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1 **Use of fallout radionuclides (^7Be , ^{210}Pb) to estimate**
2 **resuspension of *Escherichia coli* from streambed sediments**
3 **during floods in a tropical montane catchment**

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21

1 **Abstract** Consumption of water polluted by faecal contaminants is responsible for 2 million deaths
2 annually, most of which occur in developing countries without adequate sanitation. In tropical aquatic
3 systems, streambeds can be reservoirs of persistent pathogenic bacteria and high rainfall can lead to
4 contaminated soils entering streams and to the resuspension of sediment-bound microbes in the
5 streambed. Here we present a novel method using fallout radionuclides (^7Be and $^{210}\text{Pb}_{\text{xs}}$) to estimate the
6 proportions of *Escherichia coli*, an indicator of faecal contamination, associated with recently eroded
7 soil particles and with the resuspension of streambed sediments (Fig. 1). We show that using these
8 radionuclides and hydrograph separations we are able to characterize the proportion of particles
9 originating from highly contaminated soils and that from the resuspension of particle-attached bacteria
10 within the streambed. We also found that although overland flow represented just over one-tenth of the
11 total flood volume, it was responsible for more than two-thirds of the downstream transfer of *E. coli*.
12 We propose that data obtained using this method can be used to understand the dynamics of FIB
13 (Faecal Indicator Bacteria) in streams thereby providing information for adapted management plans
14 that reduce the health risks to local populations.

15 **Keywords:** FIB, South East Asia, Hydrological modelling, erosion, land use, overland flow, beryllium-
16 7, lead-210.

17 **Introduction**

18 It has been estimated that almost 2 million people die annually as a consequence of waterborne
19 diarrheal diseases caused by the use of untreated water. The majority of the victims are children
20 between 0 to 5yr old living in developing countries (WHO 2012). The rural areas of South East Asia
21 (SEA) are particularly affected because many of the poorest households living in the montane agro-
22 ecosystems use contaminated river water for basic domestic needs such as washing, bathing and
23 cooking (Ribolzi et al. 2011a, Rochelle-Newall et al. 2015, Saqalli et al. 2015). Moreover, in areas
24 where proper sanitation infrastructures are lacking, open area human defecation and the use of shallow
25 latrines is frequent (Guiteras et al. 2015, Sturma and Becerra 2012). Furthermore, in the absence of
26 animal husbandry practices that protect streams through the installation of fences, the soil surface in
27 rural areas of SEA is often contaminated with microbial faecal pollutants of human and animal origins
28 (WorldBank 2009). Indeed, in tropical and temperate systems, land use is known to play an important

1 role in determining the degree of faecal contamination in a watershed (Ekklesia et al. 2015, Goto and
2 Yan 2011).

3 The presence of faecal contamination in soil, sediment and water samples is generally
4 determined using indicator bacteria. These faecal indicator bacteria (FIB) are used because they are
5 much easier to culture and are considered to have a lower health risk than the pathogens for which they
6 are an indicator (Ishii and Sadowsky 2008). Of the FIB generally used, *Escherichia coli* is often cited
7 as being a good indicator of faecal contamination although some authors have questioned the relevance
8 of its use in tropical systems (Byappanahalli and Fujioka 1998, Byappanahalli et al. 2012).
9 Nevertheless, most of the methods used to determine the presence of *E. coli* are easy to employ,
10 relatively cheap, and do not require complex analytical equipment and are therefore adapted for use in
11 sites without access to high grade laboratory facilities.

12 Bacteria in aquatic systems can be either free-living or attached to particles and the relative
13 percentages of each fraction vary as a function of turbidity and particle quality (Lemee et al. 2002,
14 Suter et al. 2011). Probably as a consequence of their intestinal origin where they are attached to
15 digested food particles, FIB are mostly particle-bound although the actual percentage associated with
16 particles does vary with TSS concentrations (Byappanahalli et al. 2012, Nguyen et al. in press, Oliver
17 et al. 2007). During storm events characterised by high solid loads (stormflow loading), FIB numbers
18 are often higher than during the inter-storm, low flow periods characterised by lower particle loads
19 (Causse et al. 2015, Chan et al. 2015, Hrdinka et al. 2012, Stumpf et al. 2010). During periods of low
20 flow, the fraction of particle-bound FIB can be deposited and stored in the river channel and stream
21 sediments to form an in-channel reservoir of FIB that can be mobilized during floods (e.g. Pachepsky
22 et al. 2011, Pandey et al. 2012).

23 There is a paucity of knowledge on the distribution and survival of FIB in tropical freshwater
24 environments (Rochelle-Newall et al. 2015). In these ecosystems higher and more stable temperatures,
25 greater reserves of nutrients and organic carbon and more stable humidity conditions may lead to
26 higher bacterial survival rates, particularly in sediments where FIB are protected from light and
27 predators (Garzio-Hadzick et al. 2010, Pachepsky and Shelton 2011). Given the risks for human health
28 associated with the consumption of contaminated water, quantifying the proportion of FIB resuspended
29 with sediments during floods is crucial in tropical catchments where informal water sources often

1 supply the majority of domestic requirements. Providing solid estimates of this resuspension is
2 essential for the validation of model simulations on FIB transport and survival (Cho et al. 2012, Kim et
3 al. 2010, Pandey et al. 2012).

4 Particle-bound FIB that are transported in streams during floods come from two main sources:
5 (1) recently eroded particles from the surface layer of soils on hillslopes and the riparian zone that are
6 supplied to the river by overland flow and (2) resuspension of streambed sediments. However,
7 estimating the relative proportions of these two sources to sediment transport during floods is a
8 difficult challenge. To discriminate recently eroded particles and resuspended material, previous
9 studies have tried to directly quantify resuspension by generating artificial floods in the stream channel
10 during dry weather conditions (Muirhead et al. 2004, Yakirevich et al. 2013). However, these
11 experiments are generally conducted on an isolated reach of the stream and the maximum intensity of
12 these artificial floods remains limited. Moreover, they do not reproduce the dilution effect of pre-event
13 groundwater, which is often a major contributor to natural streamflow (Kirchner 2003). Therefore they
14 are unlikely to recreate the real conditions observed during a natural flood event.

15 The objective of this short communication is to estimate the fraction of particle attached *E.*
16 *coli* resuspended from streambed sediments during a natural flood event in a tropical montane system
17 in SEA. An original method based on previous applications using fallout radionuclide measurements
18 (i.e. ^7Be , $^{210}\text{Pb}_{\text{xs}}$) is proposed (Evrard et al. 2010, Matisoff et al. 2005). These radionuclides,
19 characterised by different half-lives (53 days and 22 years, respectively), are supplied to the soils by
20 rainfall. Upon soil contact they are quickly and almost irreversibly bound to fine soil particles (Taylor
21 et al. 2013). These two radionuclides were recently used to quantify the proportions of recently eroded
22 sediment (sediment labelled by short-lived ^7Be) and particles remobilised from the riverbed/channel
23 (depleted in ^7Be) in the study area (Gourdin et al. 2014b). In addition to the quantification of the
24 contribution of both sources (soil erosion and sediment resuspension), this method can be used to
25 estimate the age of resuspended sediments to which particle-bound bacteria are attached.

1 **Material and methods**

2 **Study site**

3 The study site is a catchment of 60 ha located in North Lao PDR (Fig. 2). It is a sub-basin of the Houay
4 Xon system, which is a tributary of the Mekong River. The geological substrate consists of argillites,
5 siltstones and fine-grained sandstones from Permian to Upper Carboniferous (Department of Geology
6 and Mines, 1990/1991). The soils developed on these bedrocks are mainly Entisol, Ultisol and Alfisol
7 (Soil Survey Staff 1999).

8 The terrain is steep with a mean slope gradient of 54%, ranging from 0% to more than 171%.
9 The climate is a wet-dry monsoon tropical climate with an average annual rainfall of ~1300 mm.year⁻¹.
10 The rainfall pattern is highly seasonal with 77% of the rainfall occurring during the rainy season from
11 mid-May to mid-October, whereas the November to March period is cooler and mostly dry.

12 The mean monthly temperature measured at the local meteorological station ranges between
13 12 to 35 °C, with the highest temperatures observed in April just before the onset of the wet season.
14 Mean monthly relative humidity varies between 17 and 100%. The watershed is characterised by the
15 occurrence of about 30 storms during the wet season of which around 20 generate a flood in the stream.
16 During these storms, rainfall reaches intensities that are commonly observed in tropical systems (up to
17 100 mmh⁻¹ over a time step of 6 mins).

18 Land use in the catchment is dominated by cropland (with a short fallow cycle) and teak
19 plantations on the hillslopes (Huon et al. 2013). In the stream zone, small domestic gardens and fish
20 ponds are found. These areas are generally uninhabited during the dry season. However, during the wet
21 season, these zones are more frequently used by farmers and their livestock. During this time the
22 inhabitants construct small semi-permanent camps where open air defecation is practiced. Livestock is
23 of relatively low density and includes pigs, chickens, goats and dogs. Wild animals also live in the
24 fallow areas and can contribute to faecal contamination. There are three swamp zones along the stream
25 length.

26 This catchment can be considered as representative of the montane agro-ecosystems of SEA.
27 Monitored since 1998, this site is part of MSEC (Multi-scale Environmental Changes, <http://msec.obs->

1 mip.fr/) South East Asian agro-hydrological observatory. It belongs to the French network of critical
2 zone observatories (RBV, Réseau de Bassins Versants, <http://rnbv.ipgp.fr/>).

3 **Hydrological measurements and water sampling**

4 Rainwater depth and intensity was measured (with a 6 minute time step) using an automatic rain gauge
5 (Cimel). Stream discharge was measured at the catchment outlet using an automated recording station
6 that consisted of a water level recorder (OTT, Thalimedes/SE200) and a V-notch weir. The control-
7 rating curve was previously determined using cross-sectional metering and slug tests. Stream water was
8 collected in plastic bottles by an automatic sampler (Automatic Pumping Type Sediment Sampler,
9 ICRISAT) installed at the catchment outlet (Fig. 2). During this study, 12 1-liter samples were
10 collected over the flood. Electrical conductivity normalized to 25°C (EC) of each sample was
11 determined with an YSI® 556 probe. The concentration of total suspended solid (TSS) was determined
12 for each sample after evaporation in an oven at 105°C for 48 h. Cumulated TSS exports were
13 calculated by integrating the flux of TSS over the flood.

14 ***Escherichia coli* determinations**

15 The standardized microplate method (ISO 9308-3) was used for *E. coli* load determinations. This
16 method is based on serial dilutions and an evaluation of the most probable number (MPN) using a
17 culture medium specific for *E. coli*. Each sample was incubated at four dilution rates (i.e. 1:2, 1:20,
18 1:200 and 1:2000) in a 96-well microplate (MUG/EC, BIODIAGNOSTICS) and incubated for 36
19 to 48h at 44°C. Ringers' Lactate solution was used for the dilutions and one plate was used per sample.
20 The number of positive wells for each microplate was noted and the MPN was determined using the
21 Poisson distribution. Each sample was incubated within a few hours after the flood. The *E. coli* load at
22 the sediment surface was also determined at four sites along the stream (Fig. 2). At each site, 10 g of
23 sediment were collected from the upper 2 cm of the sediment surface and were resuspended in 11 of
24 Ringers' Lactate solution. The resulting solution was agitated and the *E. coli* load was determined
25 using the microplate method. This microplate method has previously been used with success in the
26 studied ecosystem (Causse et al. 2015).

1 Radionuclide analyses

2 Radionuclide activities were measured in a total of five suspended sediment samples. Samples were
3 packed into 15–60-ml (depending on the quantity available) polyethylene containers and sealed
4 airtight. The ^7Be and ^{210}Pb activities were determined by gamma spectrometry using the very low-
5 background coaxial N- and P-type GeHP detectors at the Laboratoire des Sciences du Climat et de
6 l'Environnement (LSCE; Gif-sur-Yvette, France; (Gourdin et al. 2014b)). All results were expressed in
7 Bq kg^{-1} . Activities were also decay-corrected for the sampling date. Uncertainties on radionuclide
8 activities were ca. 10% for $^{210}\text{Pb}_{\text{xs}}$ and 20% for ^7Be . All samples were analysed within less than 53 days
9 (^7Be half-life) following the rainfall event.

10 EC-based hydrograph separation

11 A tracer-based approach similar to the one described by Ribolzi et al. (1997) and Collins and Neal
12 (1998) was used to separate storm hydrographs into event water, which mainly includes infiltration
13 excess overland flow (Patin et al. 2012, Ribolzi et al. 2011b), and pre-event water, i.e. ground water
14 pre-stored in the catchment (Vigiak et al. 2008). This approach uses a simple mixing model with two
15 reservoirs and the electrical conductivity (EC) of water as a tracer. It is described by the following
16 equations:

$$17 \quad Q_{\text{sw}} = Q_{\text{of}} + Q_{\text{gw}} \quad (1)$$

$$18 \quad Q_{\text{sw}} \text{ EC}_{\text{sw}} = Q_{\text{of}} \text{ EC}_{\text{of}} + Q_{\text{gw}} \text{ EC}_{\text{gw}} \quad (2)$$

19 Where Q_{sw} represents the instantaneous stream water discharge at the catchment outlet, Q_{of} ,
20 the instantaneous discharge of overland flow and Q_{gw} , the instantaneous discharge of groundwater
21 (expressed in l s^{-1}). EC_{sw} is the instantaneous EC measured in the stream. EC_{of} and EC_{gw} are the
22 average values respectively for overland flow and groundwater during the storm event. EC_{of} was
23 measured on samples of overland flow that were collected from the soil surface on hillslopes draining
24 to the stream during the rainfall event (Patin et al. 2012). Since groundwater feeds the stream during
25 inter-stormflow periods (Ribolzi et al. 1997), EC_{gw} was approximated from stream measurements
26 carried out prior to storm event.

1 Solidograph separation and estimate of *E. coli* resuspension

2 Due to their different half-lives (53 days and 22 years respectively), ^7Be and $^{210}\text{Pb}_{\text{xs}}$ are particularly
3 relevant for quantifying the respective contributions of (1) particles detached from the soil surface
4 along hillslopes and the riparian zone (i.e. recently tagged with ^7Be) and (2) resuspension of stream bed
5 sediments (i.e. isolated from recent fallout and depleted in ^7Be). The percentage of “new” suspended
6 particles (P_{new} , recently labelled particles detached from the soil surface) in the total stream load at a
7 given sampling time was estimated following the method of Matisoff et al. (2005):

$$8 \quad P_{\text{new}} = 100 [(A/B) / (A_0/B_0)] \quad (3)$$

9 where A and B are the ^7Be and $^{210}\text{Pb}_{\text{xs}}$ activities (Bq kg^{-1}) of the suspended particles collected from the
10 stream and A_0 and B_0 are the ^7Be and $^{210}\text{Pb}_{\text{xs}}$ activities in rainfall (Bq l^{-1}). The value 5.4 was used to
11 estimate the ratio A_0/B_0 . It was obtained by averaging signature of 4 rain showers that occurred
12 immediately prior to the flood (i.e. 6-April, 23-May, 31-May and 1-Jun 2012). This value is consistent
13 with other work from the same study area (Gourdin et al. 2014a, Gourdin et al. 2014b).

14 The proportion of “old” suspended particles (P_{old} , resuspended streambed sediments isolated
15 from recent fallout) was estimated by the following equations:

$$16 \quad P_{\text{old}} = 100 - P_{\text{new}} \quad (4)$$

$$17 \quad F_{\text{old}} = F_{\text{sw}} P_{\text{old}} / 100 \quad (5)$$

18 Where F_{sw} and F_{old} are, respectively, the total and old suspended solid particle flows (kg s^{-1}) at
19 time t. The flow of *E. coli* from streambed sediment resuspension ($F_{\text{E.coli_old}}$; expressed in MPN s^{-1}),
20 mainly composed of old sediment, was then estimated from:

$$21 \quad F_{\text{E.coli_old}} = F_{\text{old}} \text{SB}_{\text{E.coli}} \quad (6)$$

22 Where $\text{SB}_{\text{E.coli}}$ is the total load of *E. coli* in streambed sediment (MPN kg^{-1}). The flow of *E.*
23 *coli* due to recently labelled particles detached from the soil surface ($F_{\text{E.coli_new}}$; expressed in MPN s^{-1})
24 was then estimated from:

$$25 \quad F_{\text{E.coli_new}} = F_{\text{E.coli_sw}} - F_{\text{E.coli_old}} \quad (7)$$

1 Where $F_{E.coli_{sw}} = F_{sw} SW_{E.coli}$ (8)

2
3 $F_{E.coli_{sw}}$ is the flow of *E. coli* in the stream at time *t* (MPN s⁻¹) and $SW_{E.coli}$ is the
4
5 concentration of *E. coli* in the stream water sample (MPN l⁻¹) at time *t*.
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9 **Results**

10 **Description of the rainfall-runoff event**

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17 The studied flood was triggered by a storm that occurred on May 15th, 2012. It was the first significant
18
19 erosive event of the rainy season. During the storm, maximum rainfall intensity occurred between
20
21 10:30 am and 10:36 am and reached 75 mm h⁻¹ (Fig. 3). The cumulated rainfall over the 48 min rain
22
23 event was 33.5 mm. This corresponds to approximately a one-year return period (34.7 mm daily
24
25 rainfall) according to Bricquet et al. (2003) for the 1950-2000 period. The lag time between the peak of
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27 rainfall and peak discharge was 12 minutes. This storm generated a stormflow of approximately 273
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29 min during which discharge peaked at 90 L s⁻¹ at 10:48. This corresponds to a maximum specific
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31 discharge of 1.5 L s⁻¹ ha⁻¹. The runoff coefficient for this event (depth of streamflow/depth of rain) was
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33 3%.
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36 **Variations in the stream flow composition**

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16 The initial EC was 533 $\mu\text{S cm}^{-1}$, characteristic of groundwater (Fig. 3). This then rapidly decreased to
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18 358 $\mu\text{S cm}^{-1}$ during the peak of storm flow, indicating the increasing contribution of overland flow that
19
20 has a lower EC (40 $\mu\text{S cm}^{-1}$). EC then progressively increased as the flood receded. TSS concentration
21
22 reached a peak of 24g L⁻¹ and TSS loss during the 425min event was 4.3 Mg.

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The activity of ²¹⁰Pb_{xs} remained relatively stable throughout the flood event at 80 Bq kg⁻¹). In
contrast, ⁷Be activity was high at the start of the rain event (398 Bq kg⁻¹) it then further increased
slightly to 438 Bq kg⁻¹ at beginning of the flood recession. Activity then dropped drastically to 62 Bq
kg⁻¹ during the flood recession to thereafter increase to more enriched values towards the end of the
flood event. *E. coli* concentrations in the floodwaters were 2100 MPN 100 ml⁻¹ during the early stage
of the rising flood. This then increased a maximum value of 49000 MPN 100 ml⁻¹ at the start of the

1 flood recession. Then, it declined to 830 MPN 100 ml⁻¹ at the end of the flood event. *E. coli*
2 concentration in streambed sediments was also determined and the average concentration was 40 000
3 MPN g⁻¹ (n=4 samples, SD=21602, CV=54%).

4 **Separation of suspended particles and *E. coli* fluxes**

5 The separation of TSS flux into recently eroded particles (“new”) and resuspended sediments flux
6 (“old”) from the streambed is shown in Fig. 4. At the beginning of the flood the major proportion of
7 particles are “new” with an apparent age of 8 days indicating that they were recently marked by ⁷Be at
8 the bare soil surface. This peak corresponds to the peak in overland flow. As the flood progressed, the
9 proportion of “old” particles reached a maximum apparent age of 158 days at 140 minutes after the
10 start of the flood and then declined.

11 The flux of *E. coli* associated with particles recently labelled with ⁷Be was always higher than
12 that associated with the older particles and this during the entire flood. The maximum flux of *E. coli*
13 associated with recently labelled particles (17.9 x 10⁶ MPN x s⁻¹) occurred at peak solid flux (77 mins
14 after the flood peak). Flux then declined steadily until the end of the flood. In contrast to the peak flux
15 of recent particles, the peak in *E. coli* associated with older particles occurred after a lag of
16 approximately 1 hour. We conducted an uncertainty analysis (Monte Carlo type) following the method
17 of Ribolzi et al (2000) to estimate the confidence levels on the proportions of *E. coli* attached to “old”
18 particles at the point of maximum contribution in the hydrograph. We found that the associated errors
19 lead to a coefficient of variation of 12% and 14% of the total fluxes of *E. coli* attached to “old” and
20 “new” particles, respectively. Over the course of the flood a total of 125 x 10⁹ MPN *E. coli* were
21 exported, of which about 89 % were associated with recently eroded particles and 11 % with particles
22 resuspended from the riverbed.

23 **Discussion**

24 Here, we present a novel method using fallout radionuclides (⁷Be and ²¹⁰Pb_{xs}) to estimate the
25 proportions of *E. coli* associated with recently eroded soil particles and with the resuspension of
26 streambed sediments. This method allows the separation of the total flux of particles into two
27 components: “new” and “old” following the model of Matisoff et al. (2005). A key assumption for the

1 use of ^7Be as a tracer is that radionuclides sorption to particles is rapid and irreversible over the
2 timescales of interest (Taylor et al. 2012). Gourdin et al. (2014a) recently validated this assumption by
3 implementing laboratory experiments and field observations during a storm event in the Houay Pano
4 catchment. These authors also used this method to quantify the sources and dynamics of suspended
5 sediment and particulate organic matter (Gourdin et al. 2014b, 2015). Here, we use an extension of this
6 work to estimate the transfer times and sources of *E. coli* in an upland stream (Fig. 4).

7 **Overland flow and groundwater**

8 As observed by Gourdin et al (2014a) we found higher contributions of recently eroded
9 sediment at the beginning of the rising flood stage and then a progressive dilution of recently eroded
10 sediment by particles remobilised from the riverbed (Fig. 4). Overland flow represented just over one-
11 tenth of the total flood volume but was responsible for more than two-thirds of the downstream transfer
12 of *E. coli* (i.e. fraction attached to recently labeled particles). This result confirms the observations of
13 Causse et al. (2015) in the same study area who concluded that the proportion of soil surface washoff
14 was critical in determining the numbers of *E. coli* in the stream during storms. It is also in agreement
15 with Kim et al. (2010) who simulated *E. coli* loads in the Little Cove Creek watershed (Pennsylvania)
16 with a version of SWAT (Soil and Water Assessment Tool) that took into account streambed bacteria
17 release. They found that surface runoff carried large numbers of *E. coli* to the stream and that sediment
18 resuspension contributed to the baseline concentration of *E. coli* in stream water.

19 Another point that should also be emphasized is the importance of the groundwater
20 contribution to flood runoff. During the studied flood event, groundwater largely dominated
21 streamflow with an estimated contribution of up to 89%. This hydrological behavior is often observed
22 in rural catchments with dense vegetation cover (e.g. Kirchner 2003). Genereux and Hooper (1998)
23 estimated in a review paper on tracer-based hydrograph separations that the pre-event groundwater
24 contribution to storms is on average $80 \pm 20\%$. Although this is well known to hydrologists and is often
25 quantified using geochemical and isotopic tracer techniques, this phenomenon is rarely discussed in the
26 literature with a view to better understanding the dynamics of FIB and environmental bacteria. In this
27 context, groundwater that is little contaminated as compared to overland flow will tend to dilute
28 streamwater during stormflow. At the same time, the contribution of groundwater adds to that of
29 overland flow to increase discharge and the resuspension of sediments in the streambed.

1 **Methodological considerations**

2 The method presented here is adapted for tropical systems with contrasted dry and wet
3 seasons. Moreover, it is particularly appropriate for events occurring at the start of the rainy season
4 when the stock of ^7Be supplied by rainfall to soils during the previous wet season has completely
5 decayed. It is also important to note that the first floods are generally the most contaminated as the
6 soils have accumulated contaminants throughout the dry season (Causse et al. 2015).

7 One of the central assumptions of this work is that *E. coli* are attached to soil particles and that
8 this does not change during the erosion, resuspension and transport processes along hillslopes and
9 downstream toward the creek. We hypothesized that enteric bacteria are more frequently associated
10 with particles both within the host and in the external environment (soils and water) and that this
11 remains relatively constant over short timescales. High proportions of environmental bacteria and
12 enteric bacteria such as *E. coli* are associated with particles in soils (Oliver et al. 2007) and in aquatic
13 systems (Lemee et al. 2002, Unanue et al. 1992). Our results, obtained during a flash flood with low *E.*
14 *coli* numbers (1 – 230 MPN 100 ml⁻¹ *E. coli*; Causse et al. 2015) and low TSS (< 20 mg L⁻¹) in the
15 stream prior to the storm support this assumption. Although it is likely valid in this case study, this
16 hypothesis should nevertheless be verified when applying this method to other study sites.

17 Our method allows the determination of the age of different particles to which *E. coli* are bound.
18 Therefore, although it does not provide an exact age for *E. coli* or of the particle, one can postulate that
19 the bacteria associated with the particles in the stream are older than those associated with the particles
20 recently marked with ^7Be (i.e. those at the soil surface on the slopes). We conducted an analysis of
21 error propagation and by far the largest proportion of the total error is by far associated with the
22 calculation of the “old” *E. coli* fraction (up to 75%). Most of this error probably arises from the
23 uncertainty associated with the $\text{SB}_{E.coli}$ determination. Therefore, given the importance of the values of
24 streambed *E. coli* in the calculation, it is critical to obtain a robust estimate of this concentration as the
25 calculations of “old” and “new” *E. coli* fractions are strongly dependent on this value.

26 **Land use**

1 In these tropical agro-ecosystems there is an intensification of activities linked to the
2 preparation of the land for annual crops at the start of the rainy season. This intensification also brings
3 with it a sharp increase in the potential for soil surface contamination due to the use of informal latrines
4 and open-air defecation by humans and livestock. This contamination is then mobilized by the erosive
5 nature of rain drop impacts (Ribolzi et al. 2011b) and is transported to the stream by overland flow
6 (Patin et al. 2012). Bare soils in teak plantations that lack an understory and in young fallow zones
7 where soils have low infiltration rates (Patin et al. 2012) have particularly high overland flows.
8 Moreover teak plantations often encroach on the riparian zones. Therefore any overland flow from the
9 upslope teak plantation may also wash off any FIB available downslope in the riparian zone located
10 between the plantation and stream proper. In the riparian zone, contaminated particles available at the
11 soil surface can also be detached and disseminated downstream within local seepage zones by
12 groundwater exfiltration in association with overland flow (Vigiak et al. 2008).

13 Domestic gardens, temporary housing and fishponds where contamination levels can be high are often
14 located in the riparian zones and the small paths that interconnect these areas can increase runoff
15 (Ziegler et al. 2000) and enhance *E. coli* transport to the stream. Moreover, riparian zones in tropical
16 systems are often characterised by swamp areas that are both a source and a trap of solid particles and
17 FIB. They therefore play an important role in controlling the dynamic of “old” and “new” *E. coli* in
18 these systems. Quantifying the contribution of these reservoirs to the total flux of *E. coli* during flood
19 events allows a better understanding of the redistribution processes and of the dynamic of particle-
20 bound FIB within catchments. This is critical if we wish to fully understand the dynamic of *E. coli* in
21 watersheds.

22 **Conclusion and perspectives**

23 Here we present a way of estimating both the relative proportion of *E. coli* resuspended from
24 streambed sediments and those transferred from the soil surface to the stream by overland flow during
25 a flood event (Fig. 1). At present, because of its analytical cost and level of expertise required for its
26 implementation, the $^{7}\text{Be}/^{210}\text{Pb}_{\text{xs}}$ method is still limited to a small number of laboratories worldwide.
27 Even though the systematic use of this method in a wide range of watersheds is limited, this method
28 remains useful for distinguishing the source of *E. coli* (resuspension of streambed sediments vs soil

1 surface washoff). Furthermore, values obtained from this method can then be applied in appropriate
2 models to understand the dynamics of FIB in streams, and then provide information for adapted
3 management plans that aim to reduce the health risks to the local populations.

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1

2 **Figure 1** - Graphical abstract showing: (1) the main water flow processes (i.e. overland flow,
3 groundwater return flow, blue arrows) and sediment flow components (i.e. resuspension and soil
4 erosion, black arrows) during floods in the Houay Pano catchment; (2) the general principle of the
5 method using fallout radionuclide markers (i.e. ^7Be and $^{210}\text{Pb}_{\text{xs}}$) to estimate *Escherichia coli* load from
6 the two main sources (i.e. streambed resuspension vs soil surface washoff); and 3) the main results
7 obtained during the 15 May 2012 storm event (i.e. relative percentage contribution of each process to
8 the total streamflow, values in parentheses).

9 **Figure 2** - Geographical location of the Houay Pano catchment, hydro-meteorological equipment,
10 water and streambed sediment sampling points, and land use in 2012.

11 **Figure 3** – Measurements during the 15 May 2012 stormflow event: rainfall intensity (6-mins time
12 step); stream discharge; electrical conductivity (EC) of the stream water; total suspended solids (TSS)
13 concentration; fallout radionuclides (^7Be and $^{210}\text{Pb}_{\text{xs}}$) activity; and *Escherichia coli* (*E. coli*) load of the
14 stream.

15 **Figure 4** – Total suspended solid (TSS) flow of “new” (i.e. recently eroded soil surface aggregates)
16 and “old” (i.e. resuspended sediment) particles estimated using the Matisoff et al. (2005) model;
17 overland flow (OLF) and groundwater (GW) discharge estimated using a tracer-based model (e.g.
18 Ribolzi et al., 1997) estimate of *Escherichia coli* (*E. coli*) flow attached to recently labelled and eroded
19 soil surface particles (new) and to resuspended particles from the streambed (old).

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