# TEMPORAL DISTRIBUTION OF ANTI-HIV SEROLOGIC TESTS DEMAND AND POSITIVITY IN A MUNICIPAL CENTRAL LABORATORY: ARE THERE INCREASED AFTER CARNIVAL?

## Distribuição temporal da demanda e positividade de testes sorológicos anti-hiv em um laboratório central municipal: há aumento depois do carnaval?

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#### ABSTRACT

Introduction: HIV infection can lead to a progressive immunosuppression and result in an AIDS-related infections complex and other manifestations in affected individuals. Data from the AIDS 2012 Epidemiological Bulletin from 1980 to 2010 reported 241,662 deaths from AIDS in Brazil. Niterói, in Rio de Janeiro State, is a medium-sized city, of 500,000 inhabitants approximately and expressive socioeconomic and cultural indicators. Objective: To evaluate the relationship between seasonal demand and positivity of anti-HIV tests in the Miguelote Viana Public Health Central Laboratory located in Niterói. Methods: This is a temporal series analytical cross-sectional study. Anti-HIV tests demand, positivity and days worked by Miguelote Viana Public Health Central Laboratory were analyzed from a database for the period from 2005 to 2010. Data was then statistically evaluated through a temporal series and hypothesis testing on tendency and seasonality. Miguelote Viana Public Health Central Laboratory is a reference center for the dosage of viral load and CD4 levels for all public health units of Niterói; also attending to the population of cities in Metropolitan Region II. This is an innovative research, since articles that relate anti-HIV tests demand increase/decrease with the respective months of the year have not been found yet. As a result, we present graphs, tables and charts. Results: From January 2005 to December 2010, we have registered 64,505 serological tests for HIV, as follows: 17.44% (11,252) in 2005; 16.36% (10,557) in 2006; 17.81% (11,494) in 2007; 17.12% (11,046) in 2008; 16.20% (10,452) in 2009; and 15.04% (9,704) in 2010. In annual average, the days worked per month were as follows: 20 in 2005; 19.5 in 2006; 19.8 in 2007; 19.6 in 2008; 19.7 in 2009 and 19.3 in 2010. The monthly average of days worked in the six years studied was: 21 in Jan; 17.3 in February; 21.6 in March; 17.3 in April, 20 in May; 18.6 in June; 21.6 in July; 22 in August; 20.3 in September; 20.3 in October; 17.3 in November and 18.6 days in December. The annual average of positivity in absolute numbers was as follows: 42.6 in 2005; 44.0 in 2006; 38.3 in 2007; 32.8 in 2008; 24.25 in 2009 and 25.25 in 2010. The average positivity per month in the six years studied was the following: 39.3 in January; 29.3 in February; 40.8 in March; 31.8 in April; 31.1 in May; 34.6 in June; 33.8 in July; 38.6 in August; 35.0 in September; 34.8 in October; 31.5 in November and 33.6 in December. The average percentage of positivity per month was as follows: January (4.35), February (3.85), March (3.95), April (3.88), May (3.56), June (2.34), July (3.54), August (3.80), September (3.79), October (3.60), November (3.92) and December (3.75). In the studied period (2005-2010), Carnival holidays occurred in the month of February, on the following days: 8, 28, 20, 5, 24 and 16, respectively. Conclusion: We observed no seasonal relation between demand and positivity of anti-HIV tests carried out at Miguelote Viana Public Health Central Laboratory. A significant statistical decrease occurred in both anti-HIV tests demand and positivity during the studied years of the 2005-2010 series. Keywords: HIV, seasonality, public health, temporal analysis, carnival.

#### RESUMO

Introdução: A infecção pelo HIV pode levar à imunossupressão progressiva e resultar em um complexo de infecções relacionadas à AIDS e outras manifestações nos indivíduos acometidos. Dados do Boletim Epidemiológico AIDS 2012, relatam, de 1980 a 2010, 241.662 óbitos por AIDS no Brasil. Niterói, no estado do Rio de Janeiro, é um município de médio porte, com cerca de 500 mil habitantes e excelentes indicadores socioeconômicos e culturais. Objetivo: Avaliar a possível relação de sazonalidade existente entre a distribuição temporal da demanda e da positividade de testes sorológicos anti-HIV no Laboratório Central de Saúde Pública Miguelote Viana (LCSPMV), de Niterói, Rio de Janeiro. Métodos: Trata-se de um estudo transversal analítico de série temporal. Foram analisados os dados de demanda, de positividade dos exames anti-HIV e dos dias trabalhados, coletados de um banco de dados referentes ao período de 2005 a 2010. Os dados foram avaliados estatisticamente por uma série temporal e testes de hipótese para tendência e sazonalidade. O LCSPMV é referência na dosagem de carga viral e níveis de CD4 para todas as unidades de saúde da rede pública de Niterói e também atende à população oriunda dos municípios que fazem parte da Região Metropolitana II. Esta é uma pesquisa inovadora, visto que ainda não foram encontrados artigos que correlacionem aumentos/diminuições das demandas de exames anti-HIV com os respectivos meses dos anos. **Resultados:** No período de janeiro de 2005 até dezembro de 2010, registramos 64.505 exames sorológicos anti-HIV, sendo em 2005, 17,44% (11.252); em 2006, 16,36% (10.557); em 2007, 17,81% (11.494); em 2008, 17,12% (11.046); em 2009, 16,20% (10.452); e em 2010, 15,04% (9.704).

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Os dias trabalhados por mês foram, em médias anuais: 20 em 2005; 19,5 em 2006; 19,8 em 2007; 19,6 em 2008; 19,7 em 2009 e 19,3 em 2010. A média mensal de dias trabalhados nos 6 anos estudados foi: 21 em janeiro; 17,3 em fevereiro; 21,6 em março; 17,3 em abril; 20 em maio; 18,6 em junho; 21,6 em julho; 22 em agosto; 20,3 em setembro; 20,3 em outubro; 17,3 em novembro e 18,6 dias em dezembro. A positividade, em números absolutos, em média anual, foi de 42,6 em 2005; 44,0 em 2006; 38,3 em 2007; 32,8 em 2008; 24,25 em 2009 e 25,25 em 2010. Já a positividade por mês nos 6 anos estudados foi em média: 39,3 para janeiro; 29,3 para fevereiro; 40,8 para março; 31,8 para abril; 31,1 para maio; 34,6 para junho; 33,8 para julho; 38,6 para agosto; 35,0 para setembro; 44,8 para outubro; 31,5 para novembro e 33,6 para dezembro. A média de porcentagem de positividade por mês foi: janeiro (4.35%), fevereiro (3.85%), março (3,95%), abril (3,88%), maio (3,56%), junho (2,34%), julho (3,54%), agosto (3,80%), setembro (3,79%), outubro (3,60%), novembro (3,92%) e dezembro (3,75%). No período estudado, o feriado de carnaval ocorreu no mês de fevereiro, nos seguintes dias: 08, 28, 20, 05, 24 e 16, dos anos de 2005 a 2010, respectivamente. **Conclusão:** Não houve relação sazonal entre a demanda e a positividade de testes anti-HIV realizados no LCSPMV. Houve queda estatisticamente significativa na demanda e na positividade dos exames anti-HIV no decorrer dos anos estudados da série de 2005-2010. **Palavras-chave:** HIV, sazonalidade, saúde pública, análise temporal, carnaval.

### INTRODUCTION

Sexually transmitted diseases are considered one of the most common public health problems worldwide in both sexes, making the body more vulnerable to other diseases, including AIDS<sup>(1)</sup>.

Since the beginning of AIDS epidemic in 1983, according to the Epidemiological Bulletin of the Ministry of Health, until June 2012 Brazil has registered 656,701 cases of the disease, although the largest number of cases lies in the Southeast (56%). From 2000 to 2011, the incidence rate in that region dropped from 23.4 to 21.0 cases per 100,000 inhabitants. However, it has not occurred in other Brazilian regions, where the incidence rate has increased. Currently, the incidence rate in Brazil is of 20.8 cases per 100,000 inhabitants<sup>(1)</sup>.

Still according to the mentioned AIDS 2012 Epidemiological Bulletin, from 1980 to 2011, 253,706 deaths from AIDS occurred in Brazil. In this period, the State of Rio de Janeiro notified 40,817 deaths from the disease, making it the second State with the highest mortality rate from AIDS<sup>(1)</sup>. These data reflect the importance of studies on the population at risk and affected by HIV.

Niterói is a medium-sized city, with about 500,000 inhabitants and great socioeconomic indicators. It has the best alphabetization level in the State of Rio de Janeiro and holds one of the richest populations of Brazil, with 30.7% belonging to upper class<sup>(2)</sup>.

In the international extent, we can say that HIV infection remains one of the main priorities when health is concerned. Despite great improvement in preventing new infections and in reducing the number of annual deaths related to the virus, it can be noted that people infected with HIV continues to increase worldwide, thus predicting that AIDS will remain one of the leading causes of death worldwide over the next decades<sup>(3)</sup>.

Approximately, 10% of those infected with HIV progress to AIDS within the first two to three years of infection<sup>(4)</sup>. In general terms, the average time from infection to AIDS lasts for about 10 years<sup>(5)</sup>. However, a percentage between 5 and 8% maintains clinical stability without immune disease progression (even in the absence of treatment), maintaining CD4+ cell counts stable showing less chance to transmit HIV to other individuals<sup>(6,7)</sup>.

Demand studies can help detect nosological tendencies and also serve as a tool to guide health planning, including medical education and allocation of financial resources. These goals are usually achieved through methods, such as epidemiological investigations in population samples, household interviews in defined geographic areas and, especially, by mortality analysis<sup>(8)</sup>.

Presently, there are more cases of AIDS among men than among women, but this difference has been decreasing over the years.

The age between 13 and 19 years is the only group in which the incidence of AIDS is higher among women in our country.

Concerning the transmission mode in individuals over 13 years, the sexual way was observed to be the prevailing mode, and the heterosexual relations in women are responsible for 83% of cases, while in men 42% of case resulted from heterosexual relations, 22% from homosexual relations and 7.7% from bisexual relations, the remaining occurred by blood transfusion and vertical transmission<sup>(9)</sup>.

In the Brazilian context lies the Carnival, which is considered one of the liveliest festivals in the world. Its origin is the historic Portuguese carnival, in which people used to throw water, eggs and flour at each other. The carnival took place in a period before Quadragesima and had a meaning of freedom, which remains to this day<sup>(10)</sup>.

The strong permissive sex appeal of Carnival times exposes people to a risky behavior that might allow the development of STD and AIDS. For this reason, the Ministry of Health has been releasing strategies, such as media campaigns and distribution of male and female condoms during Carnival festivities and other places of public manifestations to prevent the increase of cases during these periods<sup>(9)</sup>.

We have decided to study the temporal demand distribution and the positivity of anti-HIV tests in a public health reference laboratory of Niterói, Rio de Janeiro, where there is a need for in-depth studies to understand the dynamics of the epidemiology of HIV infection involving Brazilian regions and popular festivals in a better way.

### **OBJECTIVE**

To evaluate the possible seasonal relation between the demand temporal distribution and the positivity of serologic anti-HIV tests at the Miguelote Viana Public Health Central Laboratory (LCSPMV), in Niterói – Rio de Janeiro, from January 2005 to December 2010.

### **METHODS**

This is an analytical cross-sectional study of the temporal series of anti-HIV tests among patients of both genders and different age groups conducted at LCSPMV, located in Niterói – Rio de Janeiro.

Demand and positivity data was analyzed for anti-HIV tests collected in a database and the days worked in the different months of each year from the retrospective survey of archived data dating from January 2005 to December 2010, in the LCSPMV Immunology and Surveillance Service located in Niterói, Rio de Janeiro.

The free consent term was not necessary, as we have used coded tables' data.

Project was approved by CEP under protocol number 244/11, dated September 2<sup>nd</sup>, 2011, and no conflict of interest was observed on this work.

The LCSPMV is a reference for CD4+ and viral load tests for all units of the public system of Niterói and the municipalities that are part of the Metropolitan Region II, including Niterói, São Gonçalo, Itaboraí, Maricá, Rio Bonito, Tanguá and Silva Jardim.

These samples arrive at the laboratory through the forwarding flow already existing on the system or they are collected in the laboratory by spontaneous demand.

The anti-HIV tests average/month in the LCSPMV is of 1,150, and release time of a negative result varies from 3 to 5 working days.

These samples and the results are forwarded likewise so that tests listed as released are available to every one of the nearly 60 units of Niterói, as well as to the other six neighboring municipalities when viral load or CD4+ are required.

The negativity and positivity criteria for HIV serologic tests used by LCSPMV are standardized by the Brazilian Ministry of Health, according to regulation 151 dated October, 2009<sup>(11)</sup>.

The research hypothesis is the increase of demand and positivity after Carnival in the city of Niterói, State of Rio de Janeiro.

We used our own and standardized forms for the collection of data from our study, containing the following items: number of days worked per month studied; tests collected per day; month and year studied; Carnival period occurred during years studied; and positive tests per month during the years studied.

Sequential graphs, boxplot, frequency histogram and decomposition of classical series described in previous study<sup>(12)</sup> were used as descriptive methods of a time series analysis.

For the inferential analysis, a linear regression model in time series and a set of indicator variables were adjusted regarding the studied months. The significance of the regression coefficients was used as a test for trend and seasonality. In addition, we use the cross-correlation coefficient followed by a significance test to evaluate the association between positivity and demands occurred in previous months<sup>(13,14)</sup>. All the hypotheses were tested with the adoption of a 5% level of significance<sup>(12,15)</sup>.

## RESULTS

Data was collected from January 2005 to December 2010, and curves graphs and tables were then elaborated. We aimed at discovering possible answers to the discrepancies found, and verify if there was a seasonality relation to the variables of the study.

From January 2005 until December 2010 we have registered 64,505 serological tests for HIV as follows: 17.44% (11,252) in 2005; 16.36% (10,557) in 2006; 17.81% (11,494) in 2007; 17.12% (11,046) in 2008; 16.20% (10,452) in 2009; and 15.04% (9,704) in 2010.

In annual averages the days worked per month were as follows: 20 in 2005; 19.5 in 2006; 19.8 in 2007; 19.6 in 2008; 19.7 in 2009 and 19.3 in 2010. The monthly average of days worked in the six years studied was: 21 in Jan; 17.3 in February; 21.6 in March; 17.3 in April; 20 in May; 18.6 in June; 21.6 in July; 22 in August; 20.3 in September; 20.3 in October; 17.3 in November and 18.6 days in December.

Positivity annual average in absolute numbers was as follows: 42.6 in 2005; 44.0 in 2006; 38.3 in 2007; 32.8 in 2008; 24.25 in 2009 and 25.25 in 2010. The average positivity per month in the six years studied was the following: 39.3 in January; 29.3 in February; 40.8 in March; 31.8 in April; 31.1 in May; 34.6 in June; 33.8 in July; 38.6 in August; 35.0 in September; 34.8 in October; 31.5 in November; and 33.6 in December.

The average percentage of positivity per month was as follows: January (4.35), February (3.85), March (3.95), April (3.88), May (3.56), June (2.34), July (3.54), August (3.80), September (3.79), October (3.60), November (3.92) and December (3.75).

In the studied period (2005–2010), all Carnival holidays occurred in the month of February, on the following days: 8, 28, 20, 5, 24 and 16, respectively.

After the statistical tests application, we observed a significant decrease in the anti-HIV tests' demand and positivity, and noticed that there was no seasonal influence on anti-HIV demand and positivity in the period between January 2005 and December 2010 in a reference laboratory in Niterói, Rio de Janeiro.

We present the results in the following graphs, charts, and tables:

| Table 1 - Months with meaningful data over the years of |
|---|
|---|

| Anti-HIV tests                         | 2005         | 2006        | 2007         | 2008                  | 2009         | 2010          |
|--|--------------|-------------|--------------|-----------------------|--------------|---------------|
| Minimum demand (absolute)              | July = 728   | May = 412   | Nov = 747    | Nov = 756             | Feb = 695    | Feb = 596     |
| Maximum demand (absolute)              | Mar = 1.150  | Aug = 1.189 | Jul = 1.172  | Dec = 1.509           | July = 1.062 | Mar = 1.054   |
| Lower daily demand                     | July = 34,66 | May = 21,68 | Aug = 44     | Jan = 38,66           | Jan = 36,4   | Dec = 35,23   |
| Higher daily demand                    | June = 52,25 | Oct = 55,05 | Jun = 54,11  | Dec = 83,83           | Nov = 55,61  | Mar = 47,9    |
| Less working days                      | Feb/Nov = 18 | Apr = 16    | Nov = 15     | Feb/May/June/Nov = 18 | Feb/Apr = 17 | Feb/Apr = 15  |
| More working days                      | Mar/Aug = 22 | Aug = 23    | Mar/Aug = 23 | July = 23             | Jul = 23     | Mar/Aug = 22  |
| Minimum number of positives (absolute) | Jan = 31     | May = 10    | Sep = 26     | Oct = 24              | Oct = 12     | June = 15     |
| Maximum number of positives (absolute) | Mar = 52     | June = 57   | Jan = 58     | Dec = 1.509           | Apr = 31     | Mar/July = 32 |
| Lower positivity                       | Jan = 3,33%  | May = 2,42% | July = 2,30% | Dec = 2,05%           | Oct = 1,37%  | June = 2,34%  |
| Higher positivity                      | Oct = 5,33%  | Jan = 6,6%  | Jan = 5,43%  | Sep = 4,88%           | Apr = 3,94%  | Feb = 3,69%   |

### Descriptive analysis of average temporal demand

As a data pretreatment, each observation made on the demand series was divided by the number of working days of the corresponding month. This procedure avoids introducing any vices in the analysis, as there are months, for which amount of tests is inferior to the usual average due to the excess of holidays.

In many statistical analysis procedures such as hypothesis testing and regression models, one works with the supposition that data are normally distributed. However, this does not always occur, and the frequency histogram is a very useful tool to check this assumption.

As observed in the Frequency Histogram in **Graph 4**, there is a tendency of concentrating the observations around the average, and there are values at the margins that correspond to the months of May 2006 and December 2008.

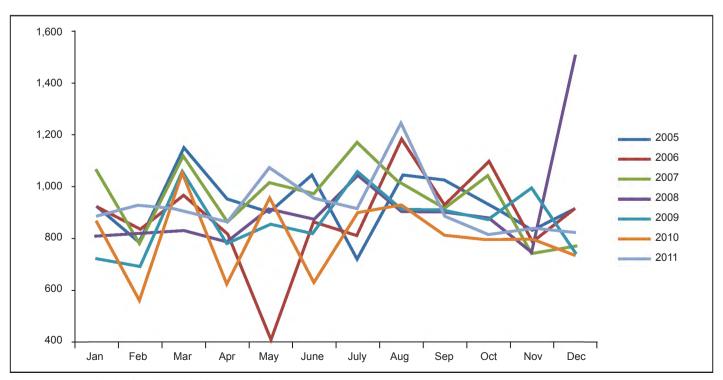
A normal curve was superposed to the graph presented, and the average and standard deviation were obtained through the observations made. The noticed distribution differs from the normal distribution in two aspects: the concentration around the average is above the expected value and the extreme values do not seem compatible with the normal distribution.

For the construction of the normal curve in **Graph 5**, we used the average demand in the period observed equal to 45.53 and standard deviation equal to 7.04.

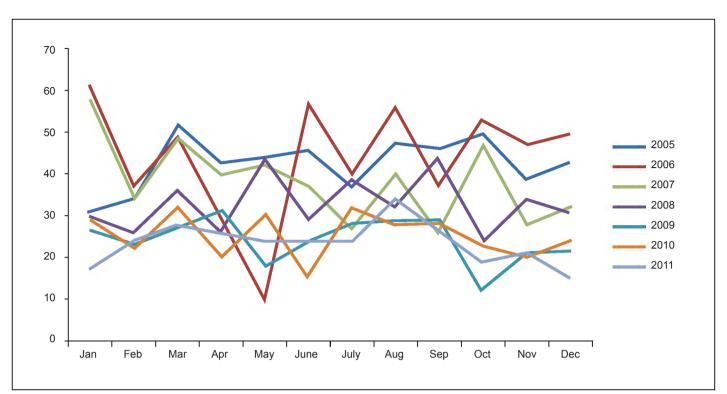
The criterion for identifying atypical points, known in statistics as outliers, uses boxplot chart boundaries. The two atypical points identified

Table 2 - Live births in Niterói of mothers resident in Niterói

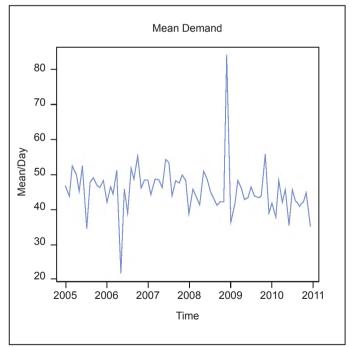
|           | 2005  | 2006  | 2007  | 2008  | 2009  | 2010  | 2011  | Total  |
|-----------|-------|-------|-------|-------|-------|-------|-------|--------|
| January   | 406   | 366   | 430   | 387   | 466   | 420   | 406   | 2.861  |
| February  | 400   | 356   | 412   | 357   | 321   | 423   | 396   | 2.665  |
| March     | 486   | 411   | 438   | 395   | 412   | 432   | 461   | 3.035  |
| April     | 418   | 406   | 459   | 395   | 463   | 423   | 435   | 2.999  |
| Мау       | 450   | 428   | 455   | 429   | 407   | 370   | 425   | 2.964  |
| June      | 425   | 423   | 430   | 380   | 397   | 401   | 446   | 2.902  |
| July      | 390   | 395   | 417   | 436   | 402   | 419   | 430   | 2.889  |
| August    | 381   | 375   | 396   | 395   | 357   | 369   | 421   | 2.694  |
| September | 365   | 379   | 388   | 417   | 446   | 386   | 393   | 2.774  |
| October   | 461   | 403   | 354   | 361   | 394   | 371   | 361   | 2.695  |
| November  | 369   | 369   | 341   | 379   | 368   | 387   | 399   | 2.612  |
| December  | 359   | 352   | 379   | 397   | 396   | 398   | 415   | 2.693  |
| Total     | 4.897 | 4.663 | 4.899 | 4.728 | 4.809 | 4.799 | 4.988 | 33.783 |



Graph 1 - Monthly absolute demand of years studied.

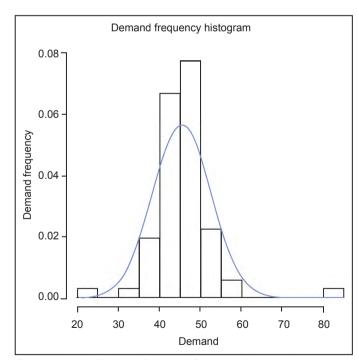


Graph 2 - Monthly absolute positive over years studied.



Graph 3 – Demand series from 2005 to 2010.

in the graph above represent the demand observed in May 2006 and December 2008. To avoid the influence of these points in future analyses, we decided to replace them by the corresponding month average calculated in the remaining data, and then perform the analysis again.



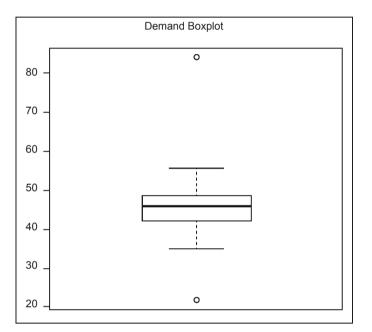
Graph 4 – Distribution of demand frequency from 2005 to 2010.

As presented in **Graph 6**, the monthly averages (central lines of boxes in boxplots) under visual inspection are not distant. In this graph specifically we did not attribute too much importance to the width of the box (which would represent the

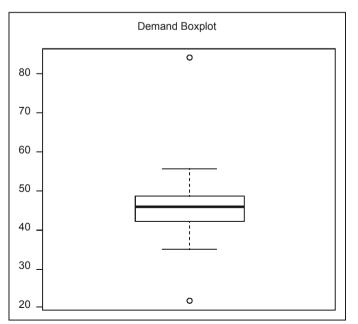
variability of a given month), as there are only six comments in each month.

### Descriptive analysis of the positivity temporal series

The same analysis was carried out with the demand series. Initially, we show in **Graph 7** the positivity time evolution. It can be noticed in this graph that there has been a strong decrease tendency in the positivity series over the years. This trend is emphasized as from 2007.



Graph 5 - Demand boxplot from 2005 to 2010.

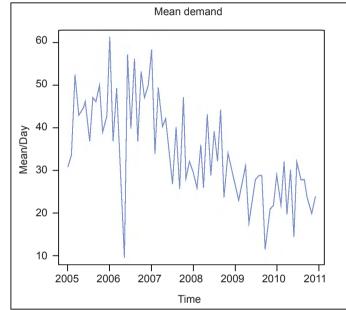


Graph 6 - Demand boxplot for HIV tests classified by observation month.

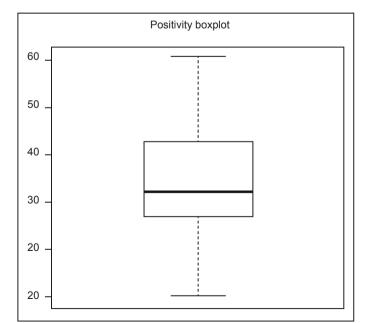
The significant boxplot usefulness during the demand series descriptive analysis is to reveal some outliers, atypical values. We repeat this procedure in **Graph 8** for the positivity series.

The analysis of the positivity series through the boxplot reveals that the comments are in a range varying from 10 to 60 cases with the median around 30 cases (the central line of the box). Contrary to what occurs with the demand series, there is no evidence of atypical observations because no point overflows the borders of the graph.

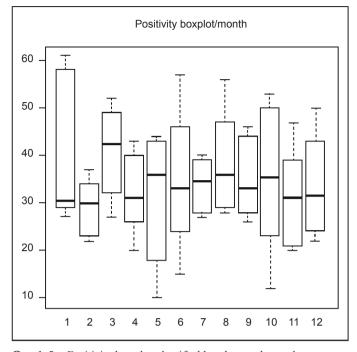
**Graph 9** shows that the median of the months of March (3) is above the other months, however, by the size of the box, we realize



Graph 7 – Positivity series from 2005 to 2010.



Graph 8 - Positivity boxplot from 2005 to 2010.



Graph 9 - Positivity boxplot classified by observed month.

there is a great data variability, although we emphasize we only have six observations for each month.

## Classic decomposition of demand and positivity series in HIV tests

According to Morettin<sup>(12)</sup>, the decomposition model (additive) of the temporal series assumes that the time series can be decomposed into three unobservable components: T(t), S(t) and a(t). These components represent Trend, Seasonality and Randomness, respectively. Therefore, an observation of a time series can be described as follows:

Z(t) = T(t) + S(t) + a(t)

if these components interact in an additive way or:

 $Z(t) = T(t) \ge S(t) \ge a(t)$ 

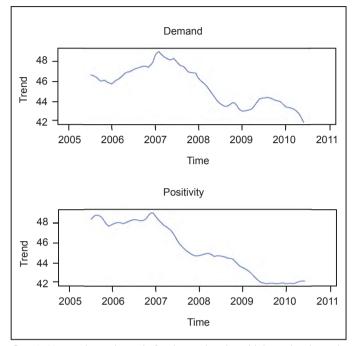
if the relation between them is multiplicative.

In this work, we will use the additive model to verify the magnitude of Trend (T(t)) and Seasonality (S(t)) components and look for an interpretation. We point out that this phase is still exploratory and that no tests of hypotheses about the results will be carried out.

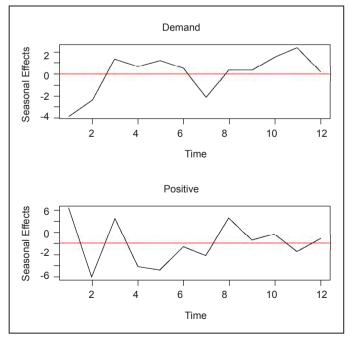
The results were obtained through the *decompose* function, available in the R statistical package. With this function, trend and seasonality are both estimated by the centered moving averages method.

**Graph 10** shows the estimated trend of both the demand and positivity series during the data observation period. In both series, it is possible to highlight a decline of values, and this decrease in the positivity series becomes more "accelerated". Apparently, 2007 is the beginning of this behavior change in both charts.

**Graph 11** illustrates the seasonal effects estimated for each month. For the interpretation of this graph, consider horizontal line represents the average behavior of the series. There is an estimate of the associated increase or decrease in demand (or positivity) for each month in relation to the global average.



**Graph 10** – Estimated trends for demand and positivity series through centered moving means (decompose function of R package).



**Graph 11** – Seasonal effects estimated for demand and positivity series through centered moving averages (decompose function of the R package).

Among other facts, we can observe in **Graph 11** that demand in January and February is below the average, while the number of positivity cases in January, March and August is above the average.

Both the analysis of the demand and positivity performed until then had a merely exploratory character. To obtain statistical significance of the results, we will adjust a regression model to both series that includes a term to describe the linear trend series and the months of the year as explanatory variables. The significance of the trend as well as seasonality will be linked directly to the parameters of the regression model.

## Linear regression adjustment to verify Trend and Seasonality in Demand and Positivity

To verify the importance of both trend and seasonal component, two linear regression models were adjusted to demand and positivity, respectively. These terms were included in the regression models (intercept and time) to measure the linear trend and indicator variables for each month using the month of December as the reference month.

The adjusted linear regression model for the demand series produced the results presented in **Table 3**.

As expected, there is a significant decrease trend in demand illustrated by the negative sign of the coefficient associated with the variable time. The value -0.0931 indicates the average decrease on demand/day in each month elapsed. The *p*-value for this parameter is below the level of significance of 1%.

Despite the evident trend importance, the effects of months' estimates did not show statistical significance. This fact indicates there is enough evidence in data to support the assumption of a difference in demand due to a given month of the year.

The same model adjusted to the demand series was also adapted to the positivity series as well. The conclusions regarding trend and seasonality significance are similar. According to **Table 4**, there is a significant decrease in positivity (-0.365 per month) and a higher acceleration if compared with the demand series. However, there is no statistical significance on the effects caused by the months of the year.

### Cross-correlation analysis between Demand and Positivity Series

In this study, we verify if there is a correlation between demand in a given month and positive cases in future months. For this purpose, we use the cross-correlation function.

The cross-correlation measure requires that two series are stationary, and due to this fact, we use the adjusted models in Section

### Table 3 - Adjustment results of multiple linear regression model for the demand series

| Coefficients | Estimates | Standard error | <i>t</i> -statistics | <i>p</i> -value |
|--------------|-----------|----------------|----------------------|-----------------|
| Intercept    | 47.6945   | 1.9825         | 24.06                | 0.0000          |
| Time         | -0.0931   | 0.0240         | -3.87                | 0.0003          |
| January      | -2.5613   | 2.4275         | -1.06                | 0.2957          |
| February     | -1.6366   | 2.4250         | -0.67                | 0.5024          |
| March        | 2.8715    | 2.4227         | 1.19                 | 0.2407          |
| April        | 1.9862    | 2.4207         | 0.82                 | 0.4152          |
| Мау          | 1.5846    | 2.4189         | 0.66                 | 0.5150          |
| June         | 2.2423    | 2.4174         | 0.93                 | 0.3574          |
| July         | -0.3680   | 2.4161         | -0.15                | 0.8795          |
| August       | 1.2734    | 2.4150         | 0.53                 | 0.6000          |
| September    | 1.0615    | 2.4141         | 0.44                 | 0.6618          |
| October      | 2.2012    | 2.4135         | 0.91                 | 0.3655          |
| November     | 3.4559    | 2.4132         | 1.43                 | 0.1574          |

Table 4 - Results of the adjustment of multiple linear regression model for the positivity series

| Coefficients | Estimates | Standard error | <i>t</i> -statistics | <i>p</i> -value |
|--------------|-----------|----------------|----------------------|-----------------|
| Intercept    | 48.8500   | 4.1002         | 11.91                | 0.0000          |
| Time         | -0.3615   | 0.0497         | -7.27                | 0.0000          |
| January      | 1.6901    | 5.0204         | 0.34                 | 0.7376          |
| February     | -7.9484   | 5.0153         | -1.58                | 0.1183          |
| March        | 3.9131    | 5.0106         | 0.78                 | 0.4379          |
| April        | -4.7254   | 5.0064         | -0.94                | 0.3491          |
| May          | -5.0306   | 5.0027         | -1.01                | 0.3187          |
| June         | -1.1690   | 4.9995         | -0.23                | 0.8159          |
| July         | -1.6409   | 4.9968         | -0.33                | 0.7438          |
| August       | 3.5540    | 4.9945         | 0.71                 | 0.4795          |
| September    | 0.2488    | 4.9928         | 0.05                 | 0.9604          |
| October      | 0.4437    | 4.9916         | 0.09                 | 0.9295          |
| November     | -2.5282   | 4.9908         | -0.51                | 0.6143          |

3.4 for the calculation of this measure. We emphasize that when working with residues, the trend components of both series were removed and, therefore, it is reasonable to assume that these series are stationary.

The cross-correlation analysis result is condensed in Graph 12.

The limits in blue (dotted line) on **Graph 12** work as critical values for the cross-correlation values. Values exceeding the limits indicate significant correlations between the demand and the positivity in an overdue moment. When the difference (Lag) is in value 0, we evaluate the immediate correlation between demand and positivity. As all values are within limits, we conclude there is no evidence of cross-correlation between the demand and positivity series. Under a practical point of view, the values observed for the demand in a given month do not help predict the positivity in future months.

Chart 1 shows the positivity percentage average by month as well as minimum and maximum per month.

## DISCUSSION

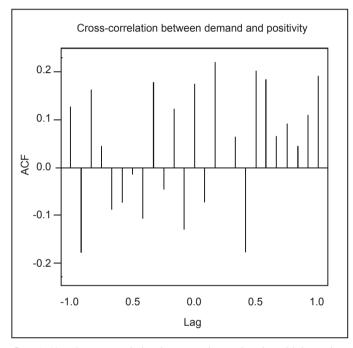
After searching in the main database (Lilacs, SciELO, MedLine, PubMed, Scopus, Web of Science) for the past ten years, we noted it was difficult to find articles focused on the relation between anti-HIV tests seasonality, demand and positivity. In fact, we found few publications on the topic, hindering the visibility of the problem, and consequently the implementation of priority interventions and subsequent evaluations of their effectiveness.

The number of working days varies from month to month, as shown in data collection and statistical analysis of the years studied, not only due to the absolute number of days, which can range from 28 to 31, but also to holidays, which occur mostly during February, April and December, and could misdirect our analysis.

In a study about risky behavior during Carnival based on questionnaire filled in by percussionists (men) of a samba school from São Paulo, Hughes *et al.* concluded that those who were at risk only during Carnival did not differed from the ones who were at risk other times<sup>(16)</sup>. PASSOS et al.

This reinforces the understanding that who is at risk in a successful event as Carnival has a huge potential to be at risk throughout the year.

To confirm the hypothesis that anti-HIV tests demand and positivity distribution does not follow a rule, in other words, it does not present a typical temporal distribution, and it does occur at random, Lima *et al.*, in a publication on massive campaigns of STD/AIDS released by the federal Government, reinforce that the calendar is fixed, only contributing to build the imaginary of AIDS in the country's scenario<sup>(17)</sup>.



**Graph 12** – Cross-correlation between demand and positivity series (ccf function of R package).

| <b>Chart 1 –</b> Positivity | Percentages 2005-2010 |
|-----------------------------|-----------------------|
|-----------------------------|-----------------------|

|            |          |          |          | Posi     | tivity   |          |       |       |       |
|------------|----------|----------|----------|----------|----------|----------|-------|-------|-------|
| Year/Month | 2005     | 2006     | 2007     | 2008     | 2009     | 2010     | Med.  | Min.  | Max.  |
| January    | 3.33%    | 6.60%    | 5.45%    | 3.69%    | 3.71%    | 3.32%    | 4.35% | 3.33% | 6.60% |
| February   | 4.32%    | 4.43%    | 4.28%    | 3.16%    | 3.31%    | 3.69%    | 3.85% | 3.16% | 4.43% |
| March      | 4.52%    | 5.04%    | 4.38%    | 4.31%    | 2.54%    | 3.03%    | 3.96% | 2.54% | 5.04% |
| April      | 4.50%    | 3.80%    | 4.60%    | 3.31%    | 3.94%    | 3.18%    | 3.88% | 3.18% | 4.60% |
| Мау        | 4.89%    | 2.42%    | 4.13%    | 4.72%    | 2.10%    | 3.14%    | 3.56% | 2.10% | 4.89% |
| June       | 4.40%    | 6.56%    | 3.80%    | 3.30%    | 2.92%    | 2.34%    | 3.88% | 3.30% | 6.56% |
| July       | 5.08%    | 4.92%    | 2.30%    | 3.73%    | 2.63%    | 3.54%    | 3.7%  | 2.30% | 5.08% |
| August     | 4.49%    | 4.71%    | 3.95%    | 3.51%    | 3.16%    | 3.01%    | 3.80% | 3.01% | 4.71% |
| September  | 4.48%    | 3.99%    | 2.84%    | 4.88%    | 3.18%    | 3.43%    | 3.79% | 2.84% | 4.88% |
| October    | 5.33%    | 4.81%    | 4.50%    | 2.72%    | 1.37%    | 2.88%    | 3.60% | 1.37% | 5.33% |
| November   | 4.71%    | 6.00%    | 3.74%    | 4.49%    | 2.10%    | 2.49%    | 3.92% | 2.10% | 6.00% |
| December   | 4.70%    | 5.44%    | 4.14%    | 2.05%    | 2.97%    | 3.24%    | 3.75% | 2.05% | 5.44% |
| Med.       | 4.56%    | 4.89%    | 4.01%    | 3.65%    | 2.82%    | 3.10%    |       | 2.60% | 5.29% |
| Min.       | 3.33%    | 2.42%    | 2.30%    | 2.05%    | 1.37%    | 2.34%    | 2.30% |       |       |
| Max.       | 5.33%    | 6.56%    | 5.45%    | 4.88%    | 3.94%    | 3.69%    | 4.97% |       |       |
| Carnival   | 08/02/05 | 28/02/06 | 20/02/07 | 05/02/08 | 24/02/09 | 16/02/10 |       |       |       |

As a way to strengthen our argumentation, we have researched the number of live births of mothers of Niterói who gave births in this city<sup>(18)</sup>. The result shows that the month of November, which comes nine months after carnivals that occurred in February, presents the lower absolute number of total births in the 2005–2010 series. Thus, it is reasonable to suppose that these data weaken the theory that there is a greater number of unprotected sexual intercourse during the Carnival period.

Still in the range of other STDs, there is an important time series study published by our research group, Passos *et al.*, in 2010. Along 12 years, our study concluded that no increase of syphilis, gonorrhea and trichomoniasis is associated with Carnival<sup>(19)</sup>.

On the data analysis of **Chart 2**, it can be noted at first that the months of August have greater absolute demands, and the months of May have smaller absolute demands, when compared to the other months of the year, except December 2008. However, these differences do not show statistical significance, a fact verified after analysis through tests of significance.

In fact, the discrepancy in the absolute demand found in December 2008 is explained from an event, by public health activities, such as the "Worldwide Day of Fight against AIDS" (December 1<sup>st</sup>), known as the governmental campaign "For the Record" (STDs, AIDS and Viral Hepatitis Department of the Health Ministry). This encourages HIV testing by general population<sup>(20)</sup>.

It is important to mention that the second edition of the same campaign, between November and December 2009, did not achieve the repercussions of the 2008 campaign in the city of Niterói. However, we point out that despite the demand increase in anti-HIV tests in 2008, there was no rise in the number nor in the percentage of anti-HIV test positivity in relation to the demand<sup>(21)</sup>.

Although the largest number of AIDS cases in Brazil's Southeast region (56%), the incidence rate in this region has been decreasing over the years<sup>(1)</sup>. Several factors may be contributing to this decrease. However, we found no publications to help us understand this situation. We believe that more people diagnosed with HIV

associated with a large number of people in use of Highly Active Antiretroviral Therapy and the dissemination of information can contribute to the stabilization/reduction of people living with HIV.

The present study shows that our initial hypothesis was not true, and that demand and positivity of anti-HIV test decreased significantly and also there was no seasonal interference throughout the period studied.

It is worth mentioning that the dissemination of useful information on sexuality issues, STD, HIV, will certainly benefit many people. However, this cannot be diffused as the primary factor to impact on the epidemiology of such a complex disease as AIDS and other STDs. Moreover when the information occur more intensely in specific times of the year (December 1<sup>st</sup> and Carnival).

Occasionally intern problems interfere (or hinder) with the dissemination of the "educational campaigns on HIV" promoted by the Brazilian Ministry of Health, which is an additional complication factor, as occurred in the last campaign of the Ministry of Health of Brazil<sup>(22–27)</sup>.

As a limitation of the study, we should mention that it was not possible to separate the repeated tests.

Another limitation of this study is that it is about a single service located in a single city. However, we emphasize that Niterói is a medium-sized city, but it is a reference to several other municipalities in the metropolitan region of Rio de Janeiro. We point out, however, that the laboratory involved in this work is available for a population of more than 1,974,911 inhabitants for viral load and CD4 levels testing. The population distributions of the cities are as follows: Silva Jardim, 21,362; Tanguá, 31,438; Rio Bonito, 56,436; Maricá, 135,121; Itaboraí, 222,618; Niterói, 491,807; and São Gonçalo, 1,016,128 inhabitants<sup>(28)</sup>.

Although our "n" has been of 64,505 tests, this is an analysis of only one laboratory, and although it is a reference laboratory in the city, the analysis cannot be amplified for the national level. Our suggestion is to encourage a research with greater scope to compare the results obtained.

Chart 2 - Absolute demand of anti-HIV test: 2005-2010

| Absolute Demand |          |          |          |          |          |          |       |       |       |
|-----------------|----------|----------|----------|----------|----------|----------|-------|-------|-------|
| Year/month      | 2005     | 2006     | 2007     | 2008     | 2009     | 2010     | Med.  | Min.  | Max.  |
| January         | 931      | 924      | 1063     | 812      | 728      | 873      | 888,5 | 728   | 1063  |
| February        | 787      | 835      | 793      | 823      | 695      | 566      | 749,8 | 566   | 835   |
| March           | 1150     | 971      | 1118     | 834      | 1060     | 1054     | 1150  | 834   | 1150  |
| April           | 956      | 815      | 870      | 785      | 787      | 629      | 807   | 629   | 956   |
| May             | 900      | 412      | 1016     | 911      | 857      | 955      | 841,8 | 412   | 1016  |
| June            | 1045     | 869      | 974      | 877      | 821      | 639      | 870,8 | 639   | 1045  |
| July            | 728      | 812      | 1172     | 1045     | 1062     | 904      | 953,8 | 728   | 1172  |
| August          | 1047     | 1189     | 1012     | 911      | 917      | 930      | 1001  | 911   | 1189  |
| September       | 1027     | 927      | 913      | 901      | 910      | 815      | 915,5 | 815   | 1027  |
| October         | 938      | 1101     | 1044     | 882      | 874      | 797      | 939,3 | 797   | 1101  |
| November        | 828      | 783      | 747      | 756      | 1001     | 802      | 819,5 | 747   | 1001  |
| December        | 915      | 919      | 772      | 1509     | 740      | 740      | 932,5 | 740   | 1509  |
| Med.            | 937,6    | 879,75   | 957,8    | 920,5    | 871      | 808,6    |       | 808,6 | 957,8 |
| Min.            | 728      | 412      | 747      | 756      | 695      | 566      | 749,8 |       |       |
| Max.            | 1150     | 1189     | 1172     | 1509     | 1062     | 1054     | 1150  |       |       |
| Carnival        | 08/02/05 | 28/02/06 | 20/02/07 | 05/02/08 | 24/02/09 | 16/02/10 |       |       |       |

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We recommend the conduction of similar studies in all Brazilian regions, so we can know the reality of this subject in Brazil.

### CONCLUSION

We observed no relation of seasonality neither with demand nor positivity of anti-HIV tests carried out in LCSPMV.

We noticed no increase in the anti-HIV serological tests demand and/or positivity for anti-HIV test after Carnival in LCSPMV, in Niterói, Rio de Janeiro.

We found a significant anti-HIV tests decrease in both demand and positivity along the years studied in the 2005-2010 series.

Hence, it follows that the main event setting the rules of variables distribution along the years was randomness and not seasonality, like common sense could think.

### **Conflict of interests**

The authors declared no conflict of interests.

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