UNIVERSITY OF THE REPUBLIC OF URUGUAY
FACULTY OF AGRONOMY
GRADUATE PROGRAM

# INITIAL EFECTS OF AFFORESTATION ON RUNOFF WATER QUALITY IN A BASIN OF TACUAREMBO RIVER

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# I. INTRODUCTION GENERAL

## ANTECEDENTS

In the last years, the traditional land use map has changed in some areas of Uruguay, due to the development of the forest sector which has been promoted by the national government through specific laws (Forest Law Nº 15.239, December of 1987). The registered increment in the forested area between 1990 and 2004 was from 45.000 to 750.000 has (Silveira et al., 2006). Contrary to other regions, the 474,076 ha planted with eucalypts and the 190,033 ha planted with pine (MGAP, 2005) have been established on soils with natural vegetation of prairies previously dedicated to extensive livestock production.

In our country the forest plantations are concentrated mainly on the Departments of Río Negro, Paysandú, Rivera and Tacuarembó (Figure 1). In this last department the company Weyerhaeuser Uruguay (before Colonvade CORP. and Los Piques CORP.) are the most important in terms of forested area, with a total of 120.000 hectares of plantations, 70% with pine and 30% with eucalypts, where the timber is destined to solid woods and laminated boards.

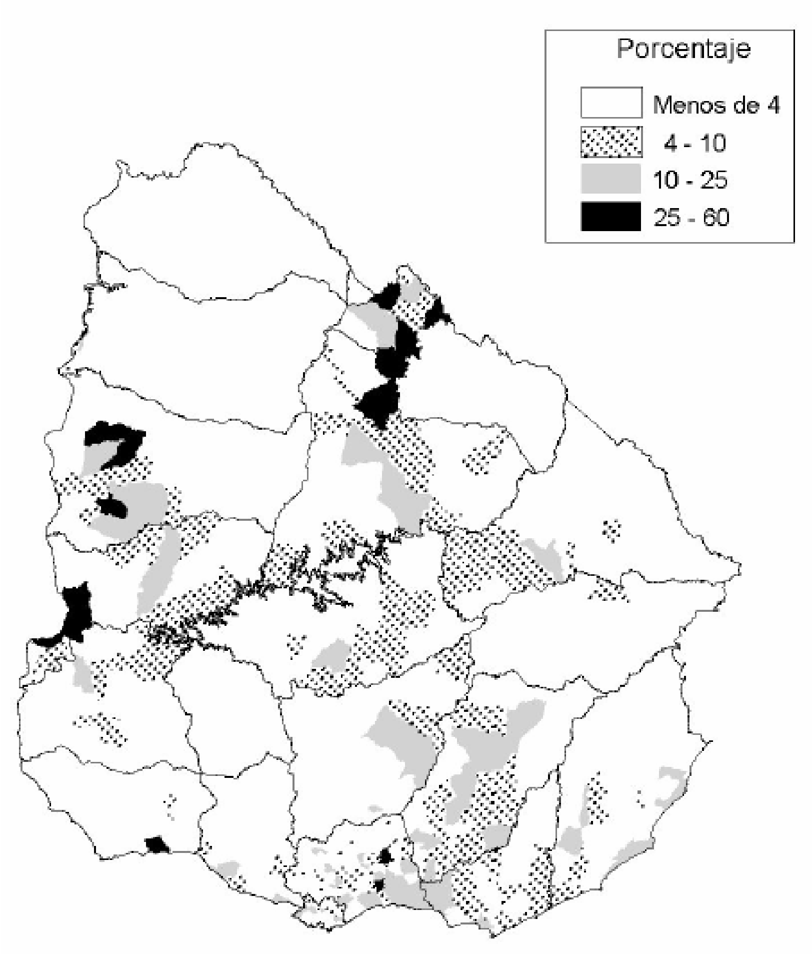


Figure 1. Proportion of areas with forest plantations in Uruguay. Adapted from MGAP-DIEA, Agricultural Census of the year 2000.

Starting from the year 1999, Weyerhaeuser Uruguay began a study to evaluate the effect of land use change from natural-prairies livestock production to forest on the sustainability of natural resources in the Department of Tacuarembó. This study was installed on a Pine plantation located in a watershed of the Tacuarembó River. The study project entitled "Effects of pine plantations on natural resources sustainability in Uruguay" is being conducted by researchers from North Carolina State University (Chescheir et al., 2003; Chescheir et al., 2004; von Stackelberg et al., 2007).

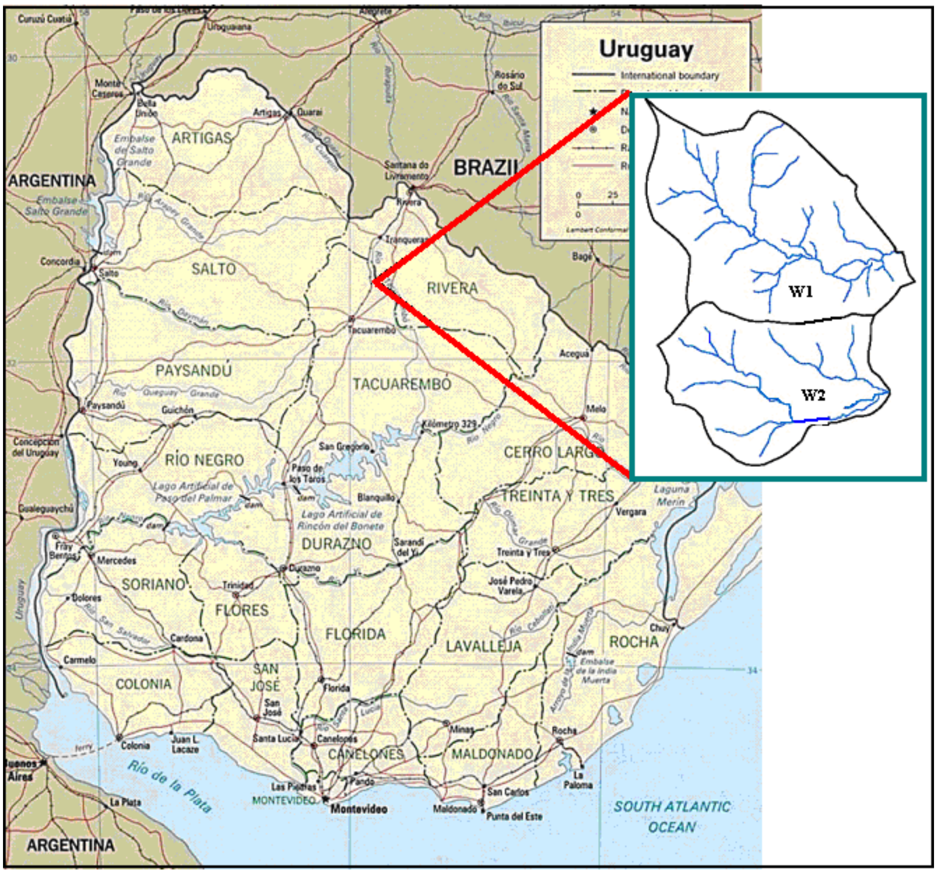


Figure 2. Location of the research site.

Our work, framed inside this project, started in 2002 and is expected to last until 2009. The research team is composed of scientist from the Soil Water Department of the Agronomy College, University of the Republic of Uruguay. The objective of the study is to evaluate the effect of this land use change on the quality of the runoff water. The water quality indicators evaluated are total phosphorus, total nitrogen, chlorides, ammonium, nitrate, total solids, turbidity, and alkalinity. This study is important not only because at a national level there is no published information about the possible impacts of afforestation on water quality, but also because there is little published information with respect to the quality of the water resources of the country.

In the thesis, only the results of total phosphorus and total nitrogen of the initial evaluation period (2002-2005) will be presented, because another implicit objective is to adjust the methodology of analysis that will be used in the future, when more information will be available. This thesis was written in the format of a scientific paper, following the specifications of the Journal of Environmental Quality, and presented in two languages, one in Spanish, directed to the local public, and another in English, intended for the international scientific community.

# LITERATURE REVIEW

Eutrophication is the nutrient-driven growth increase of algae and aquatic plants which produce anoxic conditions in lakes and water courses causing death to fish and other species. The algae growth can also reduce light penetration and release substances toxic to humans which further reduce water quality. In natural conditions, this process occurs at a slow rate, but human activities can greatly accelerate the rate of nutrient release into water bodies (Khan y Ansari, 2005).

Even at low concentrations, phosphorus (P) and nitrogen (N) are two of the most influential nutrients of the eutrophication process. The concentration increase of P in surface waters could cause eutrophication even at levels as low as 10-35 $\mu g\ L^{-1}$, while N could be a limiting factor for algae growth in estuaries and oceans (Zachary y Petrovic, 2004). It is generally considered that P limits the phytoplankton production, while N restricts their accumulation (Rabalais, 2002) and determines the composition of species of present algae (Young et al., 1996).

In soils, most of the native P and that added by fertilization are strongly associated to its solid phase, both in organic and inorganic forms. The amount present in solution at a given time, on the other hand, is much smaller, the phosphate ion ($H_2PO_4^-$) being the prevalent form. As water moves through the soil, it transports both the P associated with soil particles and that in solution. Generally, more of the P is transported downstream toward surface water bodies and less to the groundwater, because solid particles are dragged by surface runoff (erosion) and the ion $H_2PO_4^-$ its highly adsorbed to the solid phase (Tisdale et al., 1994; Sharpley et al., 2002; Zaimes y Schultz, 2002).

Nitrogen enters the soil mainly by biological fixation and/or fertilization and most of it is transformed into the soil where remains organically bound. Annually, a small part of the organic matter is mineralized to ammonium ($NH_4$), which is quickly oxidized to nitrate ($NO_3$) for nitrifier bacteria. For this reason the mineral soluble forms that prevail in the soil solution are $NO_3$ and $NH_4$, while $NO_2$ (nitrite) is generally present only in trace amounts. The high mobility of $NO_3$ determines that the largest movement of soluble N occurs through the soil profile (lixiviation) and ends up in groundwater, while exports of these N forms by water runoff are relatively minor, except under conditions of high subsurface flow (Randall and Mulla, 2001). The organic N associated to soil particles, however, also moves with the runoff water and can contaminate surface waters, although this N form is less available to microorganisms than the inorganic soluble forms. Therefore, the amounts of both N and P lost by surface runoff are favored by an increase in the transport of suspended organic matter (Delgado, 2002).

In many countries, the concentration levels of N and P have been used as indexes to classify the trophic status or to identify the risk of eutrophication of aquatic ecosystems. Some authors have established stricter simpler water quality limits based not on statistics but only on biological data, and consider that TP concentrations higher than 5 $\mu g\ L^{-1}$ can cause eutrophication (Hinesly and Jones, 1990; Zachary y Petrovic, 2004). One classification system for surface water quality based on statistical data was proposed by Dodds et al., (1998). This author compiled a database with averages of total nitrogen (TN) and total phosphorus (TP) of more than 200 rivers and streams of North America and New Zealand (Table 1), and used the accumulated distribution of the log-concentration of P and N to set up limits among trophic classes. In this classification, the highest value of the lowest third of that distribution was established as the limit between the oligotrophic and mesotrophic categories, while the inferior value of the highest third was considered as the limit between the mesotrophic and eutrophic categories.

Table 1. Concentration ranges of nitrogen and phosphorus in rivers and streams for different trophic status, according to Dodds et al., (1998).

| Status | Total Nitrogen | Total Phosporus |
|---|---|---|
| | $\mu g\ L^{-1}$ | |
| Oligotrophic | 0- 700 | 0- 25 |
| Mesotrophic | 700- 1500 | 25-75 |
| Eutrophic | > 1500 | >75 |

The United States Environmental Protection Agency (USEPA) uses another system which establishes different critical levels according to the hydric resource (course or lake) and ecoregion under consideration (USEPA, 2002). Ecoregions differ according to their geology, soil type, geomorphology and predominant land use. The definition of critical levels is based on a by-season analysis of the historical changes (10 years or more) of the concentration of the water quality indicators in an ecoregion. First, for the available series of years and for each river within an ecoregion, the seasonal median concentration of each indicator is estimated. Secondly, all medians from the same seasons are grouped and from that median distribution the 25$^{th}$ percentile (P25) is computed. This procedure is repeated for all seasons. Third, the reference condition is estimated as the mean of these four P25 values, one for each season. Finally, a multidisciplinary panel of experts defines the critical concentration, which can differ from the reference condition if the panel of experts considers it suitable. Ultimately, this critical level represents the achievable value under the current conditions of each ecoregion, since the pursued objective is to reduce in the water bodies the concentrations of the indicators from a non-desirable situation to more suitable condition. Adopting a very low critical level in an ecoregion with rich soils and intensive agriculture would not be realistic, since the possibility of adopting management measures to reduce those levels would be scarce. For this reason, in this system the critical levels vary among regions from 120 to 2180 $\mu g\ L^{-1}$ for TN and from 10 to 128 $\mu g\ L^{-1}$ for TP.

Another system similar in its philosophy to that of USEPA is the one defined by the Australian and New Zealand Environment Conservation Council (ANZECC, 2000), which also uses different reference values for each aquatic ecosystem and geographical region of the country (Southeast, Southwest, Center, Tropical and New Zealand). The reference value corresponds with the percentile 80 and/or 20 of the data distribution, depending on the degree of distortion (smoothly or moderately) of the aquatic system. As in the previous system, the critical level is defined by a panel of experts; for example for superficial waters the TP critical level varies from 10 to 100 $\mu g\ L^{-1}$ and that for TN from 150 to 1200 $\mu g\ L^{-1}$ (ANZECC, 2000).

## REFERENCES


ANZECC. 2000. Australian and New Zealand guidelines for fresh and marine water quality [Online]. Available at http://www.environment.gov.au/water/quality/nwqms/ (verified 14 Feb. 2008)

Chescheir, G.M., D.M. Amatya, and R.W. Skaggs. 2003. Effects of afforestation on the hydrologic behavior of a basin in the Tacuarembó River. Progress report for 2002-03 submitted to Weyerhaeuser Foundation. Raleigh, N.C.: North Carolina State University, Department of Biological and Agricultural Engineering.

Chescheir, G.M., N.O. von Stackelberg, D.M. Amatya, and R.W. Skaggs. 2004. Effects of afforestation on the hydrologic behavior of a basin in the Tacuarembó River. Progress report for 2003-04 submitted to Weyerhaeuser Foundation. Raleigh, N.C.: North Carolina State University, Department of Biological and Agricultural Engineering.

Delgado, J.A. 2002. Quantifying the loss mechanisms of nitrogen. J. Soil Water Conserv. 57:389–398.

Dodds, W.K., J.R. Jones, and E.B. Welch. 1998. Suggested classification of stream trophic state: Distributions of temperate stream types by chlorophyll, total nitrogen, and phosphorus. Water Res. 32:1455–1462.

Hinesly, T. D. and R. L. Jones. 1990. Phosphorus in waters from sewage sludge amended lysimeters. Environ. Pollut. 65:293–309.

Khan, F.A., and A. Ansari A. A. 2005. Eutrophication: An Ecological Vision. Botan. Rev. 71:449–482.

MGAP. 2000. (In Spanish) 2000 Agricultural Census. [Online]. Available at http://www.mgap.gub.uy/DIEA/CENSOVOL2/indice.htm (verified 5 Apr. 2008)

MGAP. 2005. (In Spanish) Statistic Bulletin. [Online]. Available at http://www.mgap.gub.uy/Forestal/DGF.htm (verified 11 Mar. 2008)

Rabalais, N. 2002. Nitrogen in aquatic ecosystems. Ambio 31:102–112

Randall, G.W. and D.J. Mulla. 2001. Nitrate nitrogen in surface waters as influenced by climatic conditions and agricultural practices. J. Environ. Qual. 30:337–344

Sharpley, A.N., T. Daniel, T. Sims, J. Lemunyon, R. Stevens, and R. Parry. 2003. Agricultural phosphorus and eutrophication, 2nd ed. U.S. Department of Agriculture, Agricultural Research Service, ARS–149, 44 pp.

Silveira, L., J. Alonso, and L. Martínez. 2006. (In Spanish) Effects of forestry plantations on the water resources of Uruguay. Agrociencia 10:75–93

Tisdale, S. L., W. L. Nelson, and J. D. Beaton. 1994. Soil fertility and fertilizers. Macmillan Publishing Company, New York

U.S.E.P.A. 2002. Water quality standards. [Online]. Available at http://epa.gov/waterscience/criteria/nutrient/ecoregions/index.html (verified 14 Feb. 2008).

von Stackelberg, N.O., G.M. Chescheir, and R.W. Skaggs. 2007. Simulation of the hydrologic effect of afforestation in the Tacuarembo River basin, Uruguay. Trans ASABE, 50:455–468.

Young, W.J., F.M. Marston, and J.R. Davis. 1996. Nutrient exports and land use in Australian catchments. J. Environ. Manag. 47:165–183

Zachary M. E. and A. M. Petrovic. 2004. Fertilizer source effect on ground and surface water quality in drainage from turfgrass. J. Environ. Qual. 33:645–655.

Zaimes, G.N. and R.C. Schultz. 2002. Phosphorus in agricultural watersheds: A literature review. Department of Forestry, Iowa State University, Ames, Iowa. 106p.

## II. INITIAL EFECTS OF AFFORESTATION ON RUNOFF WATER QUALITY IN A BASIN OF TACUAREMBO RIVER


### SUMMARY

Afforestation of areas previously destined to extensive livestock production could increase the concentration of total phosphorus (TP) and total nitrogen (TN) in surface water and lead to eutrification. In Uruguay there is a lack of information with respect to the effects of land use change on N and P concentrations and loads, and no references with respect to the expected levels of these variables under natural prairies with livestock production. The main objective of this work was to determine the effects of land use change from extensive livestock to forestry production on surface water concentration and load of TP y TN in two paired watersheds (W1 and W2) located in the Departament of Tacuarembó. A secondary goal was to collect information about the characteristic TP and TN values in these two systems. At the end of each watershed, we measured flow and collected surface water samples by using both an automatic (AM) and a manual (MM) sampling method. In the water samples we determined TP and TN concentration and estimated loads. There were no significant differences in TP and TN concentration during the evaluation period between watersheds (47.4 vs. 49.8 μg $L^{-1}$ of TP and 273.4 vs. 228.6 μg $L^{-1}$ of TN in W1 and W2 respectively). There were, however, significant differences between sampling methods (28.5 vs. 82.8 μg $L^{-1}$ of TP and 184.0 vs. 339.7 μg $L^{-1}$ of TN for the MM and the AM respectively), and loads followed a

similar trend. Therefore, during the evaluation period this land use change did not affect water quality, although these results were observed in young plantations and they could not be used to predict effects on water quality of older and more mature plantations.

**Abbreviations:** AYL, accumulated yearly loads; AM, automatic method; CDB, complete date base; MM, manual method; N, nitrogen; P, phosphorus; PCal, period of calibration; PDB, paired date base; PTreat, period of treatment; TN, total nitrogen; TP, total phosphorus; TNC, total nitrogen concentration; TPC, total phosphorus concentration; TPL, total phosphorus load; TNL, total nitrogen load;W1, cattle-raising watershed; W2, afforested watershed.

# INTRODUCTION

The forestation of land previously used for extensive livestock production on natural pastures (afforestation) can increase the nutrient concentration of surface water (Oyarzun and Huber, 2003), due to a higher fertilization, an increase in soil mineralization and to the largest soil-water contact favored by tillage during plantation. Tillage increases the fraction of soil that remains uncovered, which leads to greater nutrient losses by erosion and surface runoff. These losses are favored in part by the higher nutrient release from the mixed zone, a narrow area (1-3 cm.) of the soil surface which interacts with rain (Fiere y Gabet, 2002; Zaimes y Schultz, 2002). If the water flow remains constant, the increment in the concentration of nutrients will increase the load or amount of transported nutrients. Within areas of similar land use, the nutrient load also varies with intrinsic characteristic of basins, such as geomorphology, geohydrology, topography, and climate (USDA, 1999; Quinton et al., 2001; Gelbrecht et al., 2004; Udawatta et al., 2004). This higher nutrient load of surface waters can finally move downstream to the adjacent water bodies (lakes, lagoons), increase their nutrient concentration and lead in some occasions to an explosive growth of aquatic flora which obstruct light penetration and reoxygenation, a process called eutrophication (Khan and Ansari, 2005). Phosphorus (P) and nitrogen (N) are two of the more influential nutrients of the eutrophication process, and they are considered as primary variables with respect to their relationships with water quality (USEPA, 2000).

The type of land use and its change can greatly affects the concentration and load of nutrients in surface water. In comparison with natural systems, both agriculture and livestock production systems tend to increase nutrient losses via water runoff (Zaimes and Schultz, 2002). Tillage left the soil surface uncovered, which becomes more susceptible to water erosion and compaction for machinery. The tillage effect on nutrient losses also depends on factors like geology, soil type, slope, type of vegetation and climate. Nearing et al., (1993) presented results of a study carried out in the state of Georgia in a basin with different land uses types (60% forested, 23% with pasture, 1% with crops and 13% with urban use), showing that TN and TP load increased with the increase in area occupied by cultivated land. Instead, other studies carried out in areas with pastures, forests, or both land use types have not detected such effect (Owens et al., 1991, Thomas et al., 1992). These different results only emphasize the importance of obtaining local information.

The presence of animals in livestock production systems could greatly affect the amounts of N and P added to soil through animal waste (dung, urine) deposition (Zaimes y Schultz, 2002; Hubbard, et al., 2004; Agouridis et al., 2005). Moreover, trampling tends to compact the soil, increasing water runoff and erosion. O'Reagian et al., 2005, evaluated during 5 years in Australia the effect of several animal loads on TP and TN concentrations and loads of runoff water in 10 10-has watersheds. The animal loads used in this work were both fixed (0,13 and 0,24 UG) and variable, adjusted to forage availability. The concentrations of TN varied from 101 to 4000 μg N $L^{-1}$ and those of TP from 14 to 609 μg P $L^{-1}$, but these variations were not related with variations in animal load. The TN loads varied from 0,01 to 1,9 kg N $ha^{-1}$ and those of TP from 0,001 to 0,080 kg P $ha^{-1}$, but these variations were neither associated with the animal loads used.

The authors concluded that would be extremely difficult to detect in short term studies water quality changes caused by management, and speculated that at least a 20-yr study period would be necessary to detect such tendencies.

Numerous studies have compared nutrient losses from cattle-grazed pastures and forested watersheds. In New Zealand Quinn and Stroud, (2002) found that TN and TP concentrations were higher in the pasture than in the forested watershed (816 vs 428 µg N $L^{-1}$ and 56 vs 40 µg P $L^{-1}$, respectively). Similar tendencies were reported by Cooper and Thomsen, (1988) who studied concentration changes in the base flow of two basins with similar land use to that in the previous case; in the pasture and afforested watersheds the total concentrations were 306 vs 156 µg N $L^{-1}$ and 44 vs 23 µg P $L^{-1}$, respectively. The loads were of 0.86 vs 0.230 kg N $ha^{-1}$ and 0.122 vs 0.038 kg P $ha^{-1}$ respectively. In the south of Chile, on the other hand, similar studies have found inverse tendencies; the concentrations of TN were higher in watershed with native forests than in those with pastures (194 vs. 153 µg $L^{-1}$ respectively). The pastures in these studies included different types, some with limited agriculture and others with heaths. The load of NT was of 3.5 and 1.8 kg $ha^{-1}$ $year^{-1}$ in the forested and pasture watershed, respectively (Oyarzun and Huber, 2003). There were considerable differences, however, in the level of TN concentrations between the studies of New Zealand and Chile, being the values of concentrations much higher in New Zealand. This variation between regions could be associated to differences in climate and soil type, but also to variations in pasture management and productivity. According to Jones and Holmes (1985), overgrazed pastures tend to have higher sediment losses, but those that are lightly grazed tend to lost even less sediment than a forest.

In forested areas of Virginia (U.S.A.) TN losses were low (1.0 to 6.3 kg of N ha$^{-1}$ year$^{-1}$) and associated to the balance between nutrient release from residue decomposition and nutrient uptake by vegetation. The losses of TP were smaller to those found in agricultural or urban areas (0.02 to 0.67 kg P ha$^{-1}$ year$^{-1}$) and varied with the soil texture; the sandy soils generally tended to lost TP quickly than loamy soils. In the east of North Carolina, Chescheir et al., (2003b), collected information from 25 year-old studies on TN and TP losses. The studies were conducted on 40 basins with natural forests, forest plantations and wetlands, completing a database with more than 100 site-years. In half of the places, the mean concentration of TN and TP was inferior to 1500 and 70 μg L$^{-1}$, respectively. In all places, annual TN loads varied between 2.3 and 23.9 kg ha$^{-1}$, but when some few places with organic soils were excluded, loads were inferior to 7.5 kg ha$^{-1}$. In these sites the annual TP load varied between 0.05 to 0.36 kg ha$^{-1}$. In this same area, the water quality of three experimental watersheds forested with *Pinus taeda* was also evaluated during 17 years (1988-2004). The mean loads of TN and TP were 6.94 and 0.17 kg ha$^{-1}$ respectively (Amatya et al., 2006); inferior therefore to those loads previously reported.

Young et al., (1996) compared the information available in Australia and North America with respect to the loads of nutrients associated to different land uses and they found that the ranges reported for North America were larger than those reported for Australia (Table 2). The authors concluded that local environmental conditions as climate (intensity of rain, frequency and magnitude of the storm events), topography (slope, basin size and drainage density) and soil type determines in great deal the magnitude of load and concentration of nutrients, and they suggest that these information could only be based on local investigation.

Table 2. Mean annual loads of total phosphorus (TP) and total nitrogen for different land uses in different world zones (Young et al., 1996).

| World Zone | Land use | TP | TN |
|---|---|---|---|
| | | ———kg $ha^{-1}$ $year^{-1}$——— | |
| Australia | Natural pasture | 0.002 – 0.4 | 0.6 – 5.1 |
| | Forest | 0.001 – 0.2 | 0.9 – 1.5 |
| North America | Natural pasture | 0.3 – 2.8 | 2.0 – 11.0 |
| | Forest | 0.1 – 0.4 | 2.0 – 3.5 |

In Uruguay there are few published reports about the land use effects on concentration and load of nutrients in surface water courses. There is also almost no information about the original nutrient levels of the Uruguayan hydric resources. There is some recent information on the PT and NT concentration levels of the Uruguay River, but only from a 35-km tract located near to the paper mill plant of Botnia. During the period of this evaluation, the mean concentration of PT varied between 30 and 145 μg P $L^{-1}$, while that of NT varied between 900 to 1250 μg N $L^{-1}$ (DINAMA, 2007). Values between 57 and 110 μg P $L^{-1}$ has also been cited as the representative TP range for the Salto Grande damn for the period between 1980 to 2002 (Chalar et al., 2002). A TP load of 0.54 kg $ha^{-1}$ has also been cited as representative for this same place (Chalar, 2006). The objective of this work were i) to determine the effects of the land use change from cattle-grazed pastures to afforestation on the concentration and load of PT and NT of the runoff water in two paired watersheds located in the Tacuarembó Department, and ii) to collect local information about the representative values of these indexes in these two land-use systems.

# MATERIALS AND METHODS

This study was done in "La Corona" Ranch, department of Tacuarembó, Uruguay, from September 2002 to August 2005. This ranch, which belongs to Colonvade S.A., began to be planted with pines and eucalypts destined to the lumber industry in 2003. Two paired watersheds with similar topography, slope, and soil types were selected and marked for this study in an area inside this ranch. The drainage area was of 69 has in watershed 1 (W1) and of 107.7 has in watershed 2 (W2). The perimeter was of 3.5 and 4.6 km, the length 1.1 and 1.7 km, the drainage density 2.0 and 1.9 km $km^{-2}$ and the elevation ranged from 130 to 204 and from 136 to 192 m for W1 and W2 respectively.

The soil mapping and classification was done by Molfino (2000) and the soils physical and chemical characterization by García et al. (2004). The description of the native flora was carried out by Marchesi (2003). During the whole study period (12/09/02 to 31/08/05), W1 was used as the control watershed and left unchanged with the same original vegetation (a native pasture, mainly a mixture of C3 and C4 grasses). The treatment watershed (W2) remained with the same original vegetation only until 30/07/03 (calibration period), and then (treatment period) was planted with loblolly pine seedlings (*Pinus taeda*). The riparian zone, which presented a high diversity of native flora, was grazed only in W1. Watershed 1 was always grazed with cows, with an animal load of 0.8 cattle unit $ha^{-1}$ from April to September and 1.2 cattle units $ha^{-1}$ from October to March. A cattle unit is equivalent to the consumption of a cow weighing 380 kg that gestates and weans a calf per year.

The plantation in W2 was planted and managed according to the usual practices of the company; plants grew during the first three month in a greenhouse and were then transplanted to the field into furrows (0.1 m depth by 0.7 m width) aligned perpendicularly to the slope and separated by 2.5 m. The stock density was of 1000 trees $ha^{-1}$ and the plantation was not fertilized. Weeds were controlled by spraying Glyphosate in bands along planting rows but the area between planted rows remained untreated and with the original vegetation. All pruning and thinning practices were also carried out according to the standards of the company.

A 3-meter tall Campbell Scientific weather station equipped with automatic sensors and a CR10X data-logger was installed on the ridge between the two catchments. The sensors continuously measured air temperature, relative humidity, wind speed, wind direction, solar radiation, and net radiation on a 30-second interval and stored data on a 15-minute basis for analysis. The weather station was also equipped with an automatic rain gauge. The flow rates at the outlet of the two experimental watersheds were measured using 1.37 m high HL flumes (Chescheir et al., 2003a) and Onset (HOBO U12) modules. These modules were set to collect and record stage elevations every two minutes (Chescheir et al, 2004).

Two water sampling methods were used in each basin; one was the automatic method (AM) carried out with an ISCO 6712 automatic water sampler and the other was the manual method (MM) performed by collecting a grab water sample once a week. The ISCO 6712 equipment was programmed to take a water sample every 1200 $m^3$ of flow in W1 and every 2000 $m^3$ in W2; this volume difference compensated for the area differences between watersheds. Automatic samples collected from each watershed in the same week were stored inside the same large bottle, forming in this way a flow proportional composite sample. For the last part of the experiment (after 26/3/04), automatic samples were collected every two weeks, so they represent the flow of two weeks. The MM sampling was generally performed the same day of the weekly AM composite sample extraction. When no samples were collected by the automatic samplers because of low flow, MM samples were still collected. Due to the differences between sampling methods, there was a larger proportion of single samples colleted during storm events in the composite AM samples than in the MM samples, hence this latter method probably better represented the flow base conditions of the course. Water samples of the Tacuarembó River were also seasonally collected by manual sampling near to the study site.

Immediately before use, 1 mL of $H_2SO_4$ 10 M was added to the collection bottles to prevent the occurrence of chemical and/or biological conversions. After extraction water samples were kept at 4ºC until analyses. Samples collected in fall of 2004 were eliminated because the extraction probes were contaminated with algae. Samples were analyzed by TP and TN; the analysis of TP was determined using an ammonium peroxidisulfate digestion followed by an ascorbic acid–molybdate procedure (Pot and Daniel, 2000). Previous to the TN analysis, a 100mL-aliquot of water was concentrated to 10 mL by evaporation at 90 ºC. To avoid $NH_3$-N losses, 2 mL of $H_2SO_4$ 5.5M was added to the aliquots before evaporation. The analysis of TN was done on the concentrated aliquots by Kjeldhal digestion (Bremmer and Mulvaney, 1982) and colorimetric determination of $NH_4$ with the blue indophenol technique (Rhine et al., 1998). The colorimetric determinations were performed with a UNICAM 5675 spectrometer.

Two databases were used for the analysis of results of both the MM and the AM. One database was the complete database (CDB) and the other was the paired-up database (PDB). The CDB differed from the PDB because in the second case the TP and TN data from W1 was weekly paired with that of W2. The PDB had then a smaller number of samples because in some weeks samples were not collected or available in one of the watersheds which caused the elimination of the TP or TN information for that week.

The load of N and P was estimated by multiplying the concentration of each sample by the water flow during the corresponding period. Due to the lack of TP or TN data at some weeks, the flow database used to estimate the TP load was different to that used to estimate the TN load; nevertheless, both flow databases had similar tendencies to that of the CDB.

Concentration data was not available in some weeks (18 wks in W1 and none in W2) because the weekly flow was near zero at these weeks. In other weeks (28 wks in W1 and 35 wks in W2), the flow was important but samples were not collected or the volume was not enough for running all analyses.

In all these cases, the load was estimated using the mean concentration of the whole evaluation period and the weekly flow of each particular week; this decision was based on the lack of a significant statistical relationship between flow and concentration.

Finally, the accumulated yearly load (AYL) was estimated as the weekly load summation of all periods.

All statistical analyses of concentration and load were conducted on log10-transformed (Log10) data, because the concentration and load of TN and TP were generally not normally distributed. In the CDB, concentration and load data for TP and TN was analyzed by ANOVAs using a complete randomized factorial design, where the treatments were the combination of the two sampling methods and the two land use types (or watersheds). In this analysis, sampling dates were considered as repetitions. The PDB was analyzed instead by Analysis of Regression (ANREG) and Covariance (ANCOVA); this is the methodology recommended by USEPA to evaluate paired watersheds (Clausen and Spooner, 1993; Grabow et al., 1998). The ANREG evaluated in each period (calibration or treatment) the statistical significance of the lineal regressions between the concentrations or loads of both watersheds. The ANCOVA evaluated the existence of differences between periods in the values of the parameters obtained previously by linear regressions. When there was a significant difference between the slopes of the two straight lines of different periods (PCal and PTreat), it was then considered that afforestation had modified the evaluated variable in W2 with respect to W1. The probability value of 0.10 was considered as the limit of statistical significance. All these statistical analyses were carried out using the statistical package SAS version 6.0 (SAS Institute, 1990).

# RESULTS AND DISCUSSION

## Rain and Runoff Water

Rain varied during the study period and was much more intense in PCal than in PTreat (Table 3). It is important to mention that the mean weekly rain in PCal was above the historical 28-year mean (29.2 mm) of the nearest meteorological station (INIA Tacuarembó), although the PTreat mean was below the historical mean (Table 3).

During the evaluation period the flow followed a similar trend to that of rains, being higher in PCal than in PTreat. When comparing the two watersheds, in both periods the flow was higher in W2 than in W1. This difference was obviously not related to the land use change, because it also occurred before afforestation (Table 3).

Table 3. Rain and water flux in two watersheds (W1 and W2) during the study and results of linear regression and covariance analyses (ANCOVA). The linear regressions were computed between the water flux of both watersheds in each period, and ANCOVAs tested differences in regression parameters between periods.

| | Rain | | | Water flux | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | Watershed | | Regression | | ANCOVA | | |
| Period† | Yearly Total | Weekly Mean | Standard Deviation | W1 | W2 | | W1 x W2 | Watershed | Period | Watershed x Period |
| | mm | | | $m^3\ ha^{-1}\ year^{-1}$ | | b | *P* value | | | |
| PCal | 2647,4 | 50,9 | 61,3 | 11987,3 | 14470,3 | 1.01 | 0,0001 (405)‡ | 0,0001 | 0,0002 | 0,0001 |
| PTreat | 1254.2 | 23,8 | 32,1 | 2577.0§ | 3680.6 | 0.93 | 0,0001 (795) | | | |

† PCal: Period of calibration (one year); PTreat: Period of treatment (two years).

‡ Númber of samples

§ Anual mean

The linear regression between the drainage in both watersheds was significant in PCal and in PTreat; therefore the changes of flow in both basins were related. The numerical relationships between watersheds, however, differed among periods, since the slope of the linear regression of PCal was significantly higher than that of PTreat (effect "period x watershed" significant in ANCOVA). The "period" and "watershed" effects were also significant in that analysis, implying respectively that independently of the watershed there was a flow difference between periods, and that independently of the period there was a relationship between the drainage of both watersheds. The smaller slope in PTreat could be associated to an early decrease of the drainage in W2 due to the higher evapotranspiration of pines in relation to that of pastures (Zhang et al., 1999; Farley et al., 2005). This analysis, however, is complicated by the large differences in flow between the PCal period and the PTreat period. The analysis is also complicated by the differences in baseflow between the two watersheds which resulted in a nonlinear flow relationship between the watersheds during low flow periods (Chescheir et al., 2008). It is interesting to note that flow from W2 was only 1.2 times greater than from W1 in PCal, but was 1.4 times greater than from W1 in PTreat.

## Nutrient concentration

### Total Phosphorus

TP concentration (TPC) varied in both watersheds during the evaluation period, but followed no clear trend (Figure 3). ANOVA conducted in CDB did not show any significant difference in TPC between watersheds during the evaluation period (Table 4). Nevertheless, differences were observed between sampling methods. In W1 and W2, the adjusted means of MM were lower than those of AM (non-significant interaction). Differences in methods were probably influenced by the fact that an important fraction of the total AM samples were collected during storm conditions, when rain was intense and promoted erosive events. Therefore, TPC values in MM more properly represented baseflow whereas those in AM represent a combination of baseflow and storm flow.

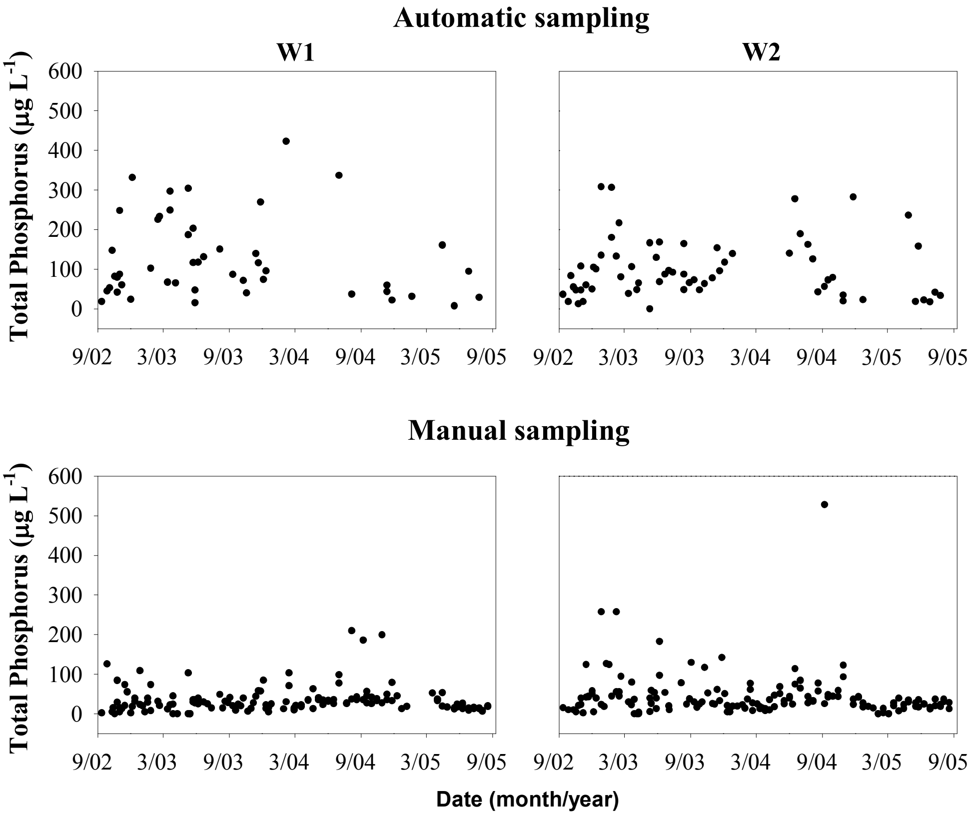


Figure 3. Total P concentration values during the evaluation period for the two sampling methods.

Table 4. Effect of sampling method and watershed on TP concentration evaluated pre (calibration) and post (treatment) afforestation. The concentration data was logarithmically transformed before ANOVAs and the means were back transformed.

| | Calibration | | | Treatment | | |
|---|---|---|---|---|---|---|
| Watershed† | MM‡ | AM | Mean | MM | AM | Mean |
| | | | $\mu g\ L^{-1}$§ | | | |
| W1 | 22.00 | 105.07 | 48.08 | 26.89 | 83.67 | 47.43 |
| W2 | 33.19 | 89.75 | 54.58 | 30.27 | 81.89 | 49.79 |
| Mean | 27.02 | 97.11 | | 28.5 | 82.8 | |

ANOVA Statistics

| Source of variation | *P* value | |
|---|---|---|
| Method | 0.0001 | 0.0001 |
| Watershed | NS¶ | NS |
| Method * Watershed | NS | NS |

† W1: Cattle-raising watershed; W2: Afforested watershed

‡ MM: Manual Method; AM: Automatic Method

§ Adjusted Means

¶ Not significantly different at $P > 0.10$

Mean TPC values of PCal and PTreat in both sampling methods were greater than the minimum critical level of all ecoregions (10 μg $L^{-1}$) cited by USEPA (2002) for rivers and streams, but were not greater than the maximum critical level (128 μg $L^{-1}$). Such critical levels would not be applicable to these sites in Uruguay, not only because of clear ecological differences, but also because W1 and W2 are narrow canyons with intermittent flow. The data USEPA used to establish the critical levels were obtained in rivers and streams with permanent water flow and deeper water columns that lead to greater sedimentation (USEPA, 2007). Other authors such as Birr and Mulla, (2001) preferred to use the mean concentration of a nearby river as a reference. This comparison avoids the interpretation problems associated with the use of critical levels of different ecological zones as reference, but keeps those associated to the difference in water column depth. By using this methodology, we compared results of both watersheds with the mean TPC-MM of the Tacuarembó River obtained by us (55 μg $L^{-1}$), and found that MM values from both watersheds were lower, whereas TPC-AM values were higher. Although TPC-MM values from both watersheds and that of the Tacuarembó River would not necessarily be comparables, the fact that the higher MM-TPC value was observed in the Tacuarembó River with the deeper water column clearly indicates that in both watersheds the TPC of the baseline water flow was not greater than that of the river. Anyway, AM values would probably be more representative of the whole fluvial basin dynamic.

In both periods, mean TPC values for both sampling methods of W1 were similar to those reported by Cooper and Thomsen (1988) for baseline waterflow in pasture watersheds (44 μg $L^{-1}$); however, they were higher than those found by those same authors for pine watersheds (23 μg $L^{-1}$). Furthermore, TPC values of W1 were within the concentration range (14-609 μg $L^{-1}$) mentioned by O'Reagian et al., (2005) for cattle-raising watersheds. Values similar to those observed at W1 and W2 were also reported by Quinn and Stroud (2002) for cattle-raising watersheds on native vegetation (41 versus 40 μg $L^{-1}$).

In PDB, the TPC regression line of both watersheds was significant in PCal and PTreat, and for both sampling methods (Table 5). The result implies that the same events that caused changes in TPC in one watershed also caused changes in the other. ANCOVA showed that neither the intercepts (period effect) nor the slopes of adjusted lines estimated for each period differed significantly, where the result was not affected by the sampling method. These results corroborate those obtained by ANOVA as previously reported in Table 4, where CDB was used.

Table 5. Regression and covariance analyses for $Log_{10}$ of total phosphorus concentration.

| Method† | Regression between W1 and W2 within each period and method‡ | | Covariance | | |
|---|---|---|---|---|---|
| | Calibration | Treatment | Watershed | Period | Watershed x Period |
| | *P* value | | | | |
| MM | 0.0050 (26)§ | 0.0001 (52) | 0.0001 (78) | 0.6042 | 0.5384 |
| AM | 0.0402 (16) | 0.0280 (13) | 0.0029 (29) | 0.5233 | 0.5833 |
| MM y AM | 0.0001 (42) | 0.0001 (65) | 0.0001 (107) | 0.9537 | 0.9373 |

† MM: Manual Method; AM: Automatic Method

‡ W1: Cattle-raising watershed; W2: Afforested watershed

§ Number of samples

Because the statistical analyses conducted in PDB showed no effect of afforestation on TPC, a new regression line was adjusted using all data obtained by both methods during the evaluation period, except for the five points considered to be outside the typical range. It must be said that the addition of these points would not have changed our conclusions. The results show the existence of a clear relationship between the TPC in both watersheds and the TPC differences between both sampling methods (Figure 4). Therefore, our results based on both databases also indicate that differences in TPC were not caused by changes in land use but by sampling methods.

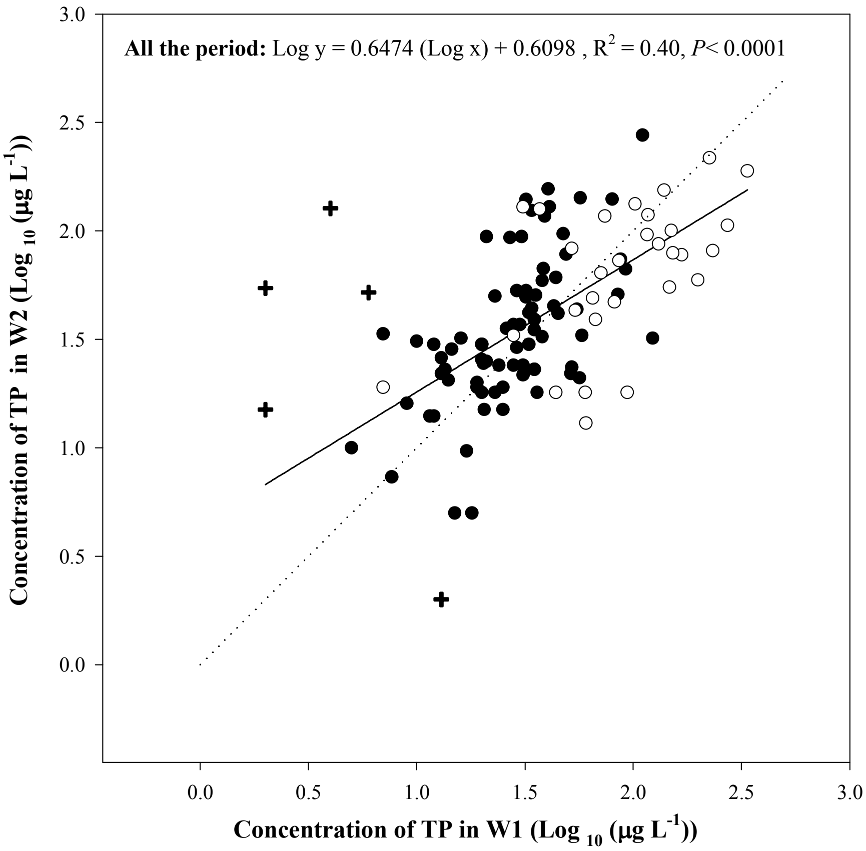


Figure 4. Relationship between TP concentrations in both watersheds during the evaluation period, obtained by Manual (MM) and Automatic (AM) sampling methods. Filled circles represent TPC values obtained by MM and empty ones by AM; crosses are the dots outside the scale. The dotted and the continuous lines represent the relation 1:1 and the regression line between the TPC of both watersheds, respectively, independent of the sampling method used.

**Total Nitrogen**

Total N concentration (TNC) tended to be higher during the first six evaluation months (pre-planting period) in both watersheds (Figure 5). Values decreased sharply soon after and then were stable until the end of the experiment. Therefore, concentration peaks were not related to changes in land use. The trend was observed in both sampling methods, and the higher TNC values observed during the first six months were not clearly associated to storm events although their magnitude was always higher in AM. As observed in TPC, TNC values obtained with AM were higher than those obtained with MM for the rest of the evaluation period. The occurrence of these peaks coincided with the more intense rain registered in PCal (Figure 5). Peaks might have been a consequence of more erosion caused by intense rain during the early stage. However, no initially high concentrations of TPC were observed, which would be expected if greater erosion occurred. Therefore, the reason for the higher initial TN concentration remains unknown.

The ANOVAs conducted in CDB showed no significant differences in TNC in PCal and PTreat between both watersheds (Table 6). Nonetheless, similar to observations of TPC, there were significant differences between the two sampling methods. As mentioned before, the MM adjusted means in W1 and W2 were lower than those of AM (non-significant interaction).

Similar to TPC and independent of the sampling method used, both watersheds exceeded the minimum critical level of TNC (120 $\mu g\ L^{-1}$) cited by USEPA (2002); nonetheless, the maximum level was not exceeded (2180 $\mu g\ L^{-1}$). The mean TNC value for both MM and AM was greater than the TNC mean in the Tacuarembó River (133 $\mu g\ L^{-1}$).

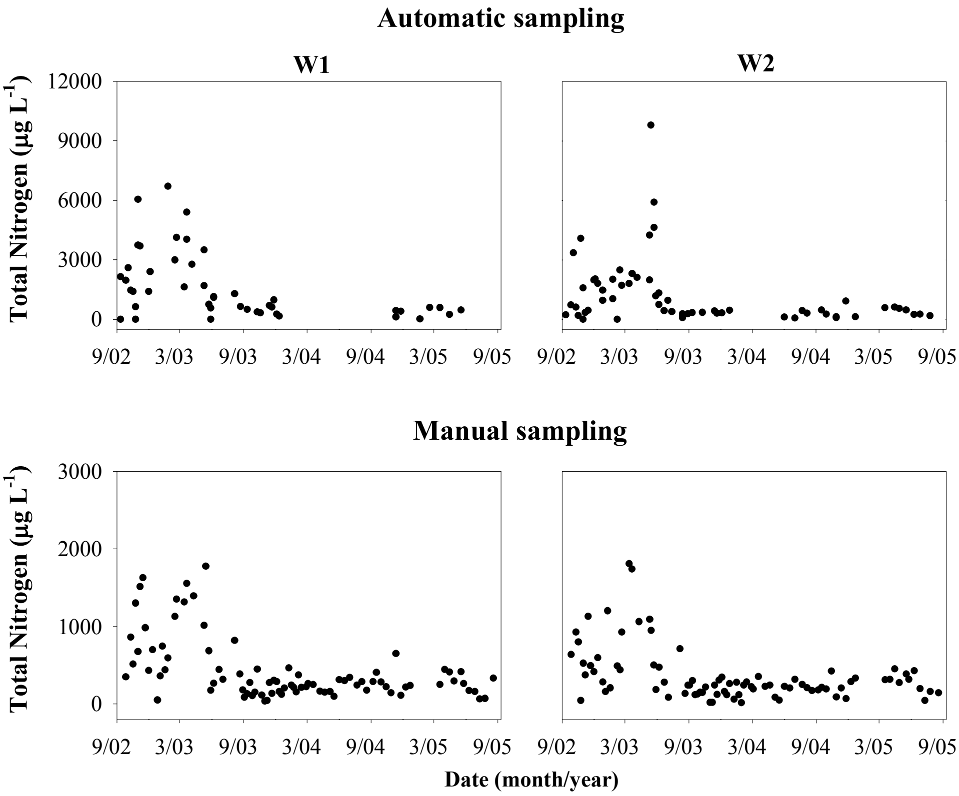


Figure 5. Total N concentration during the evaluation period for two sampling methods.

Table 6. Effects of sampling method and watershed on TN concentration evaluated pre (calibration) and post (treatment) afforestation. The concentration data was logarithmically transformed before ANOVAs and the means were back transformed.

| Watershed† | Calibration | | | Treatment | | |
|---|---|---|---|---|---|---|
| | MM‡ | AM | Mean | MM | AM | Mean |
| | $\mu g\ L^{-1}$§ | | | | | |
| W1 | 552.3 | 1776.7 | 990.6 | 201.73 | 370.63 | 273.44 |
| W2 | 518.2 | 1301.9 | 821.4 | 167.89 | 311.35 | 228.63 |
| Mean | 535.0 | 1520.9 | | 184.04 | 339.70 | |

ANOVA Statistics

| Source of variation | *P* value | |
|---|---|---|
| Method | 0.0001 | 0.0001 |
| Watershed | NS¶ | NS |
| Watershed method * | NS | NS |

† W1: Cattle-raising watershed; W2: Afforested watershed

‡ MM: Manual Method; AM: Automatic Method

§ Adjusted means

¶ Not significantly different at $P > 0.10$

The TNC range in W1 and W2 were similar to the range reported by Quinn and Stroud (2002) in cattle-raising and afforested watersheds. Soon after afforestation, TNC in W1 was similar to the range mentioned by Cooper and Thomsen (1988) for baseline water flow in pasture watersheds; on the other hand, TNC in W2 was lower than the range cited by the same authors for the baseline flow in forested watersheds. In all, TNC values in both watersheds were within the order of magnitude observed by other authors.

In PDB, the regression line for the TNC of the two watersheds was significant in PCal and Ptreat (Table 7), both in MM and in the combined methods (MM and AM). On the other hand, this relation was not significant in AM. The results of ANCOVA showed that the linear slope of PTreat was not significantly different from that observed in PCal, and therefore land use did not affect TNC. A decrease in PTreat-TNC values with respect to those of PCal was observed for both methods, although the difference of intercepts (period effect) was close to being significant ($P = 0.1131$) only when both methods were evaluated together. This decrease was related to the highest TNC values observed during the first six months of evaluation.

Similar to TPC, a regression line between the two watersheds was adjusted for synthesis, with data obtained by both sampling methods during the evaluation period (Figure 6). The acceptable relationship between the TNC of both watersheds was again observed, as was higher TNC for AM when compared to MM.

Table 7. Regression and covariance analyses for $Log_{10}$ of the total Nitrogen concentration.

| Method† | Regression between W1 and W2 within each period and method‡ | | Covariance | | |
|---|---|---|---|---|---|
| | Calibration | Treatment | Watershed | Period | Watershed x Period |
| | *P* value | | | | |
| MM | 0.0081 (30) | 0.0001 (55) | 0.0001 (85) | 0.1964 | 0.2688 |
| AM | 0.8420 (15) | 0.6528 (11) | 0.8759 (26) | 0.4582 | 0.7444 |
| MM y AM | 0.0002 (45) | 0.0001 (66) | 0.0001 (111) | 0.1131 | 0.2094 |

† MM: Manual Method; AM: Automatic Method

‡ W1: Cattle-raising watershed; W2: Afforested watershed

§ Number of samples

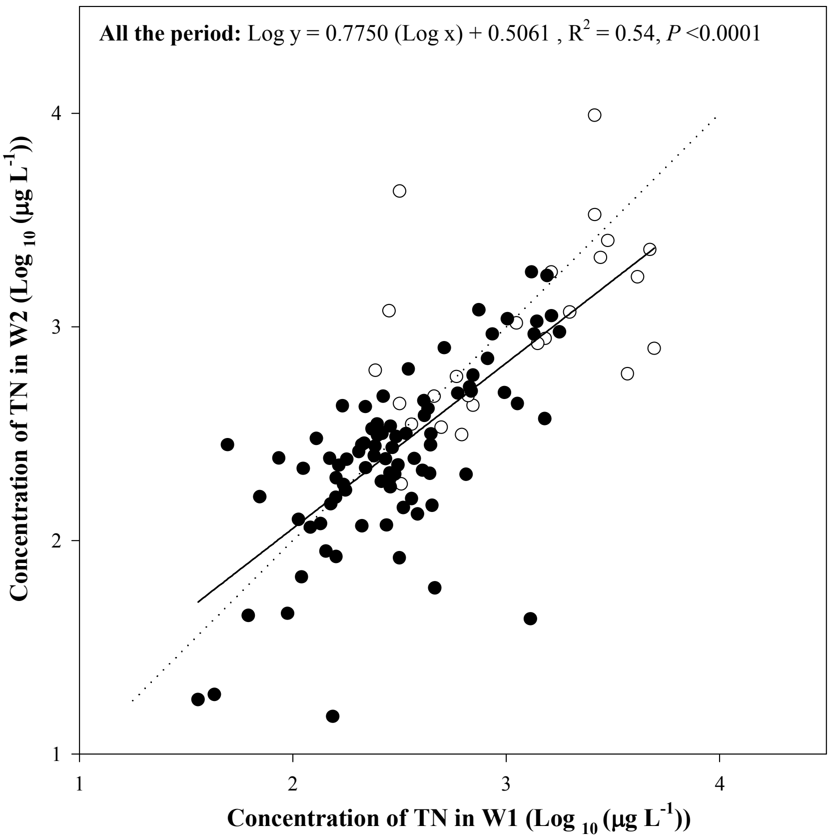


Figure 6. Relationship between TN concentrations in both watersheds during the evaluation period, obtained by manual (MM) and automatic (AM) sampling methods. Filled circles represent TNC values obtained by MM and empty ones by AM. The dotted and the continuous lines represent the relation 1:1 and the regression line between the TNC of both watersheds, respectively, independent of the sampling method used.

## Nutrient Load

### Statistical analyses

According to the ANOVAs conducted in CDB, there were no significant effects of land use changes on loads of TP (TPL) or TN (TNL). However, differences between sampling methods in both evaluation periods were found, where the adjusted MM means were lower than those of AM (Table 8). Furthermore, this difference between methods was found in both watersheds (non-significant interaction), an expected result considering the differences between the existing methods in TNC and TPC.

Table 8. Effect of the sampling method on TP and TN load at the cattle-raising and the afforested watersheds in the periods of calibration and treatment The load data was logarithmically transformed before ANOVAs and the means were back transformed.

| | | Calibration | | | Treatment | | | Statistics | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Index | Watershed† | MM‡ | AM | Mean§ | MM | AM | Mean | Source of variation | | |
| | | g $ha^{-1}$ $week^{-1}$ | | | | | | | *P* value | |
| TPL | W1 | 4.0 | 52.6 | 28.0 | 1.5 | 7.8 | 4.7 | Method | 0.0424 | 0.0047 |
| | W2 | 18.1 | 69.4 | 43.5 | 3.0 | 16.0 | 9.5 | Watershed | NS¶ | NS |
| | Mean | 11.0 | 61.0 | 32.2 | 2.3 | 11.9 | 5.3 | Method x Watershed | NS | NS |
| TNL | W1 | 127.1 | 576.1 | 332.9 | 7.9 | 53.4 | 30.4 | Method | 0.0001 | 0.0001 |
| | W2 | 164.2 | 405.3 | 279.1 | 13.2 | 42.9 | 27.9 | Watershed | NS | NS |
| | Mean | 145.4 | 488.5 | 270.9 | 10.5 | 48.1 | 24.1 | Method x Watershed | NS | NS |

† W1: Cattle-raising watershed; W2: Afforested watershed

‡ MM: Manual Method; AM: Automatic Method

§ Adjusted means

¶ Difference not significant at $P > 0.10$

Analyses conducted in the TPL and TNL data of the PDB showed significant linear regressions between the two watersheds in the two periods, these results were alike when using MM, AM, or the combination of MM and AM (Table 9). ANCOVA showed that the slopes of the regression lines within each group were significantly different (interaction watershed x period significant), as were the intercepts (period effect significant), except for TPL-AM. Irrespectively of statistics, however, the trends were similar in all cases, because with respect to PCal, the slopes in PTreat were higher and closer to one and the intercepts smaller and closer to zero (Table 9). This result implies that loads in both watersheds tended to be similar in PTreat.

Table 9. Regression (ANREG) and covariance (ANCOVA) analyses for TP (TPL) and TN (TNL) loads obtained with the manual (MM), automatic (AM), and both (MM and AM) sampling methods. The number of samples in ANREG for the calibration and treatment period were 35 and 51; 15 and 13; 50 and 64; 35 and 53; 14 and 11; and 49 and 64; for TPL-MM; TPL-MA; TPL-MM and MA; TNL-MM; TNL -MA; TNL -MM and MA; respectively. The numbers of samples in ANCOVA were the summation of the number of samples in both methods.

| | | ANREG† | | | | ANCOVA | | |
|---|---|---|---|---|---|---|---|---|
| | | Intercept | | Slope | | | | |
| Index | Method | Calibration | Treatment | Calibration | Treatment | Watershed | Period | Watershed x Period |
| | MM | Value of *P* | | | | | | |
| TPL | AM | 0.0001 (0.53) | 0.0001 (0.18) | 0.0011 (0.65) | 0.0001 (1.01) | 0.0001 | 0.0004 | 0.0645 |
| | MM y AM | 0.0089 (0.51) | 0.0152 (0.34) | 0.0063 (0.48) | 0.0002 (0.75) | 0.0001 | 0.4293 | 0.2339 |
| | MM | 0.0001 (0.55) | 0.0001 (0.19) | 0.0001 (0.53) | 0.0001 (0.94) | 0.0001 | 0.0001 | 0.0018 |
| TNL | AM | 0.0001 (0.97) | 0.0001 (0.37) | 0.0001 (0.57) | 0.0001 (0.84) | 0.0001 | 0.0001 | 0.0110 |
| | MM y AM | 0.0001 (1.74) | 0.0161 (0.60) | 0.0009 (0.31) | 0.0003 (0.70) | 0.0001 | 0.0004 | 0.0168 |
| | MM§ | 0.0001 (1.07) | 0.0001 (0.38) | 0.0001 (0.54) | 0.0001 (0.82) | 0.0001 | 0.0001 | 0.0005 |

† The number within parenthesis is the value of the intercept or the slope.

The slopes and intercepts of both indexes were significantly different, in general, between periods. Therefore, both methods (AM and MM) were compared, by estimating within each period the regression coefficients (intercepts and slopes) for each method, and then evaluating by ANCOVA if those coefficients were statistically different. The results showed that these coefficients were not different between methods in TPL-PCal and in TNL-PTreat, but they were different in the other two cases (Table 10). Therefore, in the two first cases the relationship between both watersheds was represented with the intercept and slope values of the combined methods of Table 9 (Figure 7 and Figure 8). In the other two cases, however, this relationship was represented separately for each method with their own corresponding intercept and slope value. These two figures summarize for both indexes the observed relationships between watersheds in the two periods, where it can be observed again that the slopes were higher and the intercepts smaller in PTreat with respect to PCal. The figures also showed that the main load difference between watersheds in PCal occurred at the lower load range, because in the higher range the values were similar.

In PTreat the slopes became closer to one, but although the intercepts decreased they still were significantly larger than zero (Table 10). Therefore, in this last period the tendency towards a larger load in W2 still existed, mainly at low loads. The reasons for this difference could be related to the higher flow observed in both periods in W2, which was not related to the land use change. It is possible that the higher resemblance between the two watersheds observed in PTreat was related with the decreasing slope between the flow in both watersheds found in this period (Table 3).

Table 10. Regression (ANREG) and covariance (ANCOVA) analyses for TP (TPL) and TN (TNL) loads in the periods of calibration (PCal) and treatment (PTreat). The number of samples in ANREG for the automatic and manual methods were 35 and 15; 35 and 14; 51 and 13; and 53 and 11; for TPL-PCal; TNL-PCal; TPL-PTreat and TNL-PTreat; respectively. The numbers of samples in ANCOVA were the summation of the number of samples in both methods.

| | | ANREG† | | | | ANCOVA | | |
|---|---|---|---|---|---|---|---|---|
| | | Intercept | | Slope | | | | |
| Period | Index | Manual | Automatic | Manual | Automatic | Watershed | Method | Watershed x Method |
| | | Value of $P$ | | | | | | |
| PCal | TPL | 0,0001(0,53) | 0,0089(0,51) | 0,0011(0,65) | 0,0063(0,48) | 0,0001 | 0,9394 | 0,5314 |
| | TNL | 0,0001(0,97) | 0,0001(1,74) | 0,0001(0,57) | 0,0009(0,31) | 0,0001 | 0,0119 | 0,0600 |
| PTreat | TPL | 0,0001(0,18) | 0,0152 (0,34) | 0,0001(1,01) | 0,0062(0,75) | 0,0001 | 0,1152 | 0,0653 |
| | TNL | 0,0001(0,37) | 0,0161(0,60) | 0,0001(0,84) | 0,0003(0,70) | 0,0005 | 0,3183 | 0,3307 |

† The number within parenthesis is the value of the intercept or the slope.

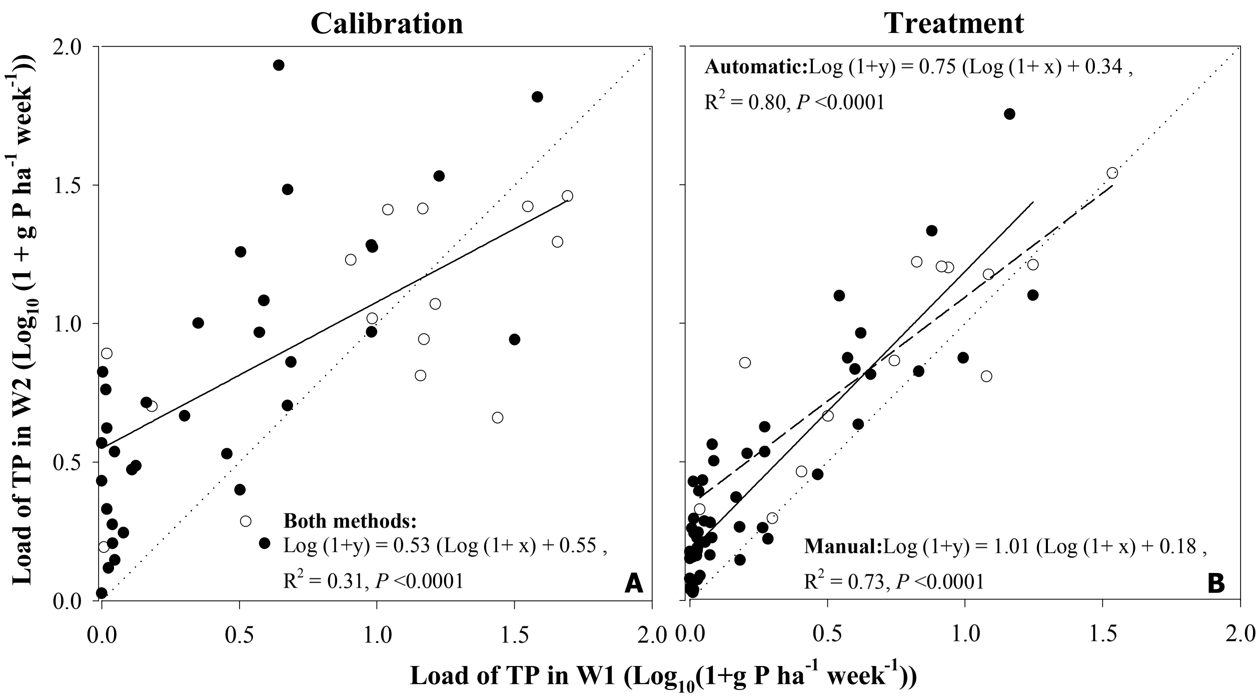


**Figure 7.** Relationships between TP loads of both watersheds for the calibration and treatment periods obtained with the automatic (AM) and manual (MM) sampling methods. The full circles represent the loads obtained with MM and the empty circles those obtained with AM. The dotted line represents the relationship 1:1, the continuous line represents the relationship between watersheds independently of the method in A) or that relationship for MM in B). The discontinuous line represents the relationship between watersheds for AM in B).

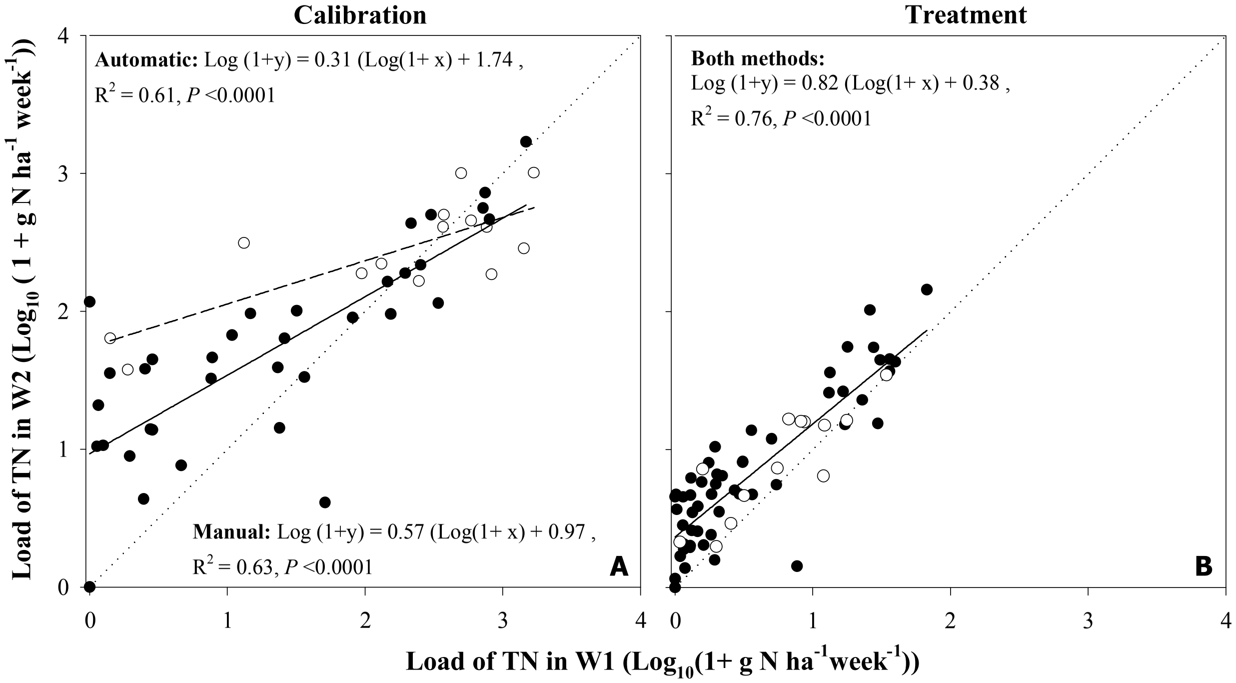


**Figure 8.** Relationships between TN loads of both watersheds for the calibration and treatment periods obtained with the automatic (AM) and manual (MM) sampling methods. The full circles represent the loads obtained with MM and the empty circles those obtained with AM. The dotted line represents the relationship 1:1, the continuous line represents the relationship between watersheds independently of the method in B) or that relationship for MM in A). The discontinuous line represents the relationship between watersheds for AM in A).

## Annual load estimates

Load estimates could vary according to the equipment used, such as automatic or manual devices to record water flow and automatic or manual extractions to determine concentration. For this reason, the incidence of three sampling strategies (SS) to estimate load using data from the CDB was evaluated, where loads were estimated as the product of flow times concentration, but using different flow and concentration values:

$$SS1 = dwf \times TC\_m \times 7 \quad [1]$$

$$SS2 = wwf \times TC\_m \quad [2]$$

$$SS3 = wwf \times TC\_a \text{ (or } wwf \times TC\_m \text{ if } TC\_a \text{ is unavailable)} \quad [3]$$

where *dwf* is the daily water flow, *TC_m* is the TP or TN concentration in water obtained by MM, *wwf* is the weekly water flow, and *TC_a* is the TP or TN concentration in water obtained by AM.

The SS1 would be applicable to situations where automatic sampling equipment is not available, although in this work the daily flow value was in fact obtained by automatic devices because manual ones were not used. This strategy is the most common in Uruguay, because automatic flow recording equipments are generally not available, although it is not recommended for load estimations (Reckhow et al., 1980). Starting from March of 2004 the sampling frequency changed from weekly to biweekly, therefore from that date the weekly load of the week of sampling was estimated as in [1], but in the previous week the load was estimated by multiplying the concentration of the sampling week by the weekly flow of the previous week. The SS2 would be applicable when only the flow was recorded automatically, and the SS3 when both the flow data registration and the water sampling were done automatically.

As expected, estimates of accumulated yearly loads (AYL) of TP and TN varied between SS (Table 11). The greatest AYL corresponded to SS3 because AM registered the highest concentrations. Sampling strategies 1 and 2 on the other hand, usually estimated lower AYL values because MM registered lower concentrations. Therefore, SS1 and SS2 might underestimate loads to some extent.

The greatest differences among SS occurred in PCal, when the most intensive rain and frequent AM events occurred. These differences among SS were similar in TN and TN, and in both indices loads were larger in W2; this result was probably influenced by the higher flux of this watershed. Also, AYL decreased from PCal to PTreat in both watersheds and in both indices, which is compatible with the decreasing flux within periods.

O'Reagian et al., (2005) presented TPL and TNL results from cattle-raising watersheds in terms of TPL or TNL per event, instead of AYL. In our work, we estimated the mean TPL and TNL per event as the ratio between the AYL mean for all the evaluation period and the number of events in this period, using in this calculation the AYL values obtained with SS3 (Table 11). Our mean W1 load values per event were lower than those reported for cattle-raising watersheds for both TN (123 versus 296 g ha$^{-1}$ per event) and TP (12 versus 14 g ha$^{-1}$ per event). In PCal the opposite trend was observed, because in this period the mean loads observed in our work were 330 and 30 g ha$^{-1}$ for TNL and TPL, respectively. Therefore, the mean SS3 of PCal and PTreat were respectively higher and lower compared to the values reported for these authors.

Table 11. Accumulated yearly TP and TN loads according to different flow and concentration sampling strategies. The estimates were obtained with the complete data base.

| | | W1† | | | W2 | | |
|---|---|---|---|---|---|---|---|
| | Period‡ | SS1§ | SS2¶ | SS3# | SS1 | SS2 | SS3 |
| | | kg ha$^{-1}$ year$^{-1}$ | | | | | |
| TPL | PCal | 0.34 | 0.56 | 1.60 | 0.77 | 0.86 | 2.37 |
| | PTreat1 | 0.06 | 0.06 | 0.12 | 0.10 | 0.12 | 0.46 |
| | PTreat2 | 0.07 | 0.09 | 0.15 | 0.14 | 0.20 | 0.18 |
| | Mean | 0.16 | 0.24 | 0.62 | 0.33 | 0.39 | 1.00 |
| TNL | PCal | 7.16 | 6.46 | 17.73 | 8.80 | 7.24 | 26.76 |
| | PTreat1 | 0.23 | 0.27 | 0.60 | 0.46 | 0.49 | 0.62 |
| | PTreat2 | 1.09 | 1.24 | 1.46 | 1.59 | 1.73 | 1.97 |
| | Mean | 2.82 | 2.66 | 6.59 | 3.62 | 3.16 | 9.78 |

† W1: Cattle-raising Watershed; W2: Afforested Watershed.

‡ PCal: Calibration Period; PTreat1: First year of treatment period; PTreat2: Second year of treatment period.

§ SS1= Weekly load estimated as the product of the daily flow recorded at the same day of manual sampling by concentration by seven. The concentration data was obtained from a single water sample collected manually.

¶ SS2= Weekly load estimated as the product of weekly flow by concentration. The concentration data was obtained from a single water sample collected manually.

# SS3= Weekly load estimated as the product of weekly flow by concentration. Concentration data was obtained from a flow-weighted water sample collected automatically during the week, or from a single water sample collected manually when automatic sampling was unavailable.

When our AYL values were compared with those reported by Cooper and Thomsen (1988), we found that our TPL values were lower in the cattle (0.62 versus 1.67 kg ha$^{-1}$ year$^{-1}$) and similar in the afforested (1.0 versus 0.95 kg ha$^{-1}$ year$^{-1}$) watershed. For TNL our estimates were also lower in the cattle (6.59 versus 11.95 kg ha$^{-1}$ year$^{-1}$) but higher in the afforested (9.78 versus 1.31 kg ha$^{-1}$ year$^{-1}$) watershed. Quinn and Stroud (2002) reported similar TPL and TNL values to those found by Cooper and Thomsen (1988) for cattle-raising and native-vegetation watersheds, although they used MM for sampling water and Cooper and Thomsen used AM.

Although as expected there were differences between the loads observed in our work and those reported by other authors for similar conditions, our values were within the reported range. This agreement could be due in part to the use of the SS3-generated data for comparison, which combines concentration data from both AM and MM and gave intermediate values between both sampling methodologies.

It was evident that in our work loads of TP and TN were not affected by changes in land use, but it is important to emphasize that our results only reflect the initial effects of afforestation on nutrient export, and therefore cannot be used to predict future trends with older trees. This observation is also applicable with respect to the afforestation effects on water quality.

## CONCLUSION

Significant TPC and TNC differences were not observed between the two watersheds during the evaluation period, indicating that afforestation did not affect these aspects of water quality. However, significant TPC and TNC differences were observed due to the sampling method; in both variables the concentration estimates obtained by AM were greater than those obtained by MM. Therefore, the effect of the water sampling method on nutrients concentration should be specifically considered when comparing water quality results.

TPC and TNC exceeded the minimum critical level reported by USEPA but not the maximum critical level. However, those reference values would not be directly applied to our results because they were obtained in rivers and streams, whereas our study was conducted in narrow canyons, with intermittent flow and sedimentation.

TP and TN loads were also not affected by changes in land use during the evaluation period although differences between sampling methods were found, similarly to those observed in TPC and TNC.

The load values obtained in our study were within the magnitude reported by other authors under similar conditions although we found some specific differences.

Our results refer only to the potential impact of the first years of afforestation on water quality. A long-term evaluation of the effect of changes in land use should be conducted to establish the real impact of mature plantations.

## REFERENCES

Agouridis, C.T, S.R Workman, R.C. Warner, and G.D. Jennings. 2005. Livestock grazing management impacts on stream water quality: a review. JAWRA 41:591–606.

Amatya, D.M., R.W. Skaggs, and J.W. Gilliam. 2006. Hydrology and water quality of a drained loblolly pine plantation in coastal North Carolina. In: W. Thomas (ed.). Hydrology and Management of Forested Wetlands. Proc. Int. Conf. ASABE, p. 15-26.

Birr, A.S. and D.J. Mulla. 2001. Evaluation of the phosphorus index in watersheds at the regional scale. J. Environ. Qual. 30:2018–2025.

Bremmer, J.M. and C.S. Mulvaney.1982. Nitrogen Total. p.595-624. In A.L. Page et al. (ed.) Methods of. soil analyses. Part 2. 2nd ed. Agron. Monogr. 9. ASA and SSSA, Madison, WI.

Chalar, G. 2006. (In Spanish) Eutrophication´s dynamics on different temporary scales: Salto Grande reservoir (Argentina-Uruguay). In Tundis, JG, Matsumura-Tundisi, T. & Sidagis, C. (ed.) Eutrophication in South America: causes, consequences and technologies for management and control. International Institute of Ecology, Inc. São Carlos, SP. p. 87–101.

Chalar, G., L. De León, E. Brugnoli, J. Clemente, and M. Paradiso. 2002. (In Spanish) Antecedents and new contributions to the knowledge of the structure and dynamic of the Salto Grande Dam. The water in South America: from Limnology to Administration in South America. Fernández-Cirelli, A. and Chalar, G. (eds). CYTED Use and Administration of Hydrologic Resources. Bs.As. 123–142p.

Chescheir, G.M., D.M. Amatya, and R.W. Skaggs. 2003a. Effects of afforestation on the hydrologic behavior of a basin in the Tacuarembó River. Progress report for 2002-03 submitted to Weyerhaeuser Foundation. Raleigh, N.C.: North Carolina State University, Department of Biological and Agricultural Engineering.

Chescheir, G.M., M.E. Lebo, D.M. Amatya, J. Hughes, J.W. Gilliam, R.W. Skaggs, and R.B. Herrmann. 2003b. Hydrology and water quality of forested lands in Eastern North Carolina, Technical Bulletin Nº 320, North Carolina Agric. Res. Service, North Carolina State University, Raleigh, NC, 79p.

Chescheir, G.M., N.O. von Stackelberg, D.M. Amatya, and R.W. Skaggs. 2004. Effects of afforestation on the hydrologic behavior of a basin in the Tacuarembó River. Progress report for 2003-04 submitted to Weyerhaeuser Foundation. Raleigh, N.C.: North Carolina State University, Department of Biological and Agricultural Engineering.

Chescheir, G.M., R.W. Skaggs, and D.M. Amatya. 2008. Hydrologic impacts of converting grassland to managed forestland in Uruguay. p. 1-9 In E.W. Tollner and A. Saleh (ed.) 21st Century Watershed Technology: Improving Water Quality and Environment. Proc. Int. Conf., Chile, 29 March – 3 April 2008. ASABE Publication Number 701P0208cd, ASABE, St. Joseph, MI. 49085-9659.

Clausen, J.C. and Spooner, J. 1993. Paired watershed study design. EPA 841-F-93-009, Office of Water, U.S. Environmental Protection Agency, Washington, DC

Cooper, A.B., and C.E Thomsen.1988. Nitrogen and phosphorus in streamwaters from adjacent pasture, pine, and native forest catchments. N. Z. J. Mar. Freshw. Res. 22:279–291.

DINAMA. 2007. (In Spanish) The quality of the Uruguay River. Results from the first three evaluation periods. 16p

Farley, K.A., E.G. Jobbágy, and R.B. Jackson. 2005. Effects of afforestation on water yield: a global synthesis with implications for policy. Glob. Change Biol. 11:1565–1576.

Fierer, N.G., and E.J. Gabet. Carbon and nitrogen losses by surface runoff following changes in vegetation. J. Environ. Qual. 31:1207–1213.

Garcia, P.F., J. Hernadez, P. Amarante, A. Califra, S. Delgado, L. Salvo, and M. Hill. 2004. (In Spanish) Effects of pine plantations in Uruguay: Annual report on soil quality. University of the Republic of Uruguay. Montevideo, Uruguay.

Gelbrecht, J., H. Lengsfeld, R. Pothing, and D. Opitz. 2004. Temporal and spatial variation of phosphorus input, retention and loss in small catchment of NE Germany. J. Hydrol. 304:151–165.

Grabow, G.L., J. Spooner, L.A. Lombardo, D.E. Line, and K.L. Tweedy. 1998. Has water quality improved?: Use of SAS for statistical analysis of paired watershed, upstream/downstream and before/after monitoring designs. U.S. EPA-NCSU-CES Grant No. X825012.

Hubbard, R.K., G.L. Newton, and G.M. Hill. 2004. Water quality and the grazing animal. J. Anim. Sci. 82:255–263.

Jones. R.C., and B.H. Holmes. 1985. Effects of land use practices on water resources in Virginia. Bulletin 144, Virginia Water Resources Res. Center, Virginia Polytechnic Institute and State University Blacksburg, 117p.

Khan, F.A., and A. Ansari A. A. 2005. Eutrophication: An Ecological Vision. Botan. Rev. 71:449–482

Marchesi, E. 2003. (In Spanish) The vegetation and flora of the watershed experiment, La Corona Ranch. Tacuarembó. COLONVADE S.A. Uruguay.

Molfino, J.H. 2000. (In Spanish) Project for installing experimental watersheds to study the impacts of Pine plantations in Uruguay. Departamento de Tacuarembó, COLONVADE S.A. Uruguay.

Murphy, J., and P. Riley. 1962. A modified single solution method for determination of phosphate in natural waters. Analyt. Chim. Acta. 229:115–120.

Nearing, M.A., R.M. Risse, and L.F. Rogers. 1993. Estimating daily nutrient fluxes to a large Piedmont reservoir from limited tributary data. J. Environ. Qual. 22:666–671.

O' Reagain, P.J., J. Brodie, G. Fraser, J.J. Bushell, C.H. Holloway, J.W. Faithful, and D. Haynes. 2005. Nutrient loss and water quality under extensive grazing in the upper Burdekin river catchment, North Queensland. Mar. Poll. Bull. 51:37–50

Owens, L.B., W.M. Edwards, and R.W. Keuren. 1991. Baseflow and stormflow transport of nutrients from mixed agricultural watersheds. J. Environ. Qual. 20:407–414

Oyarzun, C.E. and A. Huber. 2003. Nitrogen exports from forested and agricultural watersheds in the South of Chile. (In Spanish.) Gayana Bot. 60:63–68.

Pote, D.H. and T.C. Daniel. 2000. Analyzing for total phosphorus and total dissolved phosphorus in water samples. p. 94–97. In Pierzynski (ed.) Methods of phosphorus analysis for soils, sediments, residuals and waters. G.M.

Quinn, J. M. and J. M Stroud. 2002. Water quality and sediment and nutrient export from New Zealand hill-land catchment of contrasting land use. N. Z. J. Mar. Freshw. Res. 36:409–426

Quinton, J.N, J.A. Catt, and T.M. Hess. 2001. The selective removal of phosphorus from soil: is event size important? J. Environ. Qual. 30:538–545.

Rhine, E.D., G.K. Sims, and R.L. Mulvaney, E.J. Pratt. 1998. Improving the Berthelot reaction for determining ammonium in soil extracts and water. Soil Sci. Soc. Am. J. 62:473–480.

SAS Institute. 1990. SAS/STAT user`s guide. Versión 6. 4th ed. Vol.1. SAS Inst., Cary, N.C.

Thomas, G.W., G.R Haszler, and J.D. Crutchfield. 1995. Nitrate-nitrogen and phosphate-phosphorus in seven Kentucky streams draining small agricultural watersheds: eighteen years later. J. Environ. Qual. 21:147–50

U.S.E.P.A. 2002. Water quality standards. [Online]. Available at http://epa.gov/waterscience/criteria/nutrient/ecoregions/index.html (verified 14 Feb. 2008).

U.S.E.P.A. 2007. The nutrient criteria technical guidance manual: rivers and streams [Online]. Available at http://www.epa.gov/waterscience/criteria/nutrient/guidance/rivers/index.html (verified 14 Feb. 2008).

Udawatta, R.P., P.P. Motavallib, and H.E. Garretta. 2004. Phosphorus loss and runoff characteristics in three adjacent agricultural watersheds with claypan soils. J. Environ. Qual. 33:1709–1719.

von Stackelberg, N.O., G.M. Chescheir, and R.W. Skaggs. 2007. Simulation of the hydrologic effect of afforestation in the Tacuarembo River basin, Uruguay. Trans ASABE, 50:455–468.

Young, W.J., F.M. Marston, and J.R. Davis. 1996. Nutrient exports and land use in Australian catchments. J. Environ. Manag. 47:165–183

Zaimes, G.N. and R.C. Schultz. 2002. Phosphorus in agricultural watersheds: A literature review. Department of Forestry, Iowa State University, Ames, Iowa. 106p.

Zhang, L., W.R. Dawes, and G.R. Walker. 2001. Response of mean annual evapotranspiration to vegetation changes catchment scale. Water Resour. Res. 37:701–708.