BOVINE RABIES INCIDENCE IN THE STATE OF MINAS GERAIS/BRAZIL, BETWEEN 2002 AND 2006*

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ABSTRACT

Rabies is an infectious contagious disease of mammals present in all continents except Oceania. It is caused by a Lyssavirus and characterized by acute fatal encephalitis. It is considered endemic in Brazil and transmission to herbivores occurs mainly via vampire bats. The purpose of this study was to evaluate the incidence of bovine rabies in the state of Minas Gerais, as well as the diagnostic methods, applying the time series to analyze trend and seasonality in order to adjust a model to disease in cattle. Between 2002 and 2006, nervous tissue samples were obtained from deceased bovines presenting neurological symptoms and clinically suspected of rabies. The direct fluorescent antibody test (FAT) detected 40.38% (1,293/3,202) positive samples. From all FAT negative tissue samples, 2.67% (51/1,909) tested positive in a mouse inoculation test (MIT). No seasonality was observed and positive cases had a decrease of 0.14 per month.

KEY WORDS: Lyssavirus, bovines, rabies, time series.

RESUMO

INCIDÊNCIA DE RAIVA BOVINA NO ESTADO DE MINAS GERAIS, BRASIL, ENTRE 2002 E 2006. A raiva é uma doença infecto-contagiosa dos mamíferos presente em todos os continentes com exceção da Oceania. É causada por um Lyssavirus e caracterizada como uma encefalite aguda fatal, sendo considerada endêmica no Brasil e a transmissão aos herbívores ocorre principalmente através de morcegos hematofagos. O objetivo deste experimento foi avaliar a incidência de raiva bovina no Estado de Minas Gerais, assim como métodos de diagnóstico, empregando séries temporais para analisar a tendência e sazonalidade, podendo assim ajustar um modelo da doença em bovinos. Entre 2002 e 2006, amostras de tecido nervoso foram obtidas de bovinos que vieram a óbito com sintomas nervosos e com suspeita clínica de raiva. A imunofluorescência direta (IFD) detectou 40,38% (1.293/3.202) de amostras positivas. Das amostras de tecido consideradas negativas na IFD, 2,67% (51/1.909) foram consideradas positivas utilizando o teste de inoculação em camundongos (IC). Não foi observada sazonalidade e houve uma queda de 0,14 casos positivos ao mês.

PALAVRAS-CHAVE: Lyssavirus, bovinos, raiva, séries temporais.

In Brazil, the establishment of guidelines for the control of rabies of herbivores is performed by the Ministério da Agricultura, Pecuária e Abastecimento (MAPA) (Ministry of Agriculture, Cattle Breeding and Provisioning), while for urban rabies (dogs and cats) control is carried out by the Ministério da Saúde (MS) (Health Ministry).

In cattle, infections of the central nervous system (CNS) include a group of important diseases, however only rabies is responsible for annual losses of around 50 million US dollars worldwide (KING; TURNER, 1993).

Only in the beginning of last century bovine rabies was described in Brazil (CARINI, 1911). Historically, its distribution and determinations were influenced by economically induced changes in the agricultural space.

Rabies is an important zoonotic viral disease to livestock and public health in Brazil, it is estimated in approximately 850,000 heads the annual loss of cattle due to rabies, approximately equivalent to 17 millions of US dollars (LIMA et al., 2005).
Clinical observation may only lead to a suspicion of rabies because signs of the disease are not characteristic and may vary greatly from one animal to another. The only way to undertake a reliable diagnosis of rabies is to identify the virus or some of its specific components using laboratory tests. The most widely used test for rabies diagnosis is the fluorescent antibody test (FAT), which is recommended by both WHO and OIE, and is sensitive, specific and cheap (OIE, 2011). The isolation of the rabies virus, through mouse inoculation test (MIT), confirms the results obtained by other techniques, although its use is expensive and slow (Bourhy; Sureau, 1990) and detects the infectivity of a tissue suspension in laboratory animals. It should be used if the FAT gives an uncertain result or when the FAT is negative in the case of known human exposure (OIE, 2011).

Rabies in herbivores in Brazil is determined mostly by the presence of the vampire bat. Bovine and equine may be included among the animal species most affected by the vampires (Peixoto et al., 2000). The techniques for herbivore rabies control are based on anticoagulant drugs (selective direct and indirect methods) and were developed according to the biology, habitat, range of action, feeding behavior, social organization and specific behavior of the vampire bat. The low reproductive rates (7 months long pregnancy with production of a single offspring per year) favor population control of this species (Brasil, 2005).

The Programa Nacional de Controle da Raiva dos Herbívoros (PNCRH) (National Program for Herbivore Rabies Control) is aimed at lowering the prevalence of the disease in domestic herbivores populations. Its strategy is based mainly on epidemiologic surveillance, targeted vaccination of domestic herbivores and control of the vampire bat Desmodus rotundus (Brasil, 2002).

In developed countries, the seasonal adjustment of time series has long been incorporated as an official practice, particularly after the advent of computerized techniques (Arita; Dias, 2000). However, the adoption of a large scale adjustment technique should be carefully elaborated in developing countries such as Brazil, which are frequently exposed to strong structural and conjunctural changes (Hotta, 1988) leading to irregularities that may compromise adjustment employment and results (Dagum, 1978).

The purpose of this study was to evaluate the incidence of bovine rabies in the state of Minas Gerais, as well as the diagnostic methods, applying the time series to analyze trend and seasonality in order to adjust a model to disease in cattle.

The samples of animals clinically suspected of rabies were collected by official and self-employed veterinarians in all 853 municipalities of the state of Minas Gerais.

Central Nervous System (CNS) samples of cattle were analyzed by direct fluorescent antibody test (FAT) and mouse inoculation test (MIT), the latter was used in all samples that were negative on FAT.

The FAT test detects viral antigens in the brain using labeled fluorescein as rabies virus antibodies. FAT is considered the gold standard for diagnosing rabies in fresh specimens. The mouse inoculation test (MIT) is traditionally used as a quality control method for FAT.

These analyses were performed at the Laboratório de Saúde Animal (LSA) (Animal Health Laboratories) of the Instituto Mineiro de Agropecuária (IMA) (Institute of Agriculture and Cattle Raising of Minas Gerais) and Laboratório de Zoonoses da Secretaria Municipal de Saúde de Belo Horizonte (LZ/SMS/BH) (Zoonosis Laboratory of the Municipal Health Secretariat of Belo Horizonte) that are reference laboratories.

A computerized database of the results was then created. Electronic charts were used to display all municipalities of Minas Gerais in columns, with the positive results, yearly, from 2002 to 2006 characterizing 60 points (months) for analysis of bovine rabies.

In this study, the statistical analysis was performed monthly along the time of positive results counting within the aforementioned timeframe, characterizing a time series. The extraction of the regularities contained in noticeable phenomena of a time series enables the construction a mathematical model like a simplified representation of reality, once there are various possible methods for the study of a time series and characterization of trend and seasonality, when pertaining. Another important factor is that time series studies are used for variables which are self-correlated, that is, those that are correlated with themselves along time.

The Shapiro; Wilk (1965) test was used to verify the hypothesis of residual normality and to allow the use of the methodology of Box and Jenkins. The Cox-Stuart signal test (Moretti; Tolo, 2006) was employed to verify the significant existence of a trend and parameters of the linear model was adjusted to represent it. The linear model was used to establish a trend, that is, if a statistical significance was observed in the linear model there was a trend.

The Box and Jenkins’ statistical model was selected among current classes of models that are considered adequate for this representation. Following formulation of the model and estimation of its parameters, it was employed to test the hypothesis concerning the generating mechanism of the stochastic process (times of greater incidence of bovine rabies) and to predict future values of the time series.
The Fisher Test proposed by Priestley (1989) was employed to test the occurrence of seasonality, with the level of significance set at 5%. The Statistica software (5.1 version from 1996) from Statsoft® was used and the following statistical model was created:

$$y_t = T_{(t)} + S_{(t)} + a_t$$

where:
- $T_{(t)}$ = effect of trend, characterized as a gradual increase or reduction of the observations along a period of time;
- $S_{(t)}$ = effect of seasonality, characterized by the presence of repetitive periods of less than one year;
- $a_t$ = random component considered a white noise.

The Box and Jenkins’ model shapes the random component and if it is not considered a white noise in the Box and Pierce test (Priestley, 1989), an SARIMA (Seasonal Auto Regressive Integrated Moving Average: $p$, $d$, $q$) must be adjusted, where $p$ is the parameter of self-correlation, $d$ the parameter of the difference, and $q$ the parameter of moving averages. For the adjustment of the model the parameters were defined using correlograms.

Between January 2002 and December 2006, 3,202 direct fluorescent antibody tests (FAT) were performed for diagnosis of bovine rabies in the state of Minas Gerais and can be seen in Table 1.

Table 1 – Number of positive rabies diagnosis by direct fluorescent antibody (FAT) and mouse inoculation test (MIT), in cattle, in the state of Minas Gerais between 2002 and 2006.

<table>
<thead>
<tr>
<th>Year</th>
<th>Tested</th>
<th>FAT</th>
<th>MIT</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>2002</td>
<td>697</td>
<td>305</td>
<td>13</td>
<td>308</td>
</tr>
<tr>
<td>2003</td>
<td>710</td>
<td>261</td>
<td>12</td>
<td>273</td>
</tr>
<tr>
<td>2004</td>
<td>620</td>
<td>245</td>
<td>16</td>
<td>261</td>
</tr>
<tr>
<td>2005</td>
<td>617</td>
<td>269</td>
<td>6</td>
<td>275</td>
</tr>
<tr>
<td>2006</td>
<td>558</td>
<td>213</td>
<td>4</td>
<td>217</td>
</tr>
<tr>
<td>Total</td>
<td>3,202</td>
<td>1,293</td>
<td>51</td>
<td>1,344</td>
</tr>
</tbody>
</table>

SARIMA $(p, d, q) (P, D, Q)_{12}$ was chosen for the adjustment of the model as it shows the significance of the seasonal effect parameter. This effect was observed even without seasonality because there was significance in lag 12 of the self-correlation function. The process was used for an additive model.

The number of positive diagnoses of bovine rabies decreases between 2002 and 2006 ($p = 0.0339$), except in 2005 when an increase in the number of cases was observed. This is opposed to the situation observed between 1976 and 1997 in Minas Gerais (Silva et al., 2001) and between 1970 and 2001 in Goiás (Jayme, 2003), when the number of cases increased progressively and is coincident with the marked reduction in the number of cases of bovine rabies in Brazil (Arellano-Sota, 1988) and in the State of São Paulo (Taddei et al., 1991) due to the control measures implemented. However, the temporal periods of analysis and the human and financial resources allocated according to the budget of each state must be considered (Silva et al., 2001).

In the present study no seasonality was observed, as opposed to reports from Minas Gerais (Luz, 1988; Silva et al., 2001) and in São Paulo (Taddei et al., 1991), where the number of positive diagnosis increased between April and August and decreased between September and March.

In 2002, only 13 of 392 negative samples by FAT were positive by MIT (6.77%). In 2003, 12 of 449 negative samples by FAT were positive by MIT (2.67%). However, in 2004 this rate increased to 4.27% of positive cases by MIT (16/375), followed by a drop to 1.72% in 2005 (6/348) and reaching 1.16% in 2006 (4/345). This variation may be related to the efficiency of FAT which depends on factors such as: state of the sample when it is received at the lab, quality of the conjugate, equipment, training of technicians, and others.

The adjustment of the model SARIMA $(0,1,1)$ $(0,0,1)_{12}$ of Box and Jenkins allows checking for residues of white noise. With this model, a seasonal variation and a trend were evidenced. A forecast for the last 12 months was made and is shown in Figure 1, together with the confidence interval of 90%.

The seasonality was not confirmed by Fisher’s test (5%) but the trend was significant (5%). For the analyzed data, the adjustment of the SARIMA model was applied using two estimated parameters and one discrepancy difference to eliminate the trend. The parameters were $q = 0.84702 (0.10765)$ and $Q = -0.4863 (0.15308)$. The trend could be demonstrated using a linear model $y = 26.75 - 0.1427t$, with standard error 1.47 and 0.04199, respectively. Thus, a decreasing trend was characterized and in every month there was a reduction of 0.14 positive cases per month. The model adjusts for a 12 period seasonal effect. By using this model, a forecast for one year’s period could be made, with the respective confidence limits of 90% shown in Figure 1.

There was a decrease in the number of cases of rabies in cattle between 2002 and 2006, with an average of 0.14 cases per month. The time series model characterizes a 12 period seasonal effect, which means that there was a self-correlation effect with a delay of 12 months.
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