batch experiments; soil structure may be more important for retention during transport

The breakthrough of oocysts released with manure particulates was higher in saturated than in unsaturated soil

The convective-dispersion model simulated the profile distributions of oocysts reasonably well, but failed to simulate breakthrough of small amounts of oocysts occurring probably due to a preferential flow

Values of retardation coefficient were most probably less than unity in saturated columns and greater than unity in unsaturated columns.

The removal rates were much higher in saturated columns as compared with unsaturated columns.

Oocyst adsorption data from batch experiments may be inapplicable in transport models that employ the adsorption equilibrium hypothesis.