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ESCHERICHIA COLI REMOVAL FROM STORMWATER  
USING STEEL CHIPS FILTER

BY  
PENG DAI

A thesis submitted in partial fulfillment of the requirements for the  
Master of Science  
Major in Civil Engineering  
South Dakota State University  
2019

ESCHERICHIA COLI REMOVAL FROM STORMWATER  
USING STEEL CHIPS FILTER

This thesis is approved as a creditable and independent investigation by a candidate for the Master of Science in Civil Engineering degree and is acceptable for meeting the thesis requirements for this degree. Acceptance of this does not imply that the conclusions reached by the candidate are necessarily the conclusions of the major department.

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## ABBREVIATIONS

Ag	Silver
ASTM	American Society of Testing and Materials
BEACH	Beaches Environmental and Coastal Health
BMPs	Best management practices
CFU	Colony-forming unit
CWA	Clean Water Act
DBP	Disinfection byproducts
DO	Dissolved oxygen
DOC	Dissolved organic carbon
E.coli	Escherichia coli
EBV	Empty bed volume
EMPT	Empty bed contact time
H <sub>2</sub> SO <sub>4</sub>	Sulfuric acid
HAAs	Haloacetic Acids
KCl	Potassium chloride
KH <sub>2</sub> PO <sub>4</sub>	Monopotassium phosphate
LB	Luria broth base

MF	Microfiltration
Na <sub>2</sub> HPO <sub>4</sub>	Disodium phosphate
NaCl	Sodium chloride
NaHCO <sub>3</sub>	Sodium bicarbonate
NaOH	Sodium hydroxide
NAWQA	National Water Quality Assessment
NF	Nanofiltration
NOM	Nature organic matter
NPS	Nonpoint source
OH <sup>-</sup>	Hydroxide
PBS	Phosphate buffered saline
RO	Reverse Osmosis
RS	Real stormwater
SDDENR	South Dakota Department of Environment and Natural Resources
SEM	Scanning Electron Microscope
SS	Synthetic stormwater
TBA	Tert-Butanol
THMs	Trihalomethanes

TiO <sub>2</sub>	Titanium dioxide
TSS	Total suspended solid
U.S. EPA	U.S. Environmental Protection Agency
UF	Ultrafiltration
ZnO	Zinc oxide

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ABSTRACT

ESCHERICHIA COLI REMOVL FROM STORMWATER

USING STEEL CHIPS FILTER

PENG DAI

2019

Media filtration has received increasing attention as an effective technology that can facilitate the removal of particles and dissolved pollutions in the stormwater. However, most traditional media filtration systems are generally ineffective in removing bacteria, causing pollution of groundwater and receiving surface water. In this study, recycled steel chips were evaluated as low-cost filter materials for bacterial removal from stormwater. Laboratory column experiments were conducted to evaluate the impact of steel chips particle size, flow rate, initial E.coli concentration, pH, natural organic matter (NOM), steady and intermittent flow on E.coli removal. The results showed that E.coli removal decreased from 99.4% to 58.1% with increasing steel chip size from 0.5-1.0 mm to 4.0-8.0 mm. The E.coli removal efficiency of 1.0-2.0 mm steel chips varied between 86.5% to 99.2% and was not greatly affected by variations of infiltration rate, influent bacteria concentration, and pH. The competitive adsorption of co-loading NOM can inhibit E.coli adsorption by up to 80.5%. Intermittent flow can lower E.coli adsorption to 54.3% at the beginning of the flow event and gradually increased to 83.2% with the increase of time. The result also showed that less than 0.4% adsorbed E.coli (total adsorbed E.coli= $6.7 \times 10^9$  MPN) were desorbed in different conditions (flow rate, NOM).

Steel chips also showed high E. coli removal capacity when treating real stormwater.

Overall, the results suggest that steel chips are an efficient E.coli adsorption material that can potentially be used for stormwater treatment under a wide range of field conditions.

## 1. INTRODUCTION

### 1.1 Source of Stormwater Pollution

Stormwater runoff resulted from rainfall or melting snow often causes flooding, erosion and pollution problems. Point source pollution and nonpoint source (NPS) pollution are the two sources leading to stormwater pollution. Since the regulations imposed by Federal Water Pollution Control Act in 1972 and the amendments of the Clean Water Act (CWA) in 1977 and 1983, point source pollution got significantly decreased (Freeman III 2010). NPS becomes the leading reason of stormwater pollution.

Unlike pollution from industrial and wastewater treatment plants, NPS pollution comes from many different sources. As rainfall, snowmelt, or irrigation water flows through the ground, the pollutants are carried and deposited into rivers, lakes, wetlands, coastal water and ground waters, which can destroy water quality. The most common NPS pollutants include salt from irrigation practice and acid wastewater from abandoned mine drainage; extra fertilizers, herbicides, and insecticides from agriculture and residential areas; oil, grease and poisonous chemical compounds from urban and factory runoff; bacteria and nutrients from livestock and manure and faulty septic systems (Baker 1992). Agriculture and urbanization are the main contributors to stormwater pollution (Novotny 1995; Pitt et al. 1995).

Agriculture in American is famous for its high productivity, quality, and efficiency. More than 330 million acres area is used as cropland (Alston et al. 2009). When stormwater passes through agriculture area, large amount of salts, nutrients such as phosphorus and nitrogen, heavy metals, herbicides, pesticides, and soil particles can be

carried into the rivers and lakes. The National Water Quality Assessment (NAWQA) indicates that agriculture is the leading source for pollution of rivers, streams, surveyed estuaries and ground water, and the third largest source for lake pollution (Carpenter et al. 1998; Puckett 1995). The pollution mostly occurs in a poorly-managed area which is lack of conservation plan such as inappropriate animal feeding operation, poor manure treatment, overgrazing, too often plowing, wrong time and improper fertilizer application (Ongley 1996).

Urbanization can significantly alter the landscape and consequently influence water quality. Physically, the expansion of urban area increases the impervious surface areas such as building roofs, parking lots, sidewalks, and roads. Those impervious surfaces do not allow water to penetrate the ground, increasing the percentage of precipitation that becomes runoff from about 10% in an undisturbed setting to around 55% in a fully urbanized setting, contributing to the increased flooding frequency and excessive streambank erosion (Figure 1) (Paul and Meyer 2001). Chemically, organic and inorganic pollutants are introduced into urban systems, which can significantly deteriorate water quality and cause human health concerns. The Department of Economic and Social Affairs announced that with the increased pace of urbanization, the world population living in urban areas is expected to reach 6.3 billion by 2050, the stormwater pollution problems are expected to get worse over time (UN 2014). Therefore, understanding the source and transportation of contaminants in urban areas is important to determine the most appropriate management method to improve the urban stormwater quality.

## 1.2 Stormwater Pollutants

Stormwater can carry many organic and inorganic pollutants including nutrients, total suspended solids, floating litter, pathogens, organic chemicals, heavy metals, and oil & grease into rivers and lakes.

Phosphorus and nitrogen are the nutrients with a lot of concerns in aquatic ecosystems because their existence will promote weed and algae growth in rivers and lakes. Excess weed growth (known as algae bloom) clogs water ways and blocks sunlight, preventing photosynthesis of the aquatic plants and leading to low dissolved oxygen (DO), the death of fish and other aquatic plants. These will significantly disrupt the ecosystems (Alabaster and Lloyd 2013; King 2013). Fertilizer, excrement, and detergents are the main sources of phosphorus and nitrogen in stormwater (Mallin et al. 2009).

Total suspended solids (TSS), generally comes from soil erosion by wind, water, and human agriculture activities. When the soil is carried into a river, it can disrupt the ecosystem in many ways. Large amounts of silt can deposit in sensitive areas such as wetlands, wildlife preserves, stream, and lake bottoms, destroying the aquatic life living habitat (Davis and McCuen 2005). TSS can also block sunlight needed by aquatic plants and carry biodegradable chemicals that lead to the depletion of the oxygen in the water (Carr and Neary 2008; Kim et al. 2012).

Million tons of human-made solid litter enter the river every year. The toxic chemicals, undegradable plastics, and bacteria in the litter can cause water pollution, leading to the death of aquatic animals and birds. Commonly observed floating litters

include cigarette flitter, plastic bags, food wrappers, beverage bottles and caps, straws, etc., in which 60-80% wastes are plastics (Rios et al. 2007). According to the United Nations Environment Assembly statistics and previous studies, nearly 4.8-12.7 million tons of plastic waste enter the world's oceans each year (Jambeck et al. 2015). Those floating debris liters are often eaten by aquatic animals and birds, high concentrated toxic chemicals can be absorbed into the bloodstream, and the debris can fill their stomachs, causing them to starve to death. Those floating litter can also have the habitat, chemical and biological impact for animals, and eventually impact human.

Microorganisms include bacteria, viruses, and parasites. When the water with disease-producing microorganisms been consumed by human, it can lead to gastrointestinal disease. The sources of microbial pollution usually include human and animal wastes, leakage of sewers, underground storage tank, and seepage from septic tanks (Jamieson et al. 2004).

Organic matters (natural or synthetic) can be carried into the surface water by stormwater runoff. The biodegradable organic matter such as protein, amino acid, carbohydrate, and fatty acid can be oxidized by bacteria under the aerobic condition and may deplete DO in the surface waters, threatening aquatic life (Manahan 2017). Some of those materials can also generate color, taste and odor problems. The non-biodegradable organic matters include tannic and lignic acids, cellulose, and phenols, when discharged into the water, can be toxic to aquatic animals and microorganisms (Volk et al. 2000). Many of them are cumulative toxins, which can be accumulated and eventually transferred into the higher end of the food chain, threatening the health of humans.

Heavy metals in water can be toxic to aquatic life and other animals, and directly or indirectly threaten human health. The most common heavy metal pollutants in the river are cadmium, copper, lead, mercury, nickel, chromium and arsenic (Mulligan et al. 2001). Heavy metals cannot be degraded naturally in the environment, instead they will be accumulated in fishes or plants, and eventually transferred into human by the food chain. The heavy metals usually come from a variety of human activities including mining, vehicle emissions, tires, and motor oil factory (Wackernagel and Rees 1998).

Oil and grease can be toxic to aquatic life. When exposed to oil and grease, fish can experience retarded growth, livers enlargement, changes in heart and respiration rates, fin erosion, and reproduction impairment (Roberts 2012). Oil and grease can also cut off the air exchange and sunlight to plants, thus interfering the plants' photosynthesis, and making the plants failed to germinate and grow (Chawla 2002). Sometimes, the dispersants (such as *procentrum texanum* and *scripsiella trochoidea*) used in oil cleaning will also result in an increase of algae bloom (Almeda et al. 2018). The sources of oil and grease in storm drains usually come from automotive leaks and spills, improper disposal of used oil and automotive products.

### **1.3 Stormwater Managements**

Detention and/or infiltration treatment systems are more practical methods for NPS pollution control. Today, stormwater management includes flooding control, erosion management, and water quality improvement. This can be accomplished by infiltration systems and filter systems.

Extended detention ponds, wet pond/detention ponds, infiltration basin, and porous pavement are the most common infiltration systems best management practices (BMPs) (Akan et al. 2003). Instead of flowing directly to a river, stormwater can be temperately transported to and stored in the detention pond until the pollutants deposit to the bottom, decreasing flooding and pollution problems (Scholes et al. 2008). Similar to extended detention ponds, wet ponds can also be used as storage for stormwater. A wet pond's permanent pool can retain runoff from stormwater, increasing pollutant removal through sedimentation and biodegradation. Infiltration basin works in a different way. Stormwater can be stored in the infiltration system and infiltrate into sand and soil. This system only works well in fine particle pollutants removal. Large amounts of coarse particles in stormwater may block infiltration basins. Porous pavement can increase stormwater runoff infiltration to the soil and help the removal of fine particles. Water quality inlets are also known as trapping catch basins, which are mainly designed to reduce sediments, oils, and graces from runoff generated from parking lots before discharging to the storm drain or infiltration basin. Normally modified trapping catch basin serves as a pretreatment step for other BMPs. It is not sufficient to be a stand-alone unit for stormwater treatment.

Filtration system includes two major units: sedimentation chamber and filter media. Sedimentation chamber is designed to remove TSS to prevent clogging problems and filter media is designed for dissolved pollutants removal. The performance of filtration system is influenced by water quality, flow rate, physical and chemical characteristics of the media. Many materials include sand, coated sand, iron filling,

zeolite, limestone, wood chip, biochar have been extensively studied as filter media to treat different types of pollutants.

The advantages and disadvantages of BMPs must be weighed individually against physical site restraints, management objectives (flow control and/or water quality control) and costs. Due to the limitations in permeable surface and groundwater pollution, infiltration systems are not suitable for urban areas. Comparing with the infiltration systems, filter systems have fewer restrictions in physical requirements such as soil characteristics, pollutant characteristics, loading rate, land use and groundwater level, and the smaller footprint make it can be designed to fit small drainage areas (less than 5 acres) (Fetter et al. 2017; Kawamura 2000). Therefore filtration is now widely applied in urban stormwater management systems.

#### **1.4 E.coli Pollution in Stormwater and Control**

E.coli is a species of bacterium that commonly found in the environment, foods, and the intestines of human and animals (Sondi and Salopek-Sondi 2004; van den Bogaard and Stobberingh 2000). E.coli are a large and diverse group of bacteria, and most E.coli are harmless or cause relatively mild diarrhea. However, several strains such as E.coli O157:H7, can significantly deteriorate water quality and cause health problems such as severe stomach cramps, diarrhea, vomiting, and fever (usually less than 101°F/38.5°C) (Abuladze et al. 2008; Croxen et al. 2013; Regulations et al. 1986). Healthy adults can recover from E.coli O157:H7 infection within a week, but it takes longer (5-10 days) for young children and older adults to recover (Cray and Moon 1995). One point three (1.3) million children under age five die every year because of diarrheal and contaminated water is one of the main spreading ways (Black et al. 2010; Thapar and

Sanderson 2004). The presence of E.coli in water samples can be considered as a reliable indicator of potential presence of fecal contamination. The U.S. Environmental Protection Agency (U.S. EPA) sets up E.coli as an indicator of fecal contamination and regulates 126 colony-forming unit (CFU)/100 mL (or 126 MPN/100 mL) as the limit concentration of freshwater (Cho et al. 2010; Noble et al. 2003). In October 2000, US Congress passed the Beaches Environmental and Coastal Health (BEACH) Act, which requires that states with marine or great lakes coastal recreational water adopt U.S. EPA Water Quality Standards, and encourages monitoring and public notification programs (Congress 2000).

E.coli can be transported into freshwater through stormwater runoff that passes through bird feces, wild and domesticated mammalian feces. Municipal sewer overflows are also considered as a major source of E.coli (Selegan et al. 2001). Although E.coli concentration is regulated, E.coli contamination in water still cause 120 million gastrointestinal and 50 million cases of severe respiratory illness per year around the world (Shuval 2003).

The Big Sioux River is a tributary of Missouri River in eastern South Dakota and northwestern Iowa. Based on South Dakota Department of Environment and Natural Resources (SDDENR) data, the E.coli average concentration in different sample sites of Big Sioux River can reach as high as 5093 MPN/100 mL, and the peak concentration during the summer rainstorm season can reach 8000 MPN/100 mL. Nearly all flow zones significantly exceeded the EPA regulation, which can cause public health concerns to the related residents (COUNTY and DAKOTA 2011; COUNTY and DAKOTA 2017).

Therefore, it is important to have a good method to control E.coli in stormwater before it enters water systems.

There are many methods that have been developed in the last decades for E.coli control. One of the most widely used E.coli disinfection methods is chlorine disinfection, by adding chlorine gas, hypochlorite solution, and other chlorine compounds in a solid or liquid phase, to oxidize cellular material to destroy target organisms. Chlorine disinfection has existed for over one hundred years. It was first formally proposed in 1894, and early attempts to implement at a water treatment plant started in 1893 in Hamburg, Germany (Turneure and Russell 1901). After many years of development, it has become a well-established technology. Chlorine disinfection is a low-cost, reliable method that can effectively treat a wide spectrum of pathogenic organisms. The residuals that remain in the water can prolong the disinfection effect. However, chlorine disinfection has its disadvantages. The chlorine residuals are toxic to aquatic life and may need dechlorination before discharging to surface waters. Due to the highly corrosive and toxic nature of chlorine, the storage, shipping, and handling of chlorine require higher safety regulations. Chlorine is a strong oxidizing agent which can react with nature organic matter (NOM) to form cancer-causing disinfection byproducts (DBP) such as trihalomethanes (THMs) and haloacetic acids (HAAs) (Krasner et al. 2006; Nieuwenhuijsen et al. 2000; Richardson et al. 2007). Those disadvantages limited the application of chlorine disinfection in stormwater.

Ultraviolet (UV) light can effectively inactivate E.coli by destroying E.coli DNA and RNA. A mercury arc lamp used for UV disinfection can generate electromagnetic energy to destroy an organism's DNA and RNA (primarily at 254nm), thereby damaging

the cell's reproduction and infection ability. Therefore, UV disinfection is effective for most viruses, spores, and cysts (Rastogi et al. 2010; Sinha and Häder 2002). The disinfection process requires 20 to 30 seconds with low-pressure lamps which is shorter than other disinfectants (Blume and Neis 2004). Since this is a physical process rather than a chemical disinfectant process, UV disinfection eliminates the requirements to generate, store or transport toxic/hazardous chemicals. And this method would not generate any byproducts that are harmful to human or aquatic life. Thus, UV disinfection is now widely used in water and wastewater treatment plants. However, there are several factors can influence UV disinfection effectiveness including the characteristics of water, the transparency of UV light, UV light dosages, and the disinfection time. The application of UV disinfection is limited to water with high light transmittance (TSS levels  $< 30\text{mg/L}$ ) which is difficult to achieve for stormwater without any pre-treatment. Also, the high energy consumed by UV light makes it expensive to be applied in large-scale stormwater treatment (Hijnen et al. 2006; Van der Bruggen et al. 2003).

Membrane filtration provides physical barriers to remove particulates greater than the membrane pore size. Membrane separation is an effective method for E.coli removal (Srivastava et al. 2004). Microfiltration (MF), Ultrafiltration (UF), Nanofiltration (NF) and Reverse Osmosis (RO) systems are the most common membrane systems in the market. The benefits of membrane filtration include high rejection rate, high recovery, small footprint, easy operation, and ability to handle high variation in water quality and quantity (Fritzmann et al. 2007; Gabelich et al. 2007; Martinetti et al. 2009; Sourirajan 1970; Spiegler and Kedem 1966). However, due to high maintenance for membrane fouling and scaling, the high price of the membrane and energy consumption, membrane

filtration is mainly used in water treatment process. It has not been extensively used in stormwater treatment (Hilal et al. 2004; Mansouri et al. 2010).

Most of the conventional filtration systems are mainly designed to control runoff volume and to reduce suspended solids. The filter materials are usually formed by soil, sand and/or pebble, which are not effective for bacteria removal. Several emerging filtration materials including nanoparticles, activated carbon, biochar, and titanium dioxide have been evaluated for bacteria removal.

Adhikari et al. (2015) used photocatalytic inactivation to remove E.coli. The authors evaluated ZnO-Ag (Ag impregnated ZnO) nanoparticles under solar radiation. The results showed that 0.25 g/L catalyst can effectively inactivate  $10^9$  CFU/mL bacteria (Adhikari et al. 2015). TiO<sub>2</sub> was used as a photocatalyst to generate OH radicals under UV light for E. coli disinfection. A linear correlation between OH radicals and E.coli inactivation rate was observed, which showed that OH radicals are the dominant oxidant species in the UV/TiO<sub>2</sub> disinfection process (Cho et al. 2004). Two large scale water treatment units (ozone-sand filtration and powdered activated carbon-ultrafiltration) were built in Lausanne wastewater treatment plant in Switzerland to compare the micropollutants control efficiency. Both units performed well and showed over 80% bacteria removal (Margot et al. 2013). Despite the high cost, powdered activated carbon-UV treatment was considered as the better option for drinking water and recreational water due to the high bacteria removal and no disinfection byproduct formation (Margot et al. 2013). Sand and biochar (1-2%) mixture was found achieved as high as 40% E.coli removal for an influent concentration of  $1.0 \times 10^7$  CFU/mL (Abit et al. 2012). In order to test whether the biochar can effectively remove E.coli in more complex situations,

Mohanty and Boehm (2014) conducted experiments to evaluate the impact of NOM on E. coli removal by column reactors with a mixture of biochar and sand. Approximately 95-99% E.coli removal was observed in biochar columns. The presence of NOM decreased E.coli removal by columns reactors with biochar (Mohanty and Boehm 2014). Some studies used metal-coated sand to improve E.coli adsorption. A long-term (4 months) experiment was conducted to test the iron sand filter's performance in E.coli removal. The test results showed that iron oxide-coated sand filter had better performance in E.coli removal (95-100% removal) than conventional sand filter (80-90% removal), and the performance improved with time (Ahammed and Davra 2011).

Many of the materials evaluated for E. coli removal requires industrial production, which may increase the cost of their application for stormwater treatment. Some of the materials such as nanoparticle and activated carbon may clog the filter or be easily washed away due to their fine sizes. Those limitations restrict their application in stormwater treatment. Therefore, it is necessary to develop effectively, and low-cost filtration materials for E.coli removal in stormwater.

Recycled steel byproducts such as steel chips are a new group of filter materials that have shown the ability to removal E.coli in stormwater (Hooshyari 2017). Steel chips are readily available in many areas. They are byproducts of the grinding, drilling or milling of steel products. The surface of the steel chip can be oxidized in the presence of water and oxygen, forming reactive layers of iron oxides for E.coli adsorption. As a material that is typically landfilled or recycled back into the steel making industry, steel byproducts potentially offer a low-cost solution for E.coli removal from stormwater.

NOM is a complex mixture of organic compounds in aquatic environments. Previous studies found that the functional groups in NOM such as carboxylic acids, hydroxyls and quinones could significantly affect the deposition of microbe to metal-oxide coated surface by weakening the electronic attraction between bacteria and media (Foppen et al. 2008; Johnson et al. 2009; Levy et al. 2007). Some studies showed that NOM would compete for the available adsorption sites for bacteria (Abu-Lail et al. 2007; Chi and Amy 2004; Fein et al. 1999; Foppen et al. 2006).

Most of E.coli removal capacity studies were conducted under continuous flow condition. However, the stormwater filters would experience wet-dry cycles in real treatment conditions. The impact of wet-dry cycles on E. coli removal by filtrations technologies has not been carefully evaluated. Therefore, it is important to study the impacts of intermittent flow on E.coli removal.

### **1.5 Research objective**

The main objective of this study was to investigate the efficiency of steel chips for E.coli removal from stormwater. The impacts of the filter size, initial bacterial concentration, flow rate, pH, NOM, and intermittent flow were studied to evaluate the potential application of the steel chips for stormwater E. coli treatment. The E.coli removal performance of steel chips in real stormwater was also investigated. Overall, the results of this study can lead to the full-scale application of steel byproduct filtration technology to control E. coli in stormwater runoff.

## 2. MATERIALS AND METHODS

### 2.1 Filter Materials

The steel chips (1018 carbon steel) obtained from Prairie Manufacturing, LLC (Sioux Falls, SD), are a byproduct of the grinding, drilling or milling of finished iron products (Figure 2). The steel chips were washed with phosphate-free soap to remove the surface oil and air-dried for 48 hours. During the air-drying process, the surfaces of the steel chips were oxidized and covered with a layer of rust. To identify the impact of particle size on E.coli removal, the steel chips were sieved with four screens with different mesh sizes (fine 0.5-1.0 mm, small 1.0-2.0 mm, medium 2.0-4.0 mm and large 4.0-8.0 mm) before the column experiment. The physical and chemical characteristics of the steel chips were tested following the American Society of Testing and Materials (ASTM) standard testing procedures and the results are given in Table 1.

### 2.2 Stormwater

Synthetic stormwater (SS) and real stormwater (RS) were used in the experiment. The SS (also be used as E.coli buffer solution) was prepared by dissolving 1.0 mM  $\text{NaHCO}_3$ , 0.1 mM KCl in deionized water and the pH was adjusted to  $7.1 \pm 0.1$  using  $\text{H}_2\text{SO}_4$ . In order to prevent E.coli multiplication in SS during the experiments, no nutrient elements were added into SS. The experimental results suggest that E.coli concentrations can keep relatively stable for 4 days in SS at room temperature (23 °C). To test the NOM effects on E.coli removal, Suwannee River NOM (International Humic Substances Society, St. Paul MN, USA) was added to SS to achieve DOC levels of 5 or 30 mg/L during the NOM experiments.

The RS was obtained from three different stormwater drainages located in south central (44°17'01.9"N, 96°47'23.1"W), eastern (44°17'10.0"N, 96°44'56.2") and western (44°14'56.9"N, 96°49'42.0"W) areas of Brookings County (SD) during the rainstorms in 15<sup>th</sup> June and 22<sup>nd</sup> July, 2017.

### **2.3 Bacterial Preparation**

The E.coli preparation procedure is summarized in Figure 3. The gram-negative *Escherichia coli* (ATCC 35218, American Type Culture Collection) were used as model bacteria. One container of 100 mL Luria Broth Base (LB, Thermo Fisher Scientific, 10 g/L peptone, 10 g/L sodium chloride, 5 g/L yeast extract) was inoculated with E.coli from a frozen stock maintained at -20 °C. E.coli was then grown in 37 °C and 150 rpm under aerobic conditions in a temperature controlled orbital shaker (Thermo Scientific MaxQ 4000) for 24 hours to reach the stationary growth phase. A centrifuge (Thermo Scientific Sorvall™ ST 8 Benchtop Centrifuge) was used to separate bacteria from LB growth media. The LB growth media was separated evenly into three 50 mL centrifuge tubes and centrifuged at 4000 rpm for 10 minutes. Then the centrifuge tubes were taken out of the centrifuge and the liquid phase (mainly LB growth media) was discarded. Thirty mL buffer solution was then added to the centrifuge tubes in which the bacteria were re-suspended. The tubes were put into the centrifuge and centrifuged at 4000 rpm for 10 minutes. The centrifuge process with buffer solution (1 mM NaHCO<sub>3</sub>, 0.1 mM KCl, pH=7.1±0.1 using H<sub>2</sub>SO<sub>4</sub>) was repeated three times to completely separate the E.coli from LB media. At last, separated E.coli was re-suspended into 100 mL stock solution. The E.coli concentration in the stock solution was estimated by a spectrophotometer (HACH DR/400 OU, Germany). This culture process resulted in E. coli concentrations in

the range of  $1.0\text{-}1.3 \times 10^9$  cells/mL in the stock solution. Then E.coli stock solution was used to add E. coli to the simulated stormwater samples.

E.coli concentration was accurately quantified by Coli-18 (IDEXX; Westbrook, Maine) method (i.e., most probably number, MPN). One pack of Colilert reagent was added to a 100 mL water sample. After that, the sample/reagent mixture was poured into a Quanti-Tray\*/2000 and sealed in an IDEXX Quanti-Tray\* sealer. The tray was placed in an oven at  $35 \pm 0.5$  °C for 24 hours and the result was read according to the result interpretation. The picture of E.coli test kits is given in Figure 4.

All dishes were soaked in phosphate-free soap 24 hours and then rinsed with reverse osmosis (RO) water. The dishes were then put into 25% sulfuric acid bath for 24 hours and rinsed with 18 MΩ nanopure water. All the dishes were autoclaved before using. To avoid any contamination, disinfection cleanses (Steris Coverage Spray HB plus) were used to sterilize all equipment.

#### **2.4 Sample Preparation for Scanning Electron Microscope (SEM)**

The moisture on the steel chips sample was removed before the SEM test. The pre-treatments included bacteria fixation, gradient dehydration (Figure 5) and freeze dry (to remove moisture while keeping E.coli cell morphogenesis). 0.01 M phosphate buffered saline (PBS) was prepared by adding 136.75 mM NaCl, 0.27 mM KCl, 10.14 mM  $\text{Na}_2\text{HPO}_4$ , and 1.76 mM  $\text{KH}_2\text{PO}_4$  in deionized water. The steel chips samples were washed twice by PBS and added into 2.5% Glutaric dialdehyde PBS solution (25% Glutaric dialdehyde mixed with 0.01M PBS) to fix bacteria onto the steel chips surfaces. The fixed samples were taken out after 24 hours and then washed twice with 0.01 M PBS

and dehydrated using graded ethanol (30%, 50%, 70%, 80%, 90%, 95%, 100%), soaked 20 minutes for each concentration ethanol. Finally, the chips were washed with liquid Tert-Butanol (TBA) three times (15 minutes each time), freeze-dried and kept in dry storage.

## 2.5 Experiment Design

The experiments were conducted using Omnifit® fixed-bed glass columns (Kinesis Inc., West Berlin, NJ) with 15 cm height and 1.5 cm inside diameter (Figure 6). Unless particularly stated, the experiments were conducted under the small size (1.0-2.0 mm) filter, SS pH=7.0, E.coli concentration= $10^6$  MPN/mL and the flow rate=1.77 mL/min (empty bed contact time (EBCT) =10 minutes) condition. All experiments were repeated three times. The experiment reactor system is shown in Figure 7.

All solutions used in this study were prepared with ultrapure water (18 M $\Omega$ -cm) produced by a Barnstead NANOpure system. The pH was measured with an Orion 290A+ advanced ISE/pH/m/OPR meter (Therom Electron, Waltham, MA). Dissolved organic carbon (DOC) concentrations were determined with a Shimadzu TOC-5000 Analyzer (Shimadzu Corp., Kyoto, Japan). E.coli concentration was measured with the Coliart 18 (IDEXX; Westbrook, Maine) method. And the surface characteristics of steel chips were analyzed by SEM (Hitachi S-3400N).

The EBCT is determined by the empty bed volume V (mL) divided by volumetric flow rate Q (mL/min).

### 2.5.1 E.coli Adsorption

The steel chips were separated into four groups according to their sizes. Steel chips with known mass (30 g fine, 30 g small, 13 g medium, 10 g large) were packed into the columns to achieve 10 cm column height.

432 empty bed volumes (EBV) SS with varying E.coli concentration ( $10$ ,  $10^2$ ,  $10^4$ ,  $10^6$  MPN/mL) and initial pH values (5.0, 7.0 and 9.0) were pumped upward through the columns at flow rates of 3.53, 1.77, 0.88 mL/min to achieve EBCT of 5, 10, 20 minutes using a peristaltic pump. The effluent was sampled every 72 EBV and analyzed for E.coli.

NOM pre-loading and co-loading experiments were conducted to study the NOM effects on E.coli removal. For NOM pre-loading experiments, 432 EBV SS (no E.coli, NOM=5 and 30 mg/L) was injected into the column followed by 432 EBV SS (E.coli= $10^6$  MPN/mL, no NOM) at flow rate 1.77 mL/min, separately. For co-loading experiments, 432 EBV SS ( $10^6$  MPN/mL E.coli) with 5 or 30 mg/L NOM was pumped into the columns at a flow rate of 1.77 mL/min. The effluent was sampled every 72 EBV and analyzed for E.coli.

Intermittent flow adsorption experiments were conducted to study the effects of wet-dry alternation on E.coli removal. In the short-term experiment, 144 EBV SS were separated into four equal flow events and pumped into two columns (column A and B) separately at a flow rate of 1.77 mL/min. After the 36 EBV SS injection, the peristaltic pump was stopped for 1h. Column A was drained (disconnecting the column bottom tubing, empty the water in the column by gravity) and column B was kept saturated.

Followed by 1-hour pause, another 36 EBV SS was pumped into the columns. The process was repeated three times to mimic natural rainfall conditions. In the long-term experiment, the suspension time was 14 days, 432 EBV SS injection per event and conducted three events total, and the flow rate was 1.77 mL/min. The short-term and long-term effluent were sampled every 6 EBV and 72 EBV, separately, and analyzed for E.coli

E.coli were also suspended in the real stormwater to achieve  $10$ ,  $10^3$ ,  $10^6$  MPN/mL concentrations. 432 EBV RS were pumped continuously into three columns separately under 1-2 mm size steel chips and 1.77 mL/min flow rate condition. The effluent was sampled every 72 EBV and analyzed for E.coli.

### **2.5.2 E.coli Desorption**

To examine the desorption potential of E.coli from small steel chips, wet-dry cycle intermittent flow and four desorption solutions (including pH 7.0, 9.0 and NOM 5, 30 mg/L) under continuous flow were used and compared. Columns were initially loaded with E.coli by pumping SS with  $10^6$  MPN/mL E.coli at a flow rate of 1.77 mL/min (EBCT=10 min) for 432 empty bed volumes (3 days). For wet-dry cycle intermittent flow experiments, 36 EBV SS without E.coli was pumped through Columns A or B. During the 1-hour suspension time, column A was drained and column B was kept saturated. The wet-dry cycles were repeated four times for each column. The effluent was sampled every 6 EBV and analyzed for E.coli. For varying desorption solution experiments, the desorption solutions were continuously pumped into the columns at a flow rate of 1.77 mL/min for 144 empty bed volumes. The effluent was sampled every 24 EBV and analyzed for E.coli.

### 3. RESULT AND DISCUSSIONS

#### 3.1 Effects of steel chips size, flow rate, initial bacteria concentration and pH on E.coli adsorption

Figure 8a compares the E.coli removal efficiency with different steel chips sizes. Among the four sizes of steel chips, fine steel chips (0.5-1.0mm) had the highest E.coli removal efficiency, which was stable at around 99%. With increasing particle sizes of 4.0-8.0 mm, the removal efficiency declined from 99.4% to 58.1%. Surface area is an important property related to adsorbent removal efficiency. Small particles typically have some surface areas and adsorption sites than large particles (Najm et al. 1990). E.coli can be attached on the steel chips outer surface or pores large than 2  $\mu\text{m}$  (average sizes of E.coli cells are 1-2  $\mu\text{m}$ ). With a higher surface area, 0.5-1.0 mm steel chips exhibited better removal efficiencies (40.5-42.0%) than 4.0-8.0 mm steel chips. The morphology of steel chips surface after the E.coli adsorption can be seen in Figure 9b. A large amount of E.coli (long rod-shaped cells) distributed on the steel chips surface. Additionally, fine steel chips have higher packing density (Table 1), and this could also increase bacteria removal by straining (Bradford et al. 2004; Sohn and Moreland 1968).

The E.coli removal performance of steel chips was tested under three different flow rates (3.53, 1.77, 0.88 mL/min, and 5, 10, 20 min EBCT, respectively) to simulate a range of detention times that may occur under field application conditions. The U.S. EPA recommends a five-minute retention time for E. coli removal from stormwater. Previous studies found that the increasing flow rate had a negative effect on colloid deposition (Li et al. 2005). The results in Figure 8b show a similar trend with the increase of flow rate.

*E. coli* removal efficiency at 5 min EBCT decreased by 10% than 20 min EBCT results. Although high flow velocity could decrease the *E. coli* removal efficiency, the removal efficiency was not substantially decreased by the increase of flow velocity. The highest EBCT (5 min) tested during this study exhibited an average of 87.6% removal efficiency of *E. coli*. This demonstrates that steel chips are effective for *E. coli* adsorption for the range of EBCTs tested in this study. Similar results were also observed in biochar amended biofilters. Varying residence times between 5 and 30 min resulted in less than 5% difference in *E. coli* removal efficiency from the filters, and it was explained by the presence of a strong adhesive force between bacteria and biochar (Mohanty and Boehm 2014). Similar to biochar, steel chips ( $\text{Fe}^{2+}$ ,  $\text{Fe}^{3+}$ ) are expected to have net positive charges on the surface. Therefore, negatively charged *E. coli* can be attached tightly on the steel chips surface. The shear stress caused by high filtration rate (EBCT=5 min) was not strong enough to detach *E. coli* from steel chips.

The *E. coli* concentration range can vary from 0 to  $10^4$  CFU/mL in natural water (Grebel et al. 2013). To test steel chip performance under different *E. coli* concentration, the initial *E. coli* concentrations was set at a range of 10 to  $10^6$  MPN/mL. As seen in Figure 8c, the removal efficiency in the steel chip columns did not show substantial changes for different initial bacteria concentrations. With the increases of *E. coli* concentration to as high as  $10^6$  MPN/mL, the removal efficiency decreased slightly from 98.1% to 89.7%, indicating that some of the adsorption sites might be exhausted at high *E. coli* concentrations. Similar results have been observed and explained by a “blocking” effects on the surface adsorption in other studies (Camesano and Logan 1998; Camesano et al. 1999; Dabros and Van de Ven 1993; Dabroś and Van de Ven 1983; Rijnaarts et al.

1996). Bacteria at higher concentration could block attachment sites more rapidly than lower concentrations.

Figure 8d shows the effect of pH on E.coli removal by steel chips. It is well known that the adsorption capacity of electrically charged matters is highly correlated to the pH values. In this experiment, the solution pH may have modified the steel chips surface through ionization of iron oxides. The surface of steel chips was more positive charged in acidic solutions, and the increase of electrostatic attraction makes the active binding sites more available for E.coli. Under alkaline conditions, high concentrations of  $\text{OH}^-$  would decrease iron surface positive charge, leading to a weaker electrostatic adsorption between steel chips and E.coli. Therefore, steel chips showed a higher E. coli removal efficiency at pH 5 than at pH 7. The E. coli adsorption by steel chips at pH 9 showed variations based on time of column reactor operations, in which E. coli removal by steel chips reduced from 94.8% to 86.5% when pH was increased from 5 to 9 after 12 hours of operation. However, the removal of E. coli by steel chips at pH 9 steadily increased beyond 12 h and reached about 92% after 48 h (Figure 8d). The possible reason is an increase of the ferric hydroxide colloid formation. Based on previous studies,  $\text{pH} > 7$  is favorable for ferric hydroxide formation (Gayer and Woontner 1956; Morgan and Lahav 2007; Stumm and Lee 1961). Visual observation of the steel chip column demonstrated that reddish aggregates gradually accumulated on the steel chips, especially at pH 9. The formation of ferric hydroxide precipitation may have contributed to the higher removal of E. coli.

### 3.2 Effect of NOM on E.coli adsorption and desorption

It's important to study the influence of NOM on E.coli removal because NOM is ubiquitous in stormwater. The experiment was separated into a pre-loading part and co-loading tests. In the NOM pre-loading experiment, the removal efficiency was 73.7-79.1% and 66.1-70.6% at the first six hours under 5 and 30mg/L NOM conditions, respectively. With the increase of time, the removal efficiency gradually increased to 80.7% and 78.9% (Figure 10b), indicating that the impact of preloaded NOM on E.coli attachment gradually reduced. Other studies have reported that the presence of NOM would decrease bacteria removal capacity (Mohanty and Boehm 2015; Mohanty et al. 2014; Yang et al. 2012). Thus, the effluent NOM concentration was analyzed to find the possible reasons (Figure 10a). The results of the effluent DOC analyses showed that the majority of attached NOM were washed out after 12 hours for both conditions. After that, the remaining NOM slowly detached from the steel chips with increasing operation time. Since more than 80% NOM was removed and no longer occupied the adsorption sites, the effect of NOM on E. coli adsorption was minimized. Compared with pre-loading, both columns suffered from the competitive adsorption from NOM, especially at a concentration of 30 mg/L. The E.coli removal efficiency by steel chip columns was reduced to 38.8% and 19.2%, respectively, for 5 and 30 mg/L DOC levels. The reduced E. coli adsorption in the presence of NOM may be attributed to the fact that the negative charged NOM and E.coli cell would repel each other. Another possible reason is that the presence of NOM in solution would generate more repulsive electrostatic force between microbe and collector, reducing E.coli adsorption. Several studies found that NOM can also influence both E.coli and steel chips surface ionization. The presence of NOM in

microbe suspensions resulted in the sorption of humic acid onto microbe surface, which induced microbe surface properties change. Bacteria showed more negative zeta potentials of oocyst (-35mV) when humic acid was added than that without NOM (-12.5mV) (Dai and Hozalski 2003). The sorption NOM alters goethite surface charge, leading a reduction in *E. coli* attachment (Foppen et al. 2008). Apart from the influence of electrostatic force, hydrophobic interaction might also play an important role in *E.coli* attachment (Chen and Walker 2012). NOM are quite hydrophobic. Steel chips and *E.coli* could be more hydrophobic in the presence of NOM, which makes the attachment more difficult.

Thus, on the basis of the experiment results shown above and in previous studies, the influence of NOM on *E.coli* adsorption may be summarized in the followings: (1) NOM can be absorbed on the steel chips surface and occupy the attachment sites; (2) the adsorption of NOM can increase electrostatic repulsion between steel chips and cell surface; and (3) the adsorption of NOM affects the steel chips and bacteria surface hydrophobicity.

### **3.3 SEM Analysis**

The steel chips surface structure and characteristics were observed with SEM analysis (Figure 9). Figure 9b, c, and d clearly illustrates the distribution density variations of *E.coli* on different steel chip filters. Figure 9 shows that the presence of NOM is capable of decreasing *E.coli* densities on steel chip surfaces, especially at high concentrations. This result is consistent with the findings of *E.coli* adsorption experiments.

### **3.4 Effect of intermittent flow on E.coli adsorption**

E.coli adsorption during short and long-term intermittent flow events are shown in Figure 11. In short-term experiments (Figure 11a), the removal efficiency in undrained column showed small fluctuation (at around 90%) during the three events. The removal efficiency in drained column varied greatly. The removal efficiency dropped to 54.3% at the beginning of the flow event and gradually increased to 83.2% with the increase of time. The result indicates that intermittent flow in drained condition can increase bacteria mobilization in the column.

However, this was not the case for the long-term experiment (Figure 11b). Drained condition column performed better at the beginning of restarting the events. It is possible that steel chips were oxidized when exposed to air, and the increased amount of iron oxide may have increased E. coli adsorption sites. The oxidants could be observed by the color and morphological changes in the column. The steel chips in drained column became more yellow compared with those in the undrained condition.

### **3.5 Effect of NOM and intermittent flow on E.coli desorption**

Figure 12 shows the influence of NOM on E. coli detachment from steel chips. The total amount of adsorbed E.coli after three days of adsorption (E.coli  $10^6$  MPN/mL, pH=7, flow velocity=1.77 mL/min) is  $6.7 \times 10^9$  MPN. With the presence of NOM, 0.24% and 0.43% of attached E.coli were detached from steel chips for NOM levels of 5 and 30mg/L, respectively, after 20h desorption. Similar to the short-term intermittent flow adsorption experiment results, desorption experiments showed the same intermittent flow impact. More bacteria was detached from the filter materials in the drained condition.

However, the amount of detached E.coli was less than 0.04% of the total attached E.coli after a 20 h flush.

Overall, only a small fraction of adsorbed E. coli detached from steel chips during the desorption experiments under different conditions. These results suggest that a strong electrostatic force existed between E.coli and steel chips surface, and the adsorbed E. coli were not easily released back into the solution. Therefore, steel chips are a practical material to be used as E.coli adsorption filter. They can effectively capture suspended E.coli and the E.coli cannot be detached easily, preventing a secondary E.coli pollution.

### **3.6 The steel chips performance in real stormwater**

The E.coli removal efficiency of steel chips in real stormwater are presented in Figure 13. A slight decrease (6-7%) in E. coli removal was observed when real stormwater was used to treat steel chips, in comparison to synthetic stormwater. Urban stormwater contains a variety of contaminants. The most common pollutants include TSS (0.2-940 mg/L), metals such as Cr (<710 µg/L), Cu (<1800 µg/L), Ni (<170 µg/L), Pb (<525 µg/L) and Zn (<13000 µg/L), nutrients such as NO<sub>3</sub><sup>-</sup> (<16 mg/L), and Total P (<2.3 g/L), organics such as fluoranthene (<130 µg/L), pyrene (<120 µg/L), and glyphosate (<140 µg/L); and pathogens such as Enterococci (<1-8×10<sup>5</sup>/100 mL), Coliphage (0.5-7600/100 mL), Giardia lamblia (0-7500/100 L), and Cryptosporidium (1-9.5×10<sup>5</sup>/100 mL) (Davis et al. 2001; Grebel et al. 2013; Hatt et al. 2007; Hong et al. 2006; Zhang et al. 2011). Some contaminants such as TSS can be intercepted in the columns, blocking the adsorption sites on the steel chips. Phosphate, nitrate, nitrite, arsenic, and chromium can be adsorbed onto steel chips (Alowitz and Scherer 2002;

Roberts et al. 2004; Seida and Nakano 2002). These interferences may lead to decreased E.coli removal by steel chips in real stormwater.

#### 4. CONCLUSIONS

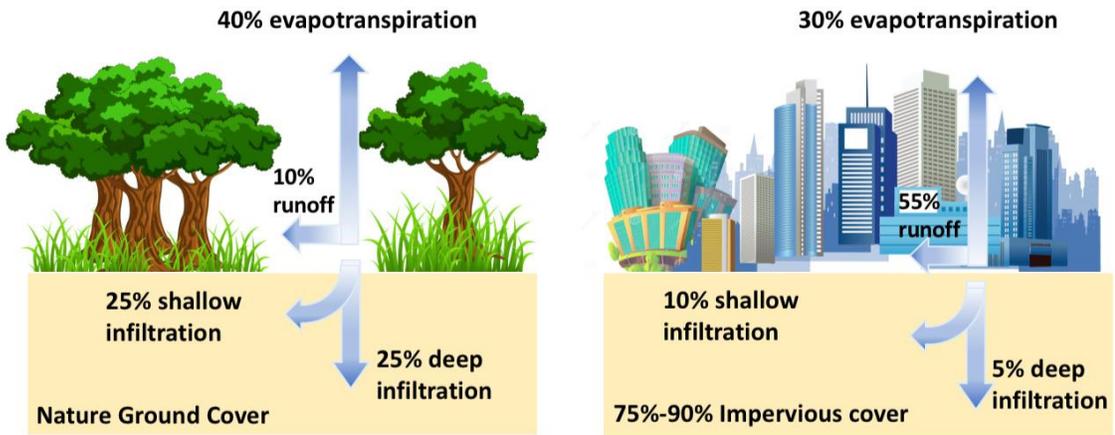
This study was designed to investigate the potential of steel chips in the removal of E.coli from stormwater. The impact of steel chips size, flow rate, initial E.coli concentration, pH, NOM, and intermittent flow on E. coli removal efficiencies of steel chips were tested. The results showed that E.coli adsorption can be greatly influenced by the size of steel chips and the concentration of co-loaded NOM. E.coli removal was decreased from 99.4% to 58.1% with an increase of steel chip size. The competitive adsorption of co-loaded NOM can inhibit E.coli adsorption by up to 80.5%. Intermittent flow can temporarily decrease E.coli removal (54.3%), but the removal efficiency can be gradually increased to 83.2% with increasing column reactor operation time. The E. coli removal efficiency by steel chips can maintain at 86.5-98.1% under different pH levels, initial E.coli concentrations and flow rates. The results of this study suggest that steel chips are effective E.coli adsorption materials that can be potentially used for stormwater treatment. In the future, field studies of steel byproduct filters are warranted to investigate their performance in real-life conditions.

**Table 1. Characteristics of steel chips.**

<b>Material</b>	<b>Size<sup>a</sup> (mm)</b>	<b>Particle</b>		<b>Packing</b>	<b>Hydraulic</b>
		<b>Density (g/cm<sup>3</sup>)</b>	<b>pH<sup>b</sup></b>	<b>Density (g/cm<sup>3</sup>)</b>	<b>Conductivity (cm/s)</b>
Steel chips	0.5-1.0	5.10	6.3	1.50	0.34
	1.0-2.0	5.20	6.3	1.35	0.87
	2.0-4.0	5.50	6.3	1.16	2.44
	4.0-8.0	5.74	6.3	0.94	2.86

<sup>a</sup> Size ranges determined by known sieve sizes

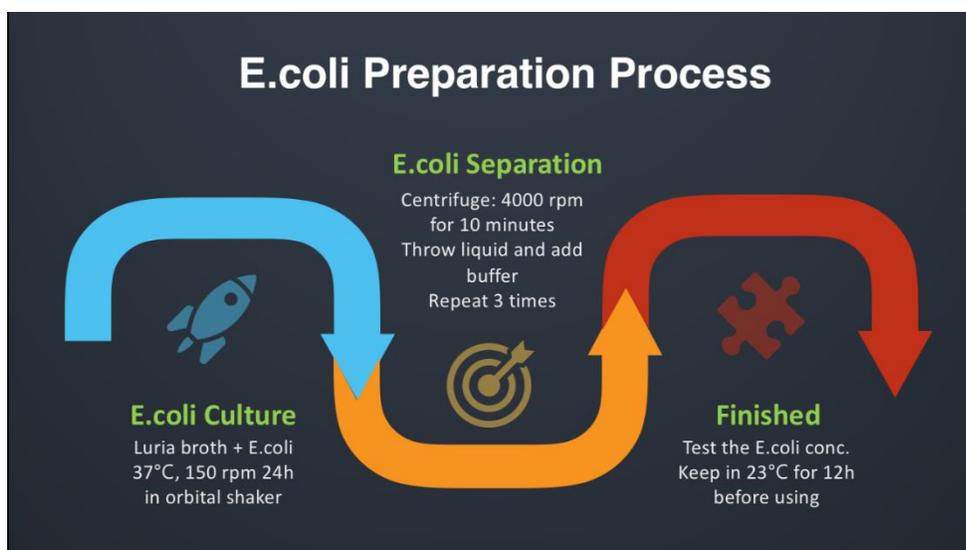
<sup>b</sup> Value of pH were obtained from a 1:1 by weight slurry mixture of material and deionized water



**Figure 1. Relationship between the impervious cover and surface runoff**



**Figure 2. Picture of different sizes of steel chips (raw materials)**



**Figure 3. E.coli preparation processes summary**



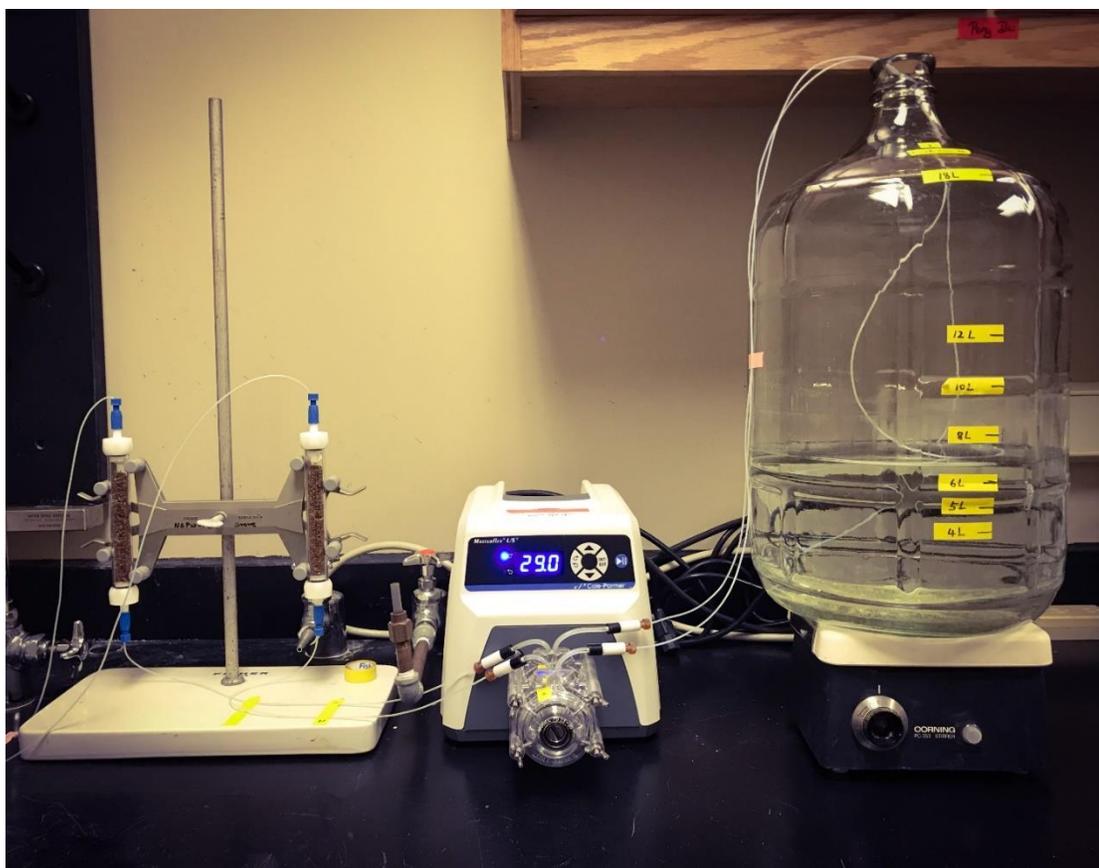
**Figure 4. Picture of E.coli concentration test kits (Colilert, tray)**



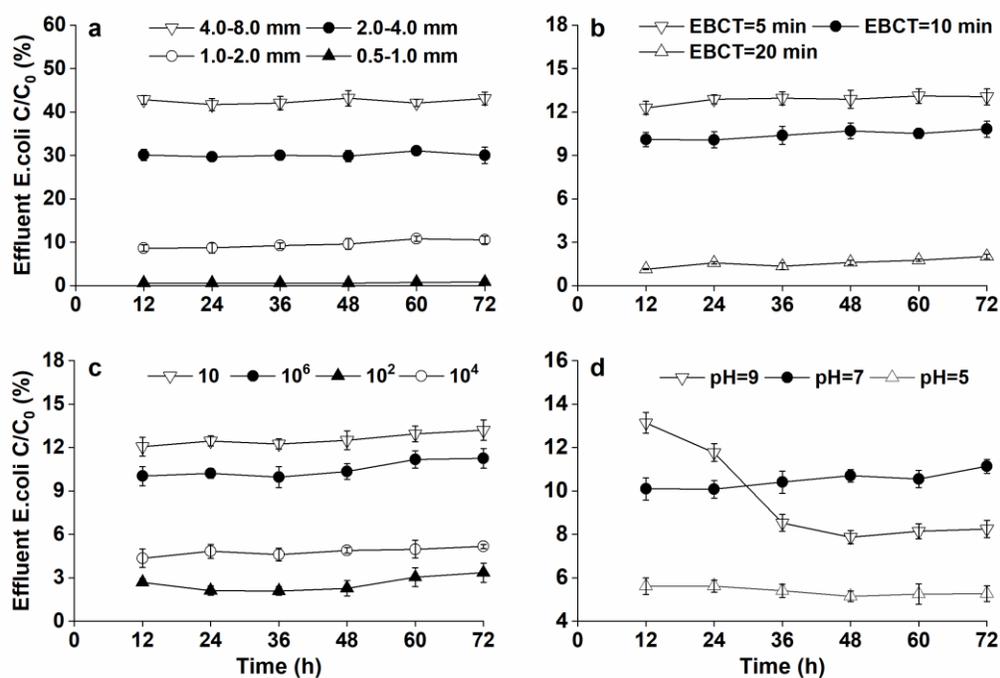
**Figure 5. Picture of SEM pre-treatment solvents.** The solvents left to right: 0.01M PBS, 2.5% GPBS, ethanol (30%, 50%, 70%, 80%, 90%, 95%, 100%), 100% TBA.



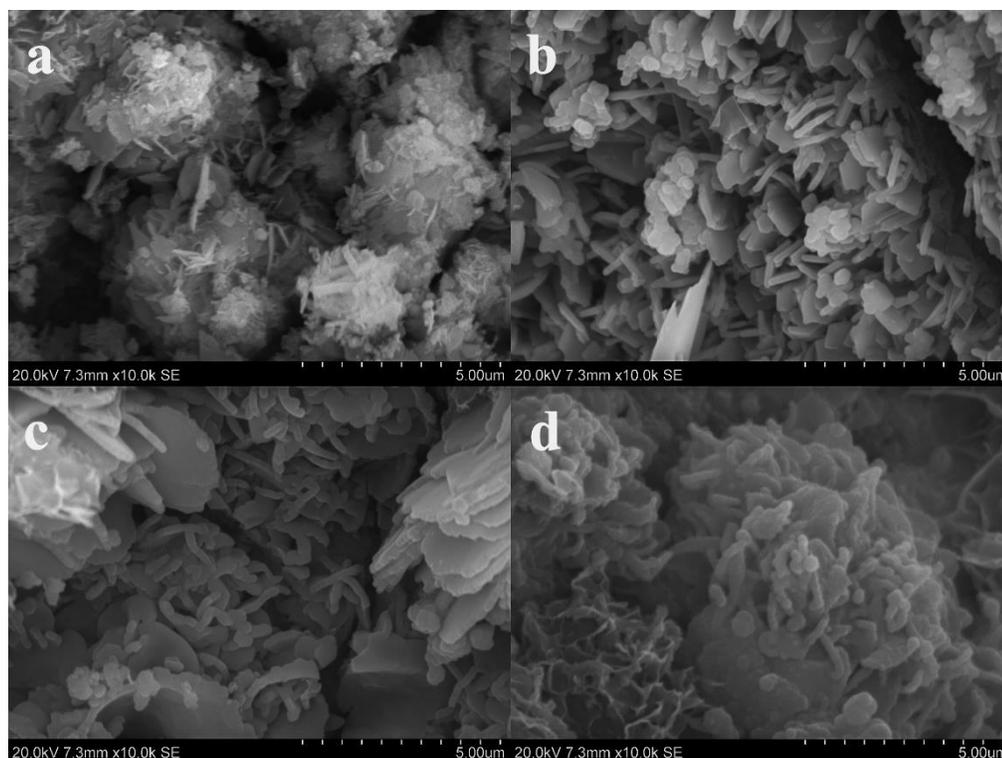
**Figure 6. Picture of the columns**



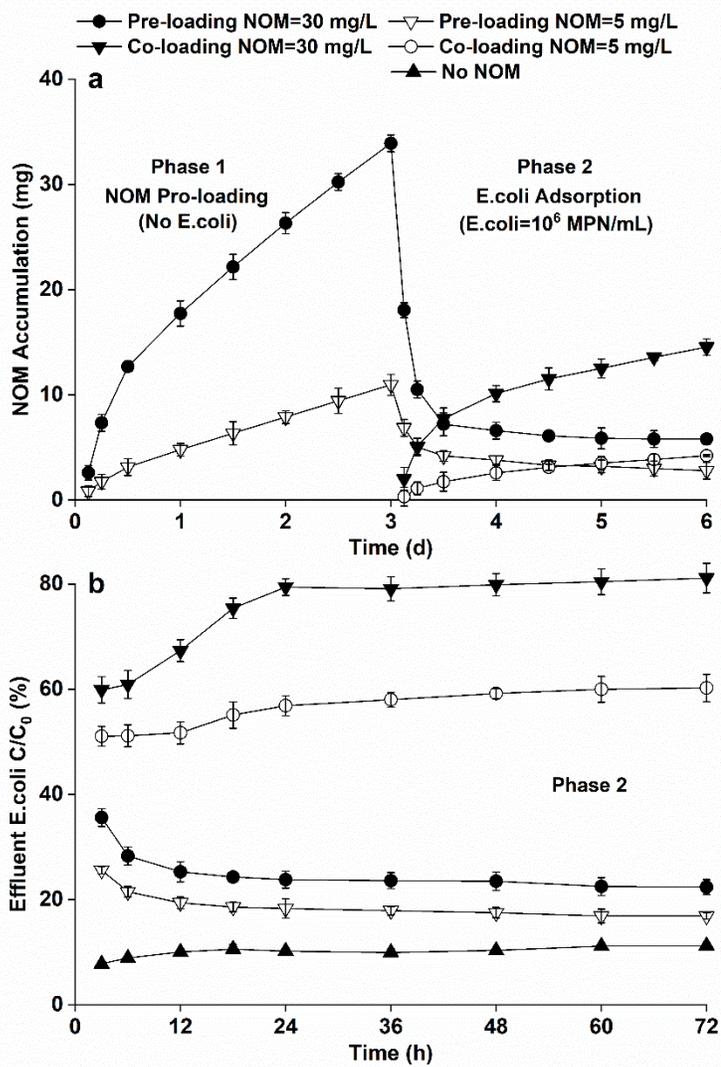
**Figure 7. Picture of the reactor**



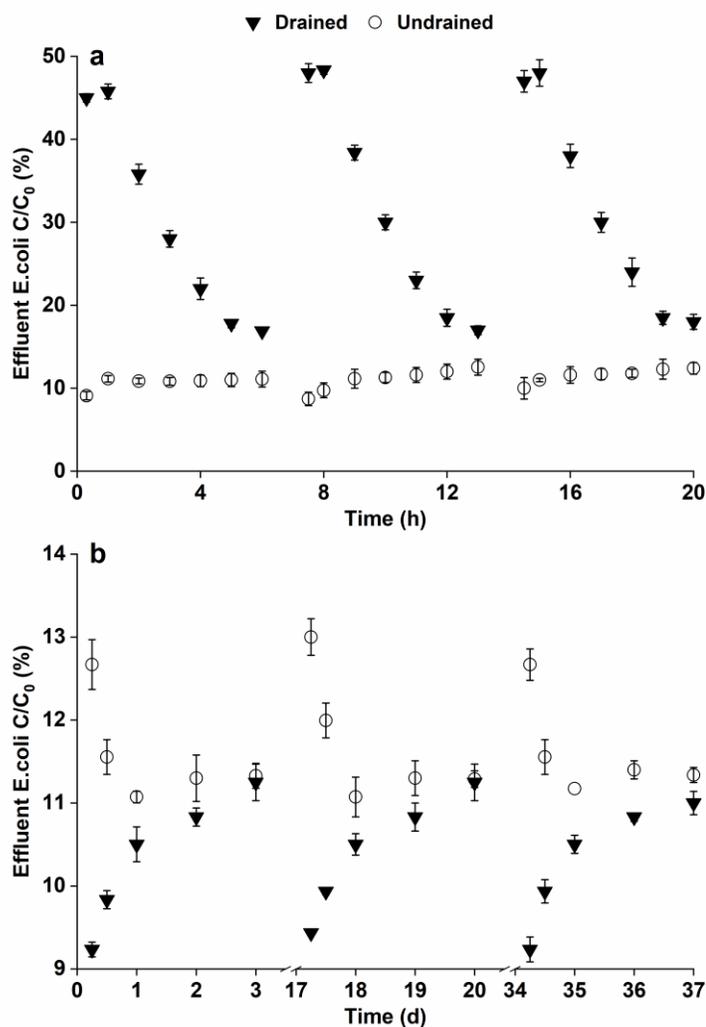
**Figure 8. Effect of different conditions for E.coli removal.**(a) steel chips size, (b) flow velocity, (c) initial E.coli concentration, (d) pH on E.coli removal. E.coli concentration in (a) (b) (d)= $10^6$  MPN/mL, steel chips size in (b) (c) (d)=1.0-2.0mm, pH in (a) (b) (c)=7.0, flow velocity in (a) (c) (d)=1.77 mL/min (EBCT=10 min). The error bar indicates standard deviation of triplicate experiments.



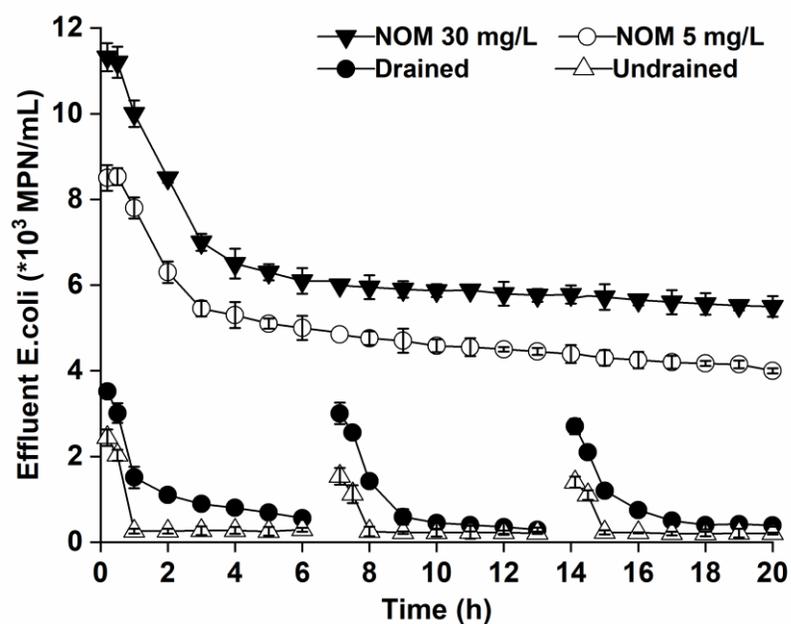
**Figure 9. SEM of steel chip surface.** (a) dry sample surface before E.coli adsorption; (b), (c), (d) shows the steel chip surface after three days E.coli adsorption experiment under different SS conditions: (b) SS without NOM; (c) SS with 5mg/L NOM; (d) 30mg/L NOM.



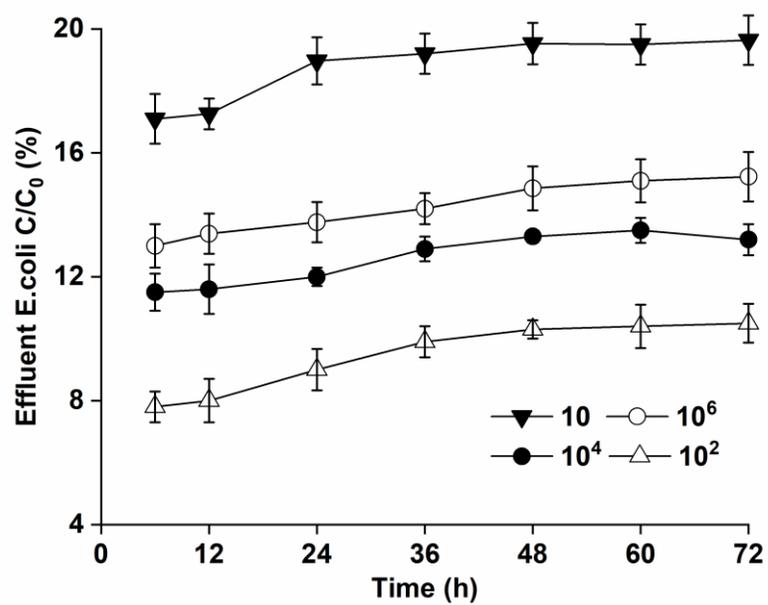
**Figure 10. Effect of NOM for E.coli Adsorption onto 1.0-2.0 mm steel chips.** (a) NOM accumulation, (b) E.coli concentration in effluent. NOM pre-loading experiments phase 2 are conducted after 432 EBV SS injection in phase 1 (NOM=5 and 30 mg/L). Experiment condition: E.coli conc.=10<sup>6</sup> MPN/mL; flow velocity=1.77 mL/min; NOM=5 and 30 mg/L in pre-loading phase 1 and co-loading phase 2, NOM=0 mg/L in pre-loading phase 2. The error bar indicates standard deviation of triplicate experiments.



**Figure 11. Effect of intermittent flow with wet-dry (undrained-drained) cycle on E.coli removal. (a) short-term (b) long-term.** Experiment condition: E.coli conc.= $10^6$  MPN/mL; flow velocity=1.77 mL/min; interval time in (a)=1h, (b)=14d. Drained and undrained indicate the column saturated condition during the 1h pause. (a) and (b) were conducted 3 days E.coli adsorption (total adsorbed E.coli was  $6.7 \times 10^9$  MPN), separately. The error bar indicates standard deviation of triplicate experiments.



**Figure 12. Effect of NOM and intermittent flow on E.coli desorption.** Experiment condition: E.coli conc.= $10^6$  MPN/mL; flow velocity=1.77 mL/min; interval time in undrained and drained condition=1h. The desorption experiments were conducted after 3 days E.coli adsorption (total adsorbed E.coli= $6.7 \times 10^9$  MPN). The error bar indicates standard deviation of triplicate experiments.



**Figure 13. E.coli removal in real stormwater.** Experiment condition: flow velocity=1.77 mL/min. The error bar indicates standard deviation of triplicate experiments.

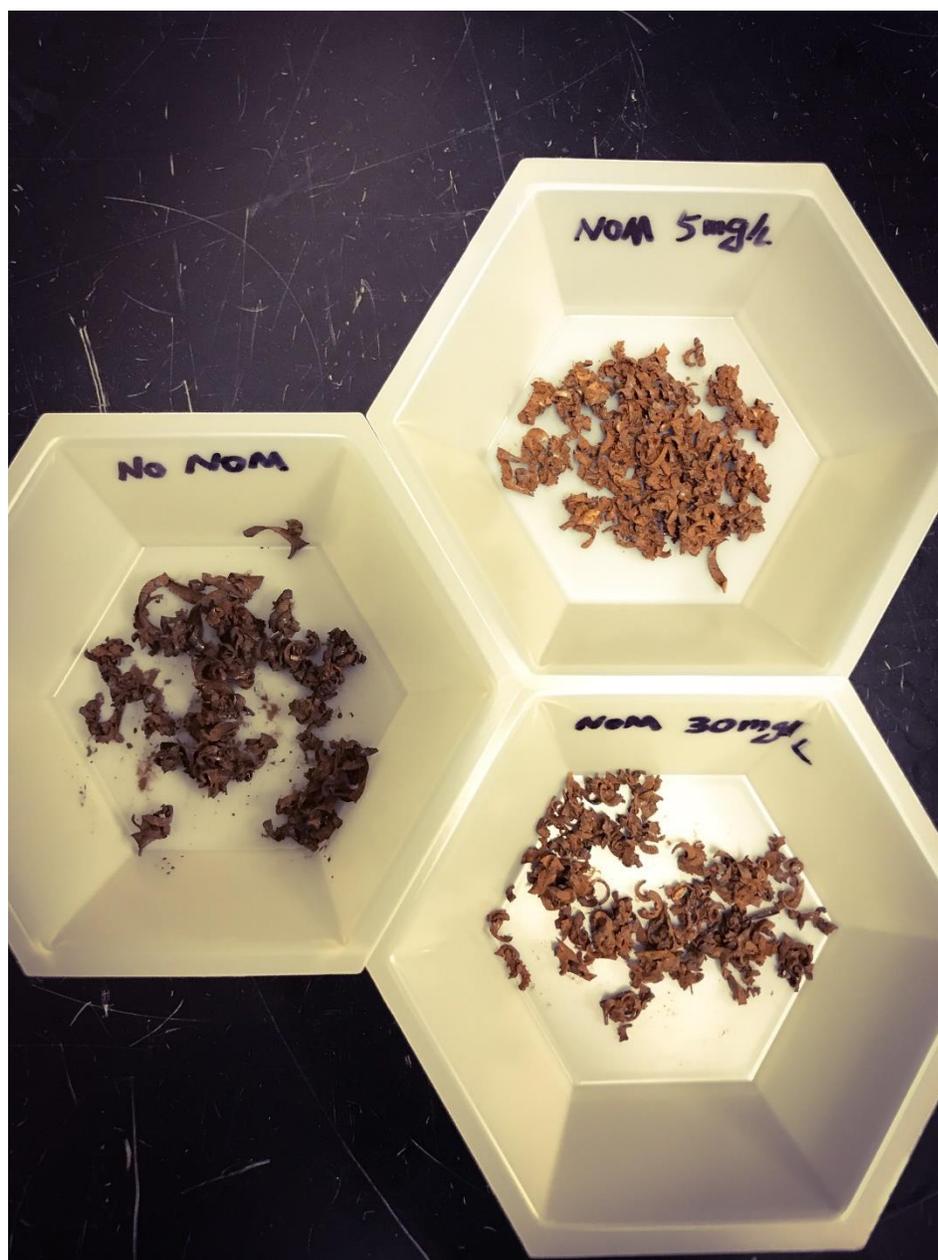


Figure 14. Picture of 2.0-4.0 mm steel chips after 3 days E.coli adsorption

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