

Supplementary materials

Decay Rate of *Escherichia coli* in a Mountainous Tropical Headwater Wetland

Paty Nakhle ¹, Laurie Boithias ^{1,*}, Anne Pando-Bahuon ², Chanthamousone Thammahacksa ², Nicolas Gallion ¹, Phabvilay Sounyafong ², Norbert Silvera ², Keooudone Latsachack ², Bounsamay Soulileuth ², Emma J. Rochelle-Newall ³, Yoan Marcangeli ³, Alain Pierret ², and Olivier Ribolzi ¹

¹ Géosciences Environnement Toulouse (GET), Université de Toulouse, CNRS, IRD, UPS, Toulouse, France; paty.nakhle@get.omp.eu (P.N.); gallion.nicolas@gmail.com (N.G.); olivier.ribolzi@get.omp.eu (O.R.)

² Institut de Recherche pour le Développement (IRD), IEES-Paris UMR 242, Sorbonne Université, c/o Department of Agricultural Land Management (DALaM), Vientiane, Lao PDR; anne.pando@ird.fr (A.P.B.); louyird@gmail.com (C.T.); phabvilay_laopdr@yahoo.com (P.S.); norbert.silvera@ird.fr (N.S.); wannaki@gmail.com (K.L.); sbounsamay@gmail.com (B.S.); alain.pierret@ird.fr (A.P.)

³ Institute of Ecology and Environmental Sciences of Paris (iEES-Paris), Sorbonne Université, Univ Paris Est Creteil, IRD, CNRS, INRA, France; emma.rochelle-newall@ird.fr (E.J.R.N.); yoan.marcangeli@sorbonne-universite.fr (Y.M.)

* Corresponding author: laurie.boithias@get.omp.eu

1. Supplementary information: calculation of *E. coli* stock variations
2. Supplementary figures

1. Supplementary information: calculation of *E. coli* stock variations

1.1 Estimate of bacterial decay due to solar radiation and to particle deposition

We applied a balance method to estimate the respective contributions of solar radiation and particle deposition to bacterial apparent decay during the experiment. In this approach, initial and final stocks of *E. coli* in the water column were compared between the treatments DL and DD, and sediment resuspension from the bottom deposit of the mesocosms was thus not considered. The control treatment was used to verify the absence of atmospheric deposits, which were considered to be negligible and were therefore also excluded. Only solar radiation-related decay and solid particle deposition were assumed to be involved in this balance. After a period of time (Δt) after the start of the experiment, the total number of decayed *E. coli* in the water column (N_t) was calculated as follows:

$$N_t = n_t^L + n_t^D \quad (S1)$$

where n_t^L and n_t^D are the number of bacteria decayed due to solar radiation and to particle deposition, respectively. Considering that treatment DL cumulates the decay effects of both solar radiation and solid particle deposition, N_t was defined as the difference between the *E. coli* stock at the start of the experiment (S_o) and after Δt for treatment DL (S_t^{DL}):

$$N_t = S_o - S_t^{DL} \quad (S2)$$

S_o was calculated by multiplying a normalized initial concentration ($C_o = 500,000$ MPN 100 mL⁻¹) by the volume of water at the beginning of the experiment ($V_o = 4,500$ mL), while S_t^{DL} was estimated by multiplying the remaining volume of water (V_t^{DL}) by the final *E. coli* concentration (C_f^{DL}) in treatment DL. C_f^{DL} was obtained using equation (1) considering the fitted value of the apparent decay rate (Table 1) for treatment DL (k^{DL}). As a result, equation (S2) can be written:

$$N_t = C_o (V_o - V_t^{DL} e^{-k^{DL} t}) \quad (S3)$$

Similarly, n_t^D was deduced from the difference between S_o and *E. coli* stock after Δt for treatment DD (S_t^{DD}):

$$n_t^D = S_o - S_t^{DD} \quad (S4)$$

S_t^{DD} was calculated by multiplying the remaining volume of water (V_t^{DD}) by the final *E. coli* concentration (C_f^{DD}) of treatment DD, and C_f^{DD} was obtained using equation (1) considering the fitted value of the apparent decay rate (Table 1) for treatment DD (k^{DD}). As a result, equation (S4) can be written:

$$n_t^D = C_o (V_o - V_t^{DD} e^{-k^{DD} t}) \quad (S5)$$

n_t^L was then obtained from the combination of equations (S1), (S3) and (S5):

$$n_t^L = C_o (V_t^{DD} e^{k^{DD} t} - V_t^{DL} e^{k^{DL} t}) \quad (S6)$$

Finally, we deduced the respective fractional contributions (expressed in percentage) of solar radiation (F_L) and solid particle deposition (F_D) in the total bacterial decay as follows:

$$F_L = \frac{n_t^L}{N_t} 100 \quad (S7)$$

$$F_D = 100 - F_L \quad (S8)$$

1.2 Uncertainty estimation of bacterial decay due to solar radiation and particle deposition

We used a Monte Carlo approach to quantify the uncertainty in n_t^D and n_t^L calculations. With the exception of t and C_o , each known parameter of equations (S5) and (S6) was assigned an estimated expectation and an estimated standard deviation. The fitted values (Figure S3) were assigned to k^{DD} and k^{DL} as mathematical expectation, whereas their respective errors were used as standard deviations (see Table 1). For V_o , the expectation corresponded to the initial volume using a 1-litre measuring cylinder, whereas the standard deviation corresponded to the cumulated inaccuracy of the measurement. Finally, for V_t^{DD} and V_t^{DL} , at each sampling time of the experiment, the mathematical expectation is considered to be equivalent to the theoretical residual volume obtained after subtracting from V_o the successive volumes of sampled water and a volume of evaporated water. An average 45% accuracy of the residual volume was used to estimate the standard deviation.

For each parameter, and at each sampling time, 10,000 random samples were taken in accordance with the normal distribution defined by the estimated expectation and the estimated standard deviation. At each random draw, n_t^D and n_t^L were calculated using equations (S5) and (S6). The 10,000 values obtained for each decay number were used to calculate the statistic descriptive parameters (i.e., mean, median, first and third quartiles and standard deviation, minimum and maximum) of n_t^D and n_t^L shown in Figure 5.

2. Supplementary figures

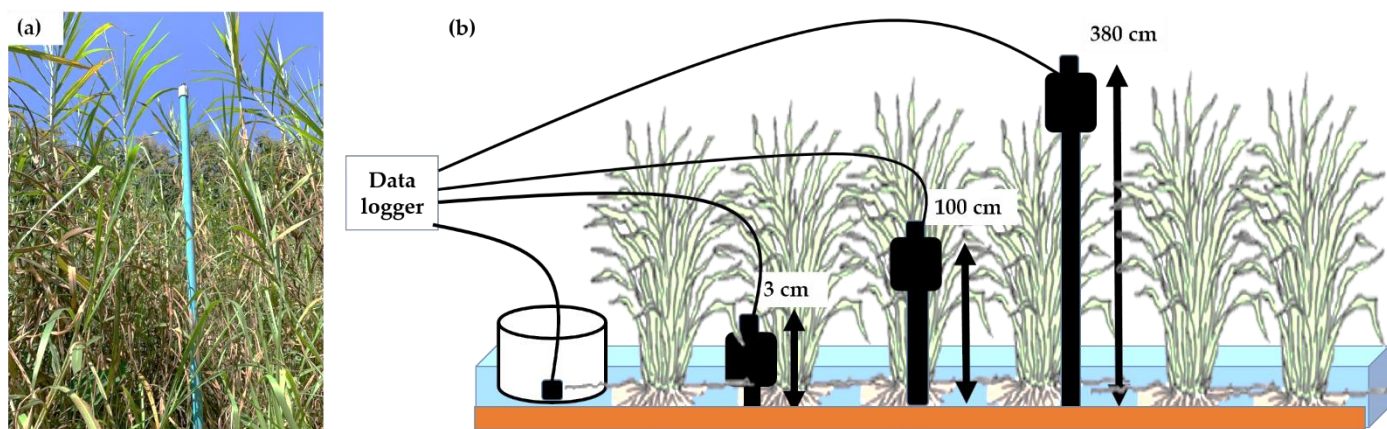


Figure S1. (a) Photo of the Napier grass and solar sensor installed in the wetland of Houay Pano catchment, northern Lao PDR; (b) diagram of the solar sensors installed at different heights (380 cm, 100 cm, 3 cm, and inside the mesocosm) to measure the solar radiation attenuation by Napier grass during two days (24 and 25 October 2020) in the wetland of the Houay Pano catchment. The pyranometers used to measure solar radiation at 380 cm height: SP110 (Campbell CS300); at 100 cm height: RG100 Solems; at 3 cm height: Li200X (LI-COR PY34392); and inside covered top mesocosm: RG100 Solems.

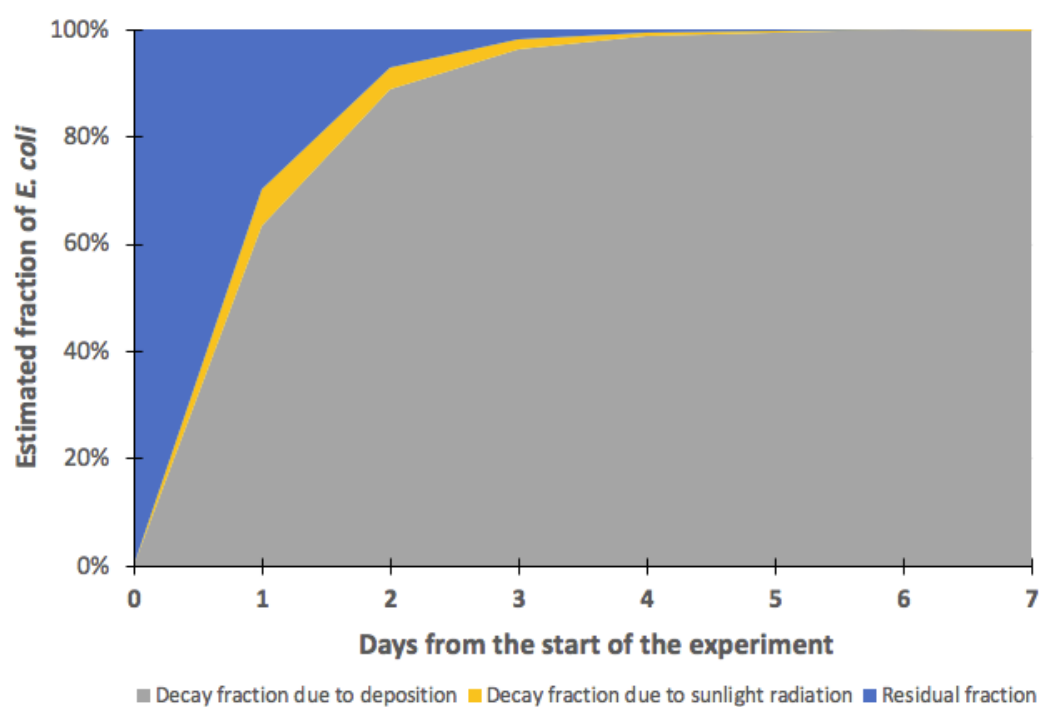


Figure S2. Stacked area graph showing the average percentage of the estimated fraction of decayed *E. coli* at daily time steps during the experiment: the grey area corresponds to the decay fraction due to particle deposition, the yellow area corresponds to the decay fraction due to solar radiation, and the blue area is the residual fraction of decayed *E. coli*.

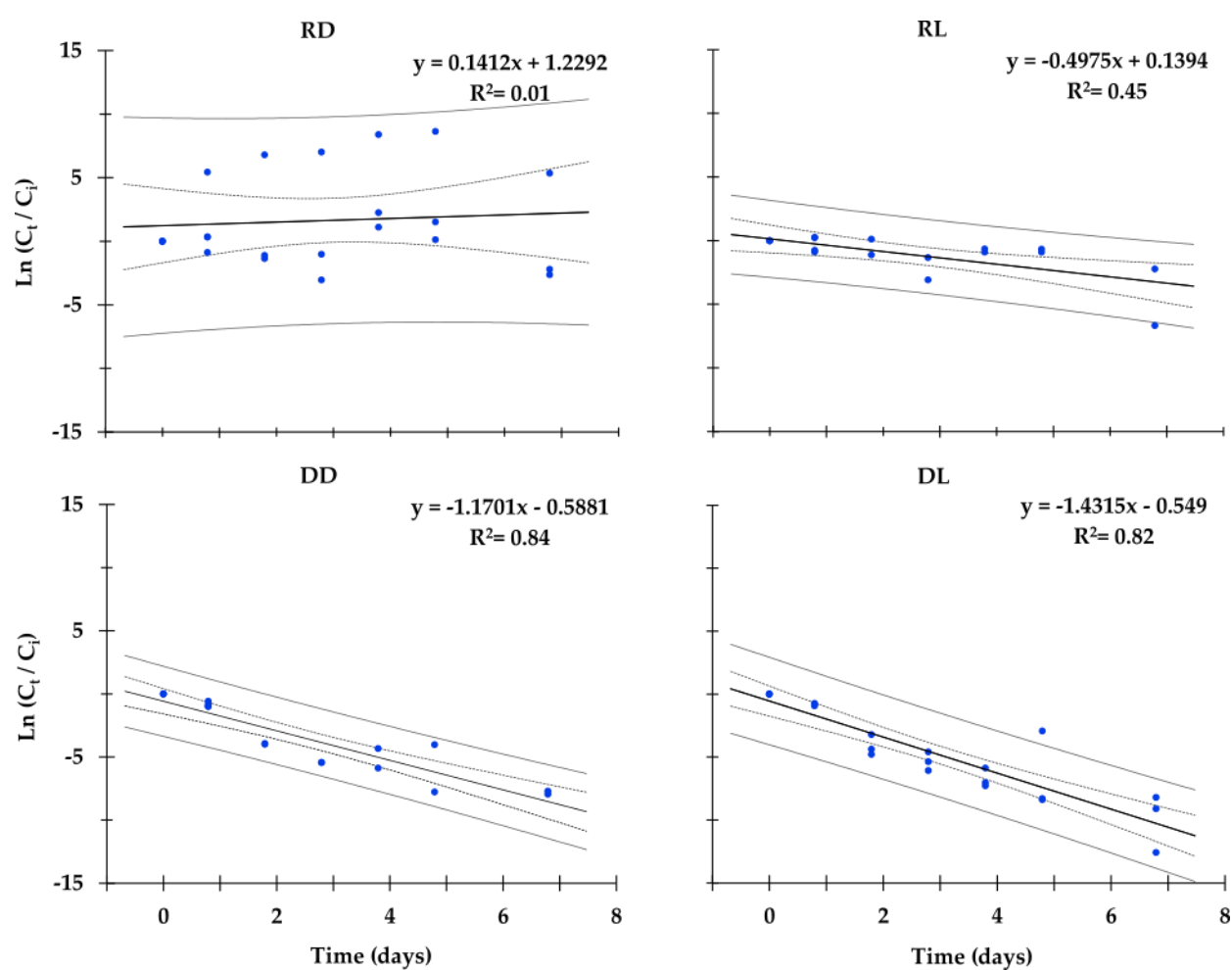


Figure S3. Plot of $\ln(C_t/C_i)$ versus time in days where C_t is the measured concentration of total *E. coli* at time t in MPN 100 mL⁻¹, C_i is the measured initial concentration of total *E. coli* in MPN 100 mL⁻¹ for the mesocosms installed in the headwater wetland of the Houay Pano catchment, northern Lao PDR, from August 9 to August 16, 2019. RD: Resuspension-Dark; RL: Resuspension-Light; DD: Deposition-Dark; DL: Deposition-Light. Dotted lines represent 95% confidence interval and continuous lines represent 95% prediction interval.