# Estimating bacterial decay in the Río de la Plata River

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**Abstract**
This paper estimates the time for a bacterial population density to decay to 90% of its initial value ($T_{90}$), for the inner region of the Río de la Plata River. A tidally influenced fluvial system with high wind influence constitutes the inner region of the Río de la Plata River. Its physical and chemical properties include: full vertical mixing, high suspended particulate matter (SPM) producing small secchi depths (20-25cm), a turbidity range between 50-400NTU, high Dissolved Organic Matter (DOM), Dissolved Organic Carbon that ranges between 5-10mg/l, and low salinity levels.

Agua y Saneamientos Argentinos S.A. (AySA SA) is a state Company that renders water and sanitation services in the city of Buenos Aires and 17 municipalities of the metropolitan area of Buenos Aires. A large portion of the treated wastewater is discharged into the inner Río de la Plata River through the Berazategui outfall. This treated wastewater discharge causes localized degradation in water quality. Therefore, accurately modeling bacterial decay within the river is a design parameter of Buenos Aires metropolitan area outfalls to preserve water quality for any use.

From 2007 onwards, *in situ* measurements and samples were collected in the Berazategui outfall plume for bacteriological and chemical analysis purposes. Samples were taken either at night or daytime, to evaluate the effect of solar radiation on bacteria survival. The starting point was at Berazategui outfall on rising tides only; samples were taken upstream every hour tracking a released surface drifter. Biological and chemical parameters were measured at AySA SA's laboratory. Bacteria (measured using E. coli as an indicator), chlorides, conductivity, and alkalinity data are presented on a semi-log plot versus elapsed time. The data were fitted with nonparametric linear regression equations. The reduction rates for bacteria and the physical dilution rates for the remaining parameters were obtained from the slopes of their respective regression equations.

The decay rate and $T_{90}$ were estimated using a first-order kinetic model. No significant differences in $T_{90}$ were observed between night and daytime. Model results show Escherichia coli reaching 10% of its initial population density in less than 6 hours for the inner region of the Río de la Plata River.

**Keywords**
Río de la Plata River, Berazategui outfall, bacterial decay, Escherichia coli, T90.

## INTRODUCTION

### Study site

The area surveyed is the inner region of the Río de la Plata River which is termed as tidally-river; even if it is not influenced by marine waters, it is dominated by tidal currents.

The Río de la Plata River is located in the lower La Plata basin (Figure 1), on the East coast of South America, approximately between 34° and 36° South Latitude and 54° and 58° West Longitude, forming the border between the República Oriental del Uruguay and the República Argentina. La Plata basin encompasses 3.1Mkm$^2$, and over 80 percent of the Argentinean inhabitants rely on the watershed for water supply. The river water stands for both the main source of water supply for approximately 11M inhabitants of Buenos Aires and its metropolitan area, and the site of municipal and industrial effluent water discharge throughout sub-aquatic outfall and rainwater drainage.

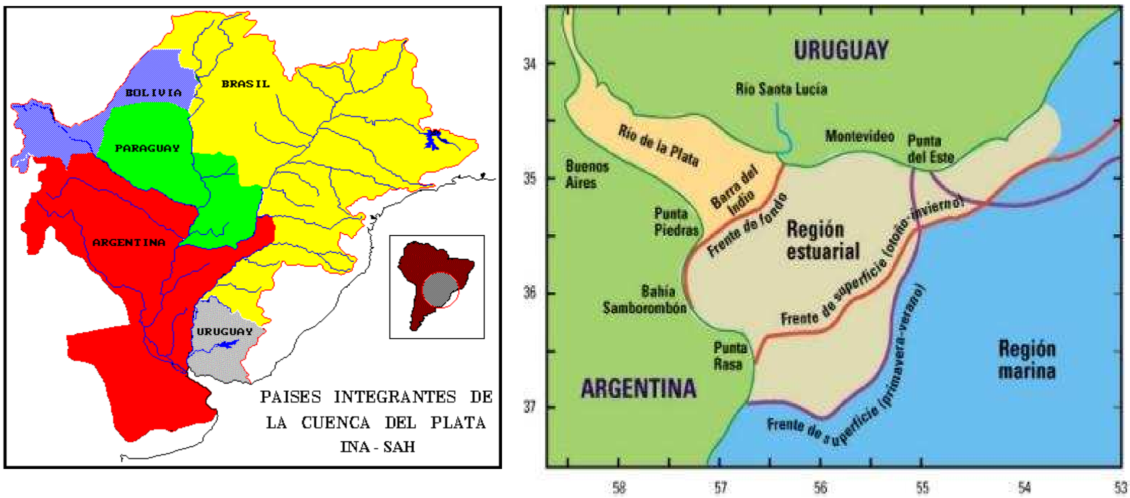


Figure 1. La Plata basin (left) and Río de la Plata (right)

The Río de la Plata River can be divided into two main regions (Figure 1), inner and outer, separated by a line called Barra del Indio that extends from Punta Piedras on the Argentinean coast to Montevideo city in Uruguay. Each region has distinct characteristics, especially depth and marine waters influence. Its main tributaries are the Uruguay River and the Paraná River, with mean discharges of about 6,000 and 16,000m$^3$/s, respectively. Given its 30,000km$^2$ of surface, during maximum discharges (fall and spring seasons) the river assimilates these alterations and there are no significant changes on the water level. In addition, there are minor contributions from several small tributaries along the Argentinean coast and the discharge from the Santa Lucia River near Montevideo city.

Even though the inner region of the Río de la Plata River is dominated by tidal currents, its originating tributaries (Paraná de las Palmas, Paraná Guazú-Bravo and Uruguay Rivers) generate water currents towards the ocean (Balay, 1961). These *drift currents* flow in three separate corridors (Figure 2) along the river namely, from South to North, Palmas, Guazú and Uruguay corridors, respectively. In terms of water quality, the inner region of the Río de la Plata River on the Argentinean coast is particularly and highly influenced by the Paraná de las Palmas waters (Simionato *et al*, 2009). Additionally, urban runoff from Buenos Aires city and its Metropolitan area directly impacts on the surroundings of the area surveyed; Jirka and Bleninger, 2004 summarized these worldwide coastal water quality problems. This area is called *Franja Costera Sur* and it extends from the shoreline to approximately 10,000m into the river, between San Fernando and Magdalena (Figure 3). Thus, water quality improves within the river while this coastal impact decreases and the Paraná River waters become dominant.

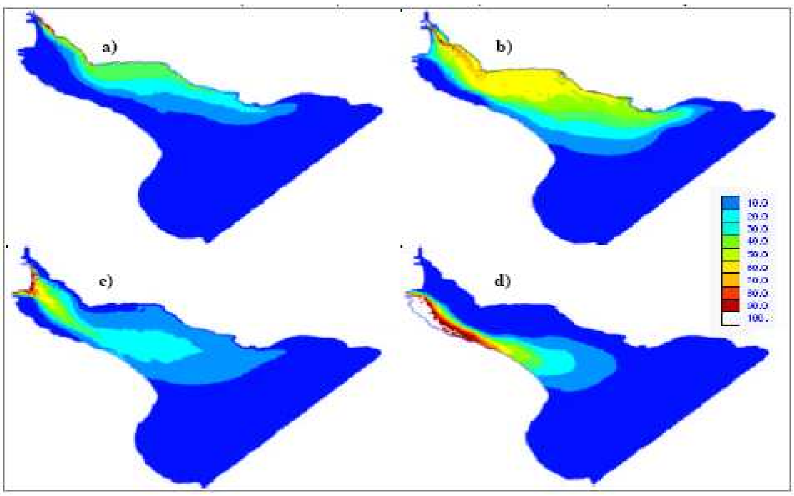


**Figure 2.** Iso-concentrated plots for substances discharged in 4 points at the headwaters of Río de la Plata River (running scenarios: spring-summer seasons) a) Uruguay River b) Paraná Bravo-Sauce River c) Paraná Guazú River d) Paraná de las Palmas River. After Piedra-Cueva, 2001.

## Berazategui outfall

The sewer-system is separated from the rainfall canalization and discharges an average effluent flow rate of about 22m$^3$/s with a maximum peak discharge of 33.5m$^3$/s. The Berazategui outfall, located at 34° 44' 41'' South Latitude and 58° 10' 3'' West Longitude (Figure 3), starts at the charging chamber on the onshore headwork, from where a feeder tunnel conveys the effluent to the 2300m long pipe on the disposal area, almost perpendicular to the coast. This conduit is on the bottom of the river (distance 4 to 6m downward approximately) and it has 10 diffusers along the last 100m; diffusers are cone arranged with 1.6m diameter.

Tidal currents dominate the velocity field (maximum velocities during tidal cycle $v_{max,}$ are about 0.3 to 0.6m/s), also there is full vertical mixing all over the area surveyed.

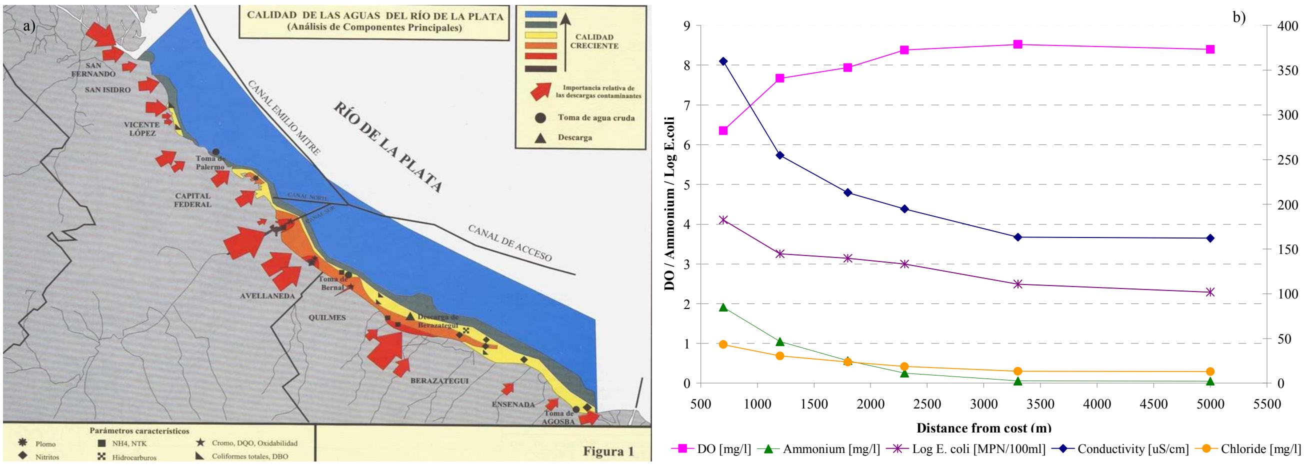


**Figure 3.** "Franja Costera Sur": a) location of runoff discharges along the coast, water intakes and Berazategui outfall discharge, and b) Plot of mean values of water quality parameters vs distance from the coast at Berazategui outfall surroundings.

## Bacterial decay, $T_{90}$

Fecal bacteria are non-conservative elements. The decay rate in aquatic environment depends on the bacterium itself (species, strain and physiological status), on physical and physicochemical characteristics encountered in the environment (sedimentation, photo-oxidation, temperature, salinity, hydrological conditions, pH) and on biochemical-biological factors (organic matter

content, presence of fecal matter, predation, viruses, competition; Chamberlin and Mitchell 1978; EPA 1985; Rozen and Belkin 2001).
Bacterial decay is usually quantified by $T_{90}$, which is the time for a bacterial population density to decay to 90% of its initial value. $T_{90}$ constitutes an important parameter for water resources conservation as for outfalls design. Usually, population dynamics for allochtone bacteria are modeled by first-order die-off kinetics, proposed by Chick (1908), because of its simplicity and success in modeling bacterial die-off in aquatic environments. However, in practical applications microorganism die-off rarely follows the Chick law and there may be a time delay before the decline in bacterial population starts (Wilkinson et al, 1995). Actually, data from enteric bacteria survival are not easily comparable, since adequate models or parameters do not exist (Gonzalez, 1995).

**Purpose**
The purpose of this paper is to analyze the field data to estimate bacterial decay within the river, particularly *E. coli* die-off, and to perform numerical modeling of the fecal bacteria present in the Berazategui outfall plume. In support of these aims, a particular field campaign has been clearly outlined. Furthermore, Pommepuy et. al. 2006, emphasizes in situ data collection for the estimation of bacterial behavior in a specific area and not only considering data from literature.

## MATERIALS AND METHODS

The technique used to estimate $T_{90}$ is *in-situ measurement in existing wastewater discharge* (Salas, 2000; Roberts et al, 2010). Measurements and samples were collected in the Berazategui outfall plume for bacteriological and chemical analysis. Samples were taken either at night or daytime, to evaluate the effect of solar radiation on the survival of bacteria. The sampling starting point was at Berazategui outfall on rising tides only and continued upstream tracking a released GPS-equipped surface drifter (Figure 4).


**Figure 4.** GPS-equipped drifter released on Berazategui outfall plume for T90 estimation.

**Sampling & Analyses**
Samples were collected on board the *Orion* Laboratory Ship, pumping water 0.7m deep under the water line, every hour tracking drifter's trajectory along the plume. The pump is located inside the ship; this device also feeds the online measuring equipment. The drifter was tracked until tide reversed its direction. Commonly, the drifter follows the tide covering distances up to 6km in 6 hours with velocities ranging between 0.05 and 0.4m/s. The starting point of the sampling was the Berazategui outfall (2300m away from coast); raw Berazategui sewage was sampled at the same time.

Even though between 2007 y 2011 eight campaigns were conducted, only four of them were considered for this study. In order to calculate the time taken for *E. coli* to die off 90% of its initial density, it is essential to avoid other sources of contamination. The main issue, to discard or to consider results from a field campaign, is the direction of the tide. In addition, aborted samplings, ever since weather turned risky for sailing, are considered non-representative data for the calculation of the *E. coli* die-off. Measurements were also discarded when the wind blew in East and South-East direction and forced the drifter to approach the coast as per the difficulty to calculate the physical dilution rate of the outfall plume. In addition, measurements on ebb tides were later excluded from the analysis given the interference of Punta Colorada coastal zone (Figure 5). As the propagation of the tidal wave upstream along the Río de la Plata River manifests as water level and velocity oscillations, mainly during ebb tides, coastal contaminated waters are carried within the river and extended to the Berazategui outfall plume. For this purpose, field experiments and collected samples were performed during rising tides only; additionally, coastal impact concentrates on the coast and does not affect the behavior of the Berazategui outfall plume. Valid measurements correspond to the months of April, May and October and March.

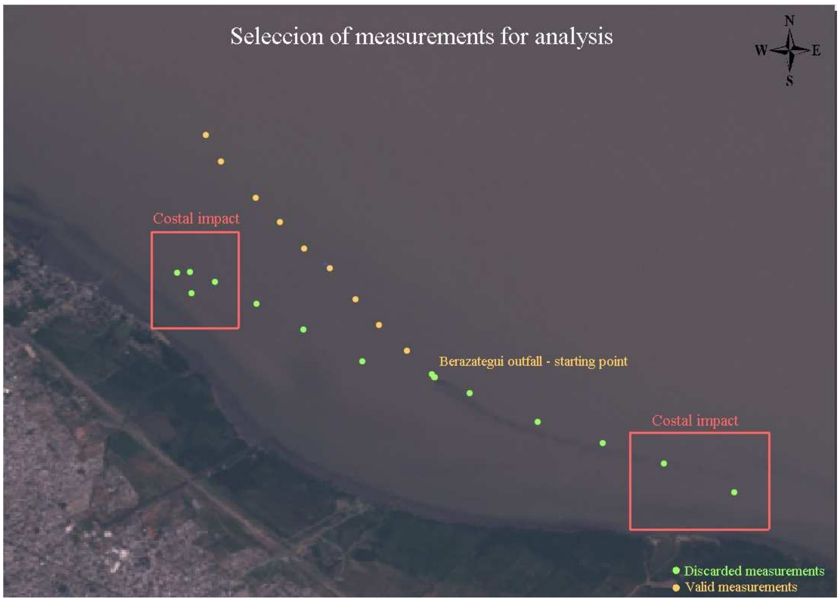


**Figure 5.** Example of discarded (green) and valid measurements (yellow) collected on field campaigns for T90 estimation.

Conductivity, temperature, pH, dissolved oxygen and turbidity were measured on board with online metering equipment (Table 1). Samples for chemical and bacteriological analysis were taken; *Escherichia coli* and coliforms were sampled in duplicates. Rino HDX 130 GPS recorded position and time and transmits it to a central location. Samples were labeled and stored at 4°C; once field campaigns were completed, they were analyzed according to standarized methods certified by ISO 17025 at AysA's Central Laboratory (*ACL)*.

Table 1. Online water quality parameters measured on Orion Laboratory Ship during field campaigns for T90 estimation.

| Parameter | On-line Measuring Equipment |
|---|---|
| Conductivity | Endress & Hausen CLS50-A1A2 |
| Dissolved oxygen | Endress & Hausen COS4 |
| pH | Endress & Hausen CPS11-2AA2ESA |
| Temperature | Endress & Hausen CLS50-A1A2 |
| Turbidity | Endress & Hausen CUS31 |

Parameters measured at ACL are: Total Coliforms, *Escherichia coli*, conductivity, alkalinity, turbidity, pH, total suspended solids, total phosphorus, total orthophosphate, chloride, nitrate, sulfate, calcium, magnesium, sodium, potassium, ammonium, oxidability, biological oxygen demand, total chromium and total lead. The quantification methods of parameters used for $T_{90}$ calculations are listed in Table 2. Analyses were performed less than 10h after the sampling. Previous experiments show no shift in bacterial density within this delay.

Table 2. Applied quantification methods of parameters for T90 estimation.

| Parameter | Quantification method | Reference |
|---|---|---|
| *Escherichia coli* | Chromogenic substrate | SM 9223 A y B Ed. 20 |
| | Multiple tube | SM 9221 F E. 20 |
| Conductivity | Conductimetry | SM 2510B Ed. 20 |
| Chloride | Ion chromatography | SM 4110B Ed. 20 |

**Decay model & Statistical analysis**

*Escherichia coli* was chosen to obtain the decay rate and $T_{90}$, because of its broad use as a tracer for fecal contamination. Decay rate, $k_m$ and $T_{90}$ were estimated using a first-order kinetic model (Chick's law, 1906), i.e. an exponential decay of *E. coli* concentration, *N* with time, *t* (equation 1).

$$\frac{\partial N(t)}{\partial t} = -k_m N(t) \quad \textbf{(1)}$$

Where, $k_m > 0$ and is expressed in $[h^{-1}]$.

Since the decay rate only encompasses disappearance of *E.coli* by death, leads from the difference of the total loss, $k_T$ and physical dilution rates, $k_d$ (equation 2). It is assumed that decay and dilution are the governing processes for reduction of bacterial concentration.

$$k_m = k_T - k_d \quad \textbf{(2)}$$

The decay coefficient is often replaced by $K = k_m/2.3$, which corresponds to the use of decimal logarithm for bacterial counts, as shown in equation 3.

$$\log N(t) - \log N(t_0) = -K(t - t_0) \quad \textbf{(3)}$$

K is thus the inverse of the period of time $T_{90}$ necessary to achieve the bacterial population 10% of its initial value.

To obtain the loss rate constant, mean values of *E. coli* at each sampling site were calculated and plotted on semi-ln plots against elapsed time. To estimate the physical dilution rate, chlorides and conductivity semi-ln plots were made. Although conductivity is not strictly conservative, it was taken into consideration because of the fairly short duration of the sampling time. Conservative substances and *E.coli* semi-ln plots were fitted with linear regression equations. Regression slopes were obtained using single Kendall-Theil Robust Line (KTRL) nonparametric model; the slope of KTRL is found by comparing each data point with all others in a pair-wise fashion and computing slopes. Therefore n(n-1)/2 slopes are obtained with *n* as the number of data points. This model was chosen because it does not depend on the normality of residuals for validity of significance test, and will not be strongly affected by outliers contrasting ordinary least squares regression (Helsel & Hirsch, 2002). Software to fit lines was obtained from the USGS website (Granato, 2006).

## RESULTS

Between 2007 and 2011, a total of eight field campaigns were carried out; four of them were valid for $T_{90}$ calculation corresponding to April and May, 2007 and March, 2011 for daytime, and to October, 2010 for nighttime. Results are shown in Table 3.

Table 3. Results of each sampling campaign for T90 estimation.

| Month / Year | Elapsed time (h) | Distance from outfall (m) | Total Coliforms (NMP/100ml) | E. coli (NMP/100ml) | Conductivity (uS/cm) | Chlorides (mg/l) | Ammonium (mg/l) | Dissolved oxigen (mg/l) | Temperature (°C) |
|---|---|---|---|---|---|---|---|---|---|
| April / 2007 DAY | 0.00 | 0 | 13960000 | 2090000 | 512 | - | 7.75 | 2.39 | 20.5 |
| | 0.26 | 1132 | 18210000 | 3200000 | 497 | - | 9.25 | 1.7 | 20.3 |
| | 0.53 | 2183 | 9436000 | 2076000 | 329 | - | 4.00 | 2.03 | 20.2 |
| | 0.78 | 3361 | 4913000 | 951000 | 316 | - | 2.80 | 2.02 | 20.0 |
| | 1.00 | 4303 | 2419600 | 721500 | 293 | - | 2.40 | 4.38 | 19.9 |
| | 1.51 | 5138 | 2202000 | 1080000 | 331 | - | 3.00 | 4.03 | 19.8 |
| | 2.00 | 5862 | 2419600 | 675000 | 274 | - | 1.60 | 6.83 | 19.6 |
| | 3.17 | 6488 | 631000 | 128150 | 221 | - | 0.39 | 6.84 | 19.5 |
| | 4.18 | 6599 | 30300 | 5390 | 211 | - | 0.10 | 5.54 | 19.5 |
| May / 2007 DAY | 0.00 | 0 | 8146000 | 1056500 | 460 | 50.00 | 3.95 | 2.79 | 16.4 |
| | 0.50 | 632 | 1121000 | 429000 | 326 | 38.00 | 2.87 | 6.03 | 15.3 |
| | 1.00 | 1255 | 509500 | 199000 | 313 | 35.00 | 1.97 | 4.52 | 15.2 |
| | 1.29 | 1825 | 720900 | 163200 | 306 | 33.00 | 1.82 | 4.62 | 15.1 |
| | 1.86 | 2419 | 485050 | 175000 | 310 | 33.00 | 1.82 | 4.8 | 15.1 |
| | 2.29 | 3009 | 476200 | 154050 | 298 | 32.00 | 1.50 | 6.51 | 15.1 |
| | 2.80 | 3575 | 393300 | 196380 | 276 | 30.00 | 1.10 | 6.60 | 15.0 |
| | 3.37 | 4165 | 98520 | 36220 | 231 | 26.00 | 0.36 | 7.96 | 14.9 |
| | 4.27 | 5007 | 3391 | 756 | 218 | 24.00 | 0.03 | 7.96 | 14.8 |
| | 4.34 | 5504 | 3116 | 232 | 218 | 24.00 | 0.03 | 7.59 | 14.9 |
| October / 2010 NIGHT | 0.00 | 0 | 8752500 | 1985000 | 862 | 88.50 | 19.2 | 3.02 | 18.6 |
| | 1.00 | 1102 | 5135500 | 1492000 | 505 | 54.40 | 11.5 | 3.85 | 18.2 |
| | 2.00 | 2151 | 696500 | 328150 | 462 | 52.10 | 1.70 | 6.1 | 17.5 |
| | 3.00 | 2989 | 6510 | 1335 | 437 | 48.30 | 1.50 | 5.75 | 17.4 |
| | 4.00 | 3714 | 8575 | 1300 | 455 | 50.20 | 1.60 | 5.28 | 17.4 |
| | 5.00 | 3857 | 5695 | 1185 | 488 | 54.40 | 1.50 | 4.98 | 17.5 |
| March / 2011 DAY | 0.00 | 0 | 1516300 | 1050150 | 632 | 68.40 | >10 | 0.18 | 26.0 |
| | 1.00 | 1070 | 169995 | 112475 | 375 | 34.50 | 7.30 | 3.44 | 25.4 |
| | 2.00 | 2642 | 198635 | 100985 | 303 | 25.60 | 0.64 | 5.03 | 23.7 |
| | 3.00 | 4007 | 9060 | 2575 | 276 | 25.00 | 0.08 | 5.86 | 23.7 |
| | 4.00 | 4865 | 12060 | 2370 | 276 | 25.20 | <0.05 | 5.27 | 23.8 |
| | 5.00 | 5382 | 4495 | 465 | 278 | 25.80 | <0.05 | 5.1 | 24.0 |

*E. coli* density (ln transformed) was plotted against elapsed time for each campaign (Figure 6a). As expected, ln *E. coli* curves were almost linear. Semi-ln plots of conductivity and chlorides are presented in Figure 6b.

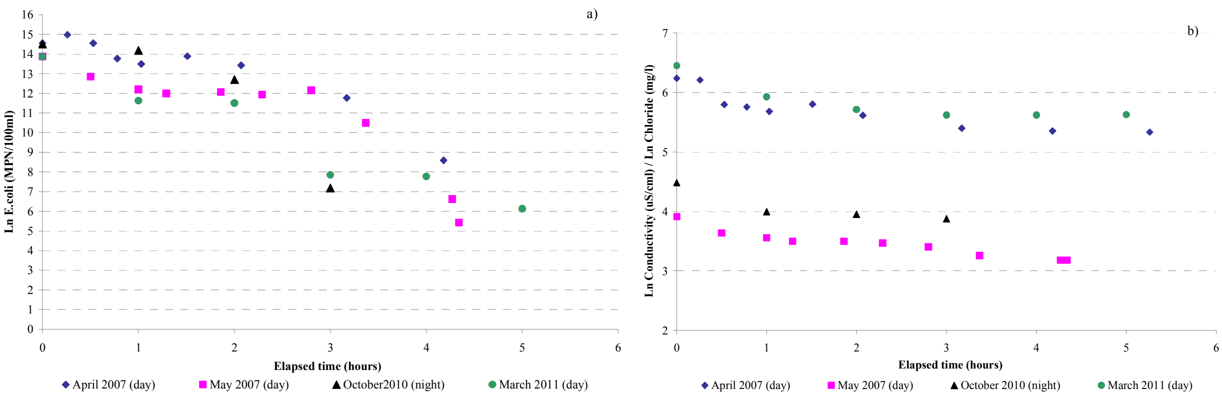


**Figure 6.** a) E. coli density (MPN/100ml) data from the different field campaigns b) Conductivity (□S/cm) data from April and March, and Chlorides (mg/l) data from May and October. Symbols: ◆ April, 2007 daytime, ■ May, 2007 daytime, ▲ October, 2010 nighttime, ● March, 2011 daytime.

In order to calculate the decay rate coefficient, $k_m$ semi-ln plots were performed by linear regression of measured data. Regression slopes, determined by Kendall-Theil Robust Line (K-TRL) nonparametric model, are summarized in Table 4 as well as calculated decay rates and $T_{90}$.

**Table 4.** Summary of estimated loss and dilution rates by linear regression using Kendall-Theil Robust Line nonparametric model and calculated decay rates and T90 for Río de la Plata River River.

| Month of sampling campaign | Loss rate, $k_T$ ($h^{-1}$) | Dilution rate, $k_d$ ($h^{-1}$) | Decay rate, $k_m$ ($h^{-1}$) | $T_{90}$ (h) |
|---|---|---|---|---|
| March | 1.542 | 0.102 | 1.44 | 1,60 |
| April | 1.426 | 0.164 | 1.262 | 1,82 |
| May | 1.181 | 0.125 | 1.055 | 2,18 |
| October | 1.797 | 0.139 | 1.658 | 4,40 |

Daytime first-order decay rates for *E.coli* in the inner region of Río de la Plata River ranged from 1.055$h^{-1}$ to 1.44$h^{-1}$, deriving in $T_{90}$ ordering between 1,6 to 2,18h. For the nighttime campaign, the decay rate is slightly larger than daytime's and $T_{90}$ doubles the times calculated in daytime campaigns. According to calculation, Figure 7 shows decay curves for *Escherichia coli* in the Río de la Plata River.

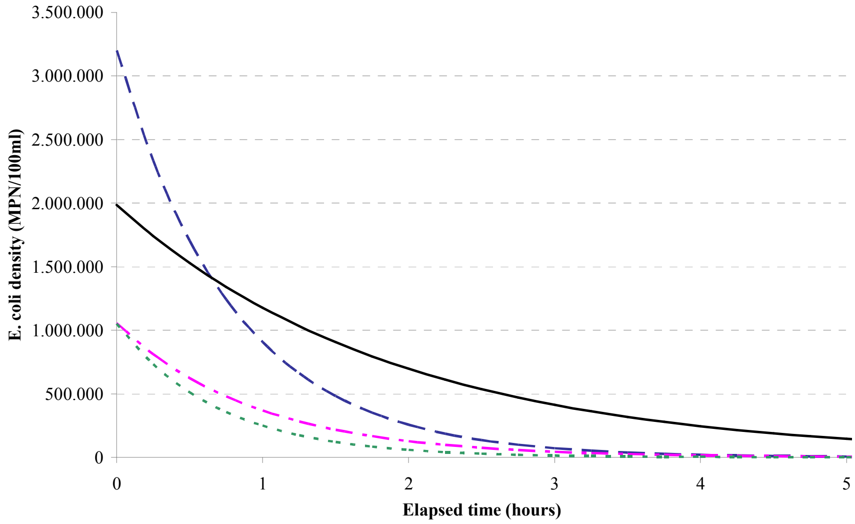


**Figure 7.** Decay curves of *E. coli* in Río de la Plata. Symbols: ---- April, 2007 daytime, ---- May, 2007 daytime, —— October, 2010 nighttime, ---- March, 2011 daytime ----

## CONCLUSIONS

According to USEPA (1985), $k_m$ values vary between 0.005 $h^{-1}$ and 1.1 $h^{-1}$ in rivers (resulting 2 h < $T_{90}$ < 7d). Moreover, local references (Menendez, EIH and Serman) propose $T_{90}$ varying from 45 to 84h. Regardless those values, this study found lower times for *E.coli* population to reduce 90% of its initial value in the inner region of Río de la Plata River River. Given the technique used to estimate $T_{90}$ (*in-situ measurement in existing wastewater discharge)* these preliminary but conclusive results show periods fewer than 6h length; between 2 to 4h, approximately.

The disappearance rates reported in this study are useful as design parameters for subaquatic sewer systems and to predict its water quality impact. Current Berazategui outfall location, with initial dilution rates around 1:2 - 1:2.5, arises an approximately *E.coli* concentration of 20,000MPN/100ml at about 3 to 4km upstream. Thus, and for greater initial dilution rates ranging 1:20 to 1:40, future location of Berazategui outfall could predict same concentration in around 0.8 to 1.5km away from sewer discharge.

Many studies suggested that incident light levels and temperature are the main factors affecting coliform disappearance rates. In this study $T_{90}$ calculated for night is greater than $T_{90}$ obtained for daytime, further, as temperature decreases $T_{90}$ increases. To confirm this trend correlation studies will be made when more results become available.

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