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Secretariat for Agriculture and Livestock Defence - SDA  
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World Health Organisation- WHO  
Pan-American Health Organisation- PAHO  
Pan-American Centre of Foot and Mouth Disease - PANAFTOSA



***Assessment of the population immunity deriving from the  
vaccination campaigns against foot and mouth disease***

***Final Report***

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## Summary

This report deals with the results of the investigation conducted in the zone free from foot and mouth disease with vaccination with the purpose of assessing the rates of vaccine coverage achieved by the veterinary service, mostly based upon the recording of the vaccination presented by cattle breeders. The activity was aimed at indirectly certifying the rate of vaccine coverage starting from the estimate of the prevailing level of immune protection for the viral strains present in the vaccine against foot and mouth disease used in the country. The estimate of protection against the foot and mouth disease virus of this animal population also allowed for assessing the strategies of vaccination used and the efficiency of the controls of the vaccination campaigns against the disease.

The region covered by the study encompassed the States of Acre (plus two municipalities of Amazonas), Bahia, Espírito Santo, Goiás, Mato Grosso, Mato Grosso do Sul, Minas Gerais, Paraná, Rio de Janeiro, Rio Grande do Sul, Rondônia, São Paulo, Sergipe, Tocantins and the Federal District. The bovine population in this region was separated per unit of the Federation according to age groups and to the strategies of vaccination practised in the respective territories. Thus, the 16 units of the Federation involved were organised in 18 independent sub-populations, according to the vaccination schemes used. For each sub-population an independent sample study was carried out.

The investigations were conducted by the Animal Health Department (DSA) related to the Secretariat for Agriculture and Livestock Defence of the Brazilian Ministry of Agriculture (MAPA) and by official veterinary services in the states involved, counting on the support of the Pan-American Centre of Foot and Mouth Disease (PANAFTOSA - PAHO/WHO), particularly at the stages of planning and interpretation of the results.

The first chapters of the report describe the different schemes of vaccination against foot and mouth disease implemented in the country, including information on the type of vaccine and on the control of the process of production and commercialisation of the vaccine, as well as on the rates of vaccination coverage for bovines achieved in the stages of vaccination carried out in the period from 2003 to 2005 in each unit of the Federation involved in the study.

The target population was characterised in domains, as to the size of the herds, and in sub-populations as to the age group of the animals. Regarding the size of the herd, three categories were considered: herds with up to 20 bovines; herds with 21 to 50 bovines and herds with more than 50 bovines. Regarding the age group, the study considered the sub-populations made up of bovines between 6 and 12 months, bovines between 13 and 24 months and bovines with more than 24 months.

The analytical method used for assessing the immunity response (protected or not protected) of each individual was the essay of competition enzymatic immune-absorption at the fluid stage (ELISA-CFL) standardised by PANAFTOSA for the detection of specific antibodies against proteins of the viral capsid. All laboratory essays were carried out at the LANAGRO of Pedro Leopoldo, MG, according to the manuals and inputs produced by PANAFTOSA. Each sub-population was tested for one of the three types of virus contained in the Brazilian vaccine (A, O and C).

Field activities were conducted in the period of July, 2005 to February, 2006, involving the collection of 20.423 samples distributed in 1.956 rural properties. Out of these samples, 1.898 (9%) were obtained in herds with up to 20 bovines; 2.477 (12%) in herds with between 21 and 50 bovines; and 16.048 (79%) in herds with more than 50 bovines. Regarding age groups, 8.565 samples were obtained (42%) from bovines between the ages of 6 and 12 months, 7.017 (34%), of bovines between the ages of 13 and 24 months, and 4.841 (24%), of bovines with more than 24 months of age.

As to the results, the analysis per sub-population showed, for nearly all the units of the Federation, excellent levels of immune coverage for the bovine population, in any of the age groups considered. The values obtained surpassed by far the initial expectations of the study, with the only exception of the East Circuit of Minas Gerais, where, clearly, an immune coverage comparatively smaller than those of the other sub-populations assessed was recorded.

Taking into account the fact that bovines with more than 12 months of age represent about 80% of the existing population in most of the sub-populations assessed, the immunisation levels recorded in animals of 13 to 24 months or more than 24 months appear compatible with the rates of vaccination coverage assessed based upon the declaration of the vaccination, and also reinforce the high level of immune coverage that exists in the bovine population of the zone free from foot and mouth disease with vaccination.

Considering all the age groups together, the lowest estimations of the prevalence of bovines protected for the virus "A", "O" and "C", with 95% of confidence, were of 87%; 98% and 98% respectively, with the exclusion of the sub-population of the East Circuit of Minas Gerais where the lowest estimate was of 68% of bovines protected for the virus "A".

As it was expected, the age group where the lowest prevalence of immunised bovines was recorded was represented by animals between 6 and 12 months of age. In spite of the expectation of 65% of protection for this group, in more than half the sub-populations assessed (72%) there was true prevalence higher than 85%, and in eight (44%), the true prevalence was higher than 95%. In the sub-populations with the lowest rates of immunity for population of bovines with ages between 6 and 12 months, only in the East Circuit of Minas Gerais the higher limit of the interval of confidence was lower than the estimated value of 65%.

In spite of the variations recorded among the types of virus assessed, the study identified a trend towards more stability, with higher levels of immunity, for the category of herds with more than 50 bovines. On the other hand, for the other categories related to the size of the herd, particularly when there was the assessment of bovines with ages between 6 and 12 months, the lowest prevalence of protection was recorded. These results coincide with the forecasts of the study, considering that the owners of herds with more than 50 bovines are more interested and ready to carry out the vaccination against foot and mouth disease, although the costs involved are higher.

Bovines not born in the properties featured vaccine coverage higher than that obtained for native animals. This phenomenon can be explained bearing in mind the fact that the animals only receive authorisation for movement if they have been vaccinated, and they are even submitted to supplementary vaccination, thus minimising the risk represented by the trade on susceptible animals.

Only the East Livestock Circuit of Minas Gerais showed a level of immune coverage lower than 80%. Considering the association recorded between immunisation levels and rates of vaccination coverage, the level of protection achieved, circa 71%, is the lowest among the sub-populations studied, deviating from the rate of vaccination coverage assessed in the region for the stage prior to the collection of the samples, of approximately 96%. This might be explained, among other reasons, by problems in the preparation of the indicators of completion of the stages of vaccination. On the other hand, this lesser immunisation coverage, when associated to the absence of records of clinical disease and to the results of the studies of viral circulation, reinforces the hypothesis of non-existence of residual virus, since there would be about 30% of susceptible bovines in this region.

Irrespective of all the activities and procedures involved in the preparation of the results of the stages of vaccination, overall the study showed the compatibility of these assessments with the immunisation levels of the population, estimated by means of the laboratory results obtained. Somehow, the levels of immune protection recorded reflect the country's tradition in terms of carrying out vaccination campaigns against foot and mouth disease, which for over three decades has been one of the main strategies of the PNEFA, and the good quality of the vaccine used, particularly starting from the 1990s.

The results obtained are consistent with the results of the assessment studies on viral circulation carried out as part of the epidemiological assessments aimed at obtaining the international recognition of the sanitary condition "free with vaccination". The conclusion of the study is that the levels of population immunity achieved were sufficient to break the epidemiological chain of circulation of the virus and to achieve the condition of "free".

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## 1. Introduction

This report presents and discusses the results of the work carried out in the zone free from foot and mouth disease with vaccination for assessing the rate of vaccine coverage of the bovine population in the units of the Brazilian Federation.

The work allowed for an estimation of the level of protection, for the viral strains present in the vaccine against foot and mouth disease used in the country, of the population of bovines of each unit of the Federation of the zone free from foot and mouth disease with vaccination, according to age groups defined and to the strategy of vaccination practised. The work was carried out by the Animal Health Department (DSA) of the Secretariat for Agriculture and Livestock Defence /MAPA, and by the bodies in charge of animal sanitary defence in the units of the Federation involved, with the support of the Pan-American Centre of Foot and Mouth Disease (PANAFTOSA-PAHO-WHO).

The systematic and mandatory vaccination against foot and mouth disease is being used in most of South America as the central tool of the national programmes of eradication of the disease, and it has been officially adopted in Brazil since the 1960s. Appropriately planned, implemented and assessed vaccination campaigns, which used vaccines with proven quality and power and that achieve high immunity-coverage levels, are able to drastically reduce the susceptibility of the population to the virus, thus reducing the risk of clinical presentation of the disease and interfering in the infection process by means of the inhibition or of the reduction of viral multiplication in exposed animals. Thus, there is a progressive and sustained reduction of the replication of the virus, both by drastically reducing the number of susceptible animals in the population and by critically reducing the viral supply, thus causing its eradication. These elements represent the conceptual basis that supports the achievement of the recognition of the sanitary condition of free from foot and mouth disease with vaccination in susceptible territories and populations submitted to systematic vaccination.

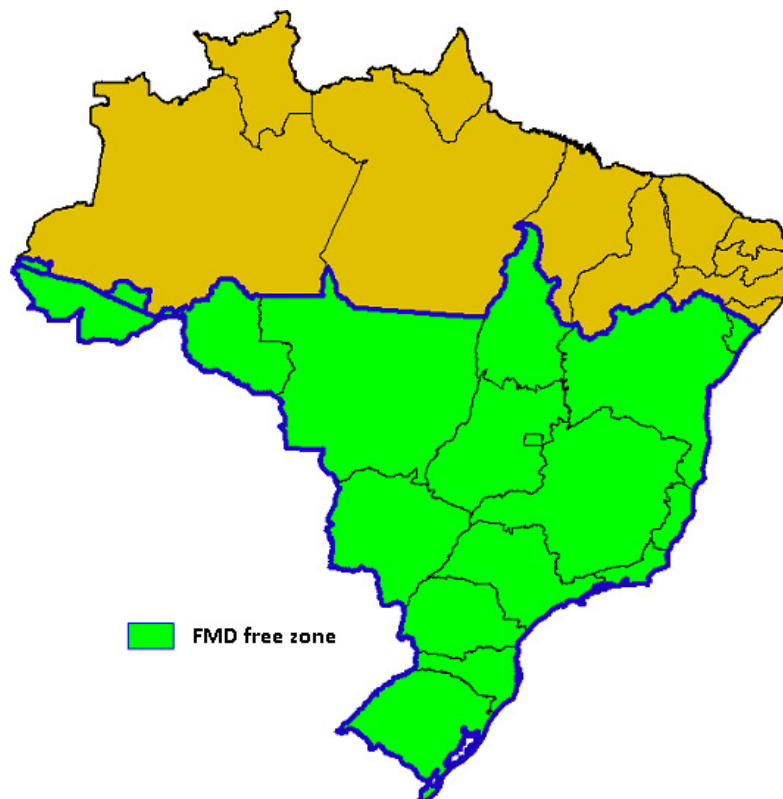
Vaccination campaigns in the country are assessed by the bodies in charge of animal sanitary defence, particularly considering the declaration of vaccination presented by rural producers in charge of its implementation, contrasted with the registry of animal-husbandry establishments available at the local veterinary units of the official veterinary service. Therefore, it depends on the effective participation of rural producers and on the quality of the registry of the official veterinary service. The results obtained based upon this control show rates of vaccination coverage higher than 90% in most of the units of the Federation involved, attaining in an important part of the locations, practically 100% of the existing bovine population. Thus, the work carried out was aimed at checking the level of immune coverage according to the strategies of vaccination against foot and mouth disease used in the free zone, as well as the efficiency of the controls and methods of assessment of the implementation of vaccination campaigns in place in the country. It represented an opportunity for assessing to what extent the rates of recording of vaccination indirectly reflect the levels of immune protection of the population of bovines in the free zone. The conduction of this work tried to additionally clarify part of the questions made by importer markets of Brazilian beef.

This report also includes a brief description of the different schemes of vaccination against foot and mouth disease implemented in the country, including information on the type of vaccine, control of the process of production and commercialisation, as well as the rates of coverage of bovines vaccinated obtained in the stages of vaccination carried out in the period from 2003 to 2005 in each unit of the Federation involved in the study. This information will be important for putting into context and discussing the findings of the study.

## 2. Information on the vaccination against foot and mouth disease in the country

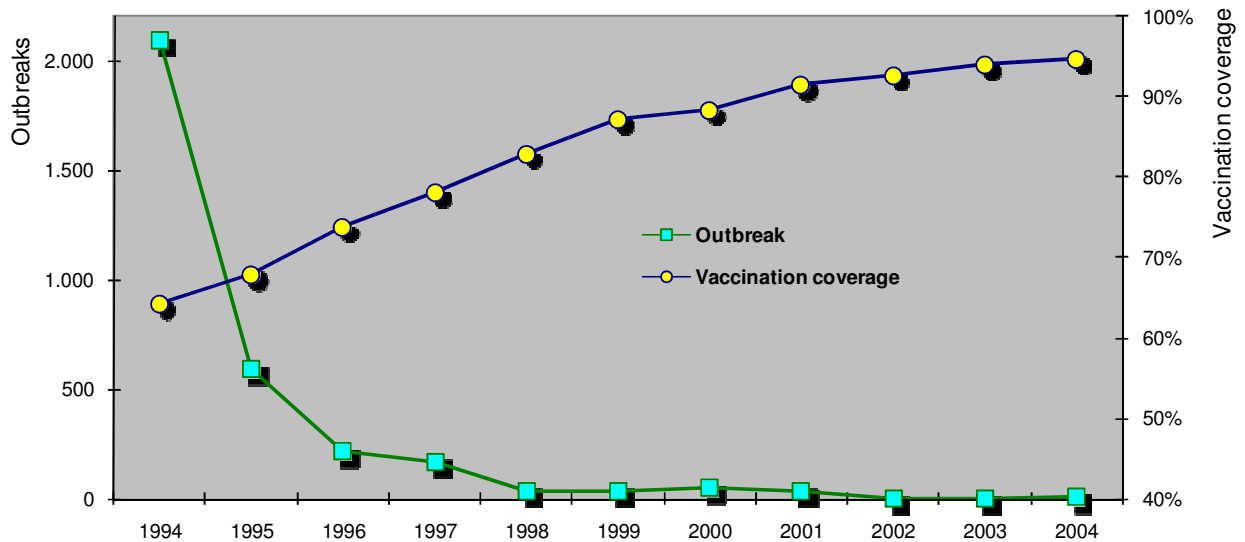
The fundamentals of the PNEFA are represented by the systematic and mandatory vaccination of the bovine and bubaline population; by the control of the movement of animals; and by the activities of epidemiological surveillance, including actions of prevention and plans of intervention against zoo-sanitary emergencies. These fundamentals are supported by the sharing of responsibilities between the public and the private sectors. In what regards vaccination, the responsibility befalls the private sector, represented by the owners of animals, and the acquisition and the inoculation of the vaccine against foot and mouth disease befall the public sector, by means of the official veterinary service, as well as guaranteeing the quality of the vaccine produced and the control, guidance and assessing of the activities of commercialisation and use of the product. As it was mentioned at the beginning, the implementation and the control of vaccination campaigns within the units of the Federation befall the state organisations of animal sanitary defence, according to general standards and procedures agreed upon with the DSA. At the option of the state organisations of animal sanitary defence, vaccination in areas of risk or in regions of small producers may be assisted or even performed by the official veterinary service.

Official vaccination campaigns began in the early 1960s. Currently they are suspended solely in the State of Santa Catarina, and they are carried out in a systematic and mandatory manner in the other units of the Federation. Vaccination associated to other sanitary activities has allowed for important progress in the fight against foot and mouth disease, and in the period from 1998 to September, 2005, the country achieved international recognition as a zone free from foot and mouth disease with vaccination for 51% of the territory, which contained 84% of the existing bovine population (Picture 1). In global terms, the occurrence of the disease diminished from over 2000 outbreaks in 1994 to 5 outbreaks in 2004, and there are wide areas of the country where the disease has not been recorded for over 10 years. The evolution of the vaccination coverage and of the annual distribution of outbreaks of foot and mouth disease in the country, for the period of 1994 to 2004, may be assessed by means of Picture 2.



**Picture 1.** Zone free from foot and mouth disease with vaccination, recognised by the OIE until September, 2005





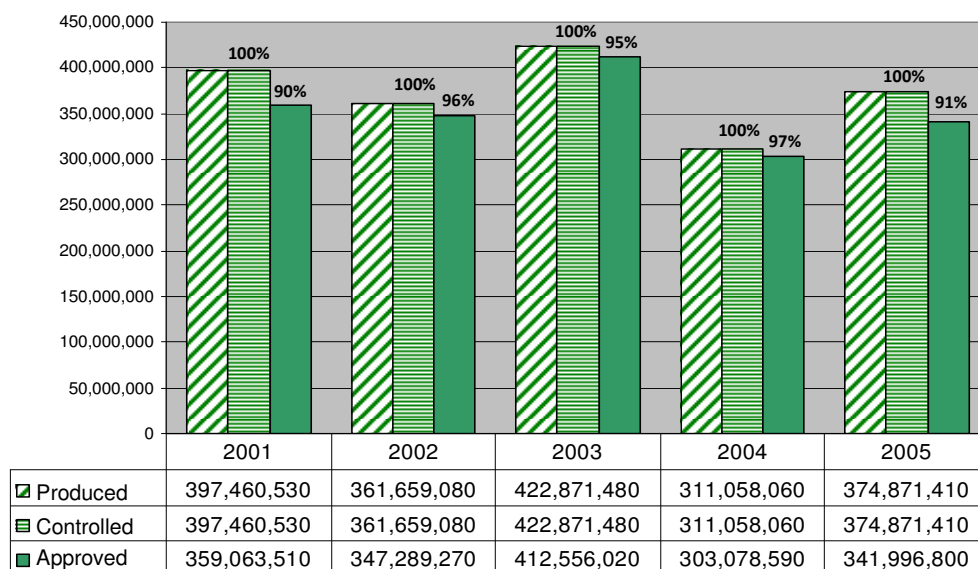
Picture 2. Number of outbreaks of foot and mouth disease and vaccination coverage, Brazil, 1994 to 2004

### 2.1. Production of the vaccine

The vaccine used in the country is trivalent and contains oily adjuvant. It is made with the strains O1 Campos, A24 Cruzeiro and C3 Indaial. This biological composition emerged starting from studies developed by PANAFTOSA in cooperation with the Centre of Animal Diseases of Plum Island, of the Department of Agriculture of the United States, which began in 1968, involving countries of South America, highlighting Brazil, where important field experiments were carried out, counting on the participation of the MAPA and of the veterinary services of the units of the Federation involved. The commercial development of the product began in the late 1970s, and the Brazilian Government put in place laboratories for the production of oily vaccine in Campinas and Porto Alegre starting from 1984, and later on PANAFTOSA transferred the technology of production to private industries. The use in Brazil was introduced gradually, and it began chiefly in regions where foot and mouth disease was endemic. Starting from 1992, the industries in place in the country only produce vaccines with oily adjuvant. As a consequence of stocks in existence on the market, in the subsequent years there has still been a reduced use of vaccines with aqueous adjuvant, which situation was maintained until 1994, with small differences among the units of the Federation.

The production of the vaccine with oily adjuvant complies with regulations established by the MAPA and recommendations of the World Animal Health Organisation (OIE). Every vaccine against foot and mouth disease used in Brazil is produced by six private laboratories, with P3+ bio-security level, located on the national territory, which, in addition to meeting the domestic demand, export vaccines for other countries of South America. This industrial complex has installed capacity for the production of over 500 million doses/year. The production of antigens is made by means of cell culture in suspension, with the use of different methods of concentration (ultra-filtration, PEG etc). Each lot of vaccine must contain at least 500.000 doses, and all lots are officially controlled and submitted to quality tests in MAPA official laboratories.

After the performance of internal quality-control tests in the industry, the vaccine is immediately packed. Out of the total lot of phials, a random sample of phials is taken by employees of the official veterinary service and the official quality tests of the lot are carried out in the laboratories of the MAPA. These tests concern the innocuity, sterility, physical-chemical condition and power of the vaccine. The power test is carried out by the indirect relative method (by means of serum essays), using 18 bovines (two testimonies and 16 vaccinated). The collection of serum samples from the animals is carried out 28 days after the vaccination and the ELISA-CFL essay of competition enzymatic immune-absorption at the fluid stage), standardised by PANAFTOSA, is used to measure the level of specific antibodies against proteins of the viral capsid. These results are then turned into percentage expectations of protection (EPP), applying a decision rule to judge the quality of the vaccine as compared to its power. The vaccine is deemed approved when it features an EPP of 80% for each of the three strains that make it up, with a level of confidence of 95%. The lot of vaccines having gone through all the other tests schedule is approved and released for commercialisation. Otherwise, the entire lot is destroyed, under the supervision of the official veterinary service. In Picture 3 it is possible to assess the total of vaccines produced and approved in the country in the period from 2001 to 2005.



**Picture 3.** Vaccine against foot and mouth disease produced by the industry and controlled and approved by the MAPA in the period from 2001 to 2005

## 2.2. Distribution of and trade on the vaccine

The distribution of the vaccine, from the industry to the dealers of veterinary products authorised by the official veterinary service to commercialise the product, is carried out by means of a central controlled by the six producing industries, which provides, in up to 48 hours, the doses required in each municipality. This central is located in the Municipality of Vinhedo (SP) and counts on appropriate logistics for storage, distribution and transport, being also responsible for the stamping of the quality seal with guarantees of inviolability, after the official approval of the lot. A computerised system allows the official veterinary service to obtain at any moment the data concerning stock, release and commercialisation of the product in all units of the Federation. The logistics of the distribution central facilitates the control of the supply and of the distribution of the product and allows for a total condition of supervision and inspection. It preserves the concept of traceability, inhibits forgery, avoids excessive manipulation of the product and minimises the possibility of occurrence of problems that jeopardise the conservation and the refrigeration of the vaccine until it reaches the sales points.

All sales points of products for veterinary use have to be registered and licensed by the official veterinary service as a condition for their operation. For the commercialisation of the vaccine against foot and mouth disease, the dealers must meet specific conditions and they are submitted a frequent controls, especially during the stages of vaccination. The procedures for control of the trade on the vaccine against foot and mouth disease are organised in the document **“Guidance for inspection of the trade on vaccines against foot and mouth disease and for the control and assessment of the stages of vaccination”**, prepared by the DSA and made available to all state organisations of animal sanitary defence. Below we highlight some points contained in said document:

- a) The authorisation for commercialisation of vaccine against foot and mouth disease is only issued by means of a technical opinion by a veterinarian of the official service certifying the conditions necessary for the conservation of the product. Special attention is paid to the alternatives used by the sales point for the conservation of the vaccine in the event of power cuts (production of ice by the sales point or by another establishment in the municipality, power generator, among others);
- b) Commercial establishments are obliged to provide, for each refrigerator, a thermometer with the records maximum and minimum temperatures, identified as being for the sole use of the official veterinary service ;
- c) The refrigerator used for the conservation of the vaccine against foot and mouth disease can only be used for this purpose;
- d) All vaccines against foot and mouth disease, in order to enter the sales point, must be inspected by the official veterinary service, which must check the sealing in all phials, the condition of conservation, the origin, the number of the lot, the validity and the number of doses;
- e) During the stages of vaccination against foot and mouth disease, the inspection of commercial establishments is intensified, with at least two inspections/establishment/week. During this period, the checking of the temperature of the refrigerators used for the conservation of the vaccines is daily, with readings in the morning and in the afternoon. Out of the stages of vaccination a minimum frequency of at least one visit a week is maintained;
- f) In the beginning and at the end of the stages of vaccination, the stocks of vaccines in authorised sales points are mandatorily checked;
- g) All activities of inspection are recorded in appropriate forms;
- h) Vaccines can only be commercialised during the official stages or with authorisation issued by the official veterinary service ;
- i) All vaccines against foot and mouth disease must be commercialised in appropriate containers capable of maintaining the ideal temperature of conservation (with 2/3 of ice), with the issuance of invoices and the respective entry in the control of the stock. After the removal of a vaccine against foot and mouth disease from the refrigerator and after the removal is recorded in the control of stock, that vaccine may not return to the sales point, and producers or any other persons are not allowed to keep the vaccine in the refrigerator of the sales point for later use;
- j) It befalls the state organisations of animal sanitary defence to maintain updated the stock of vaccine against foot and mouth disease available in authorised sales points.

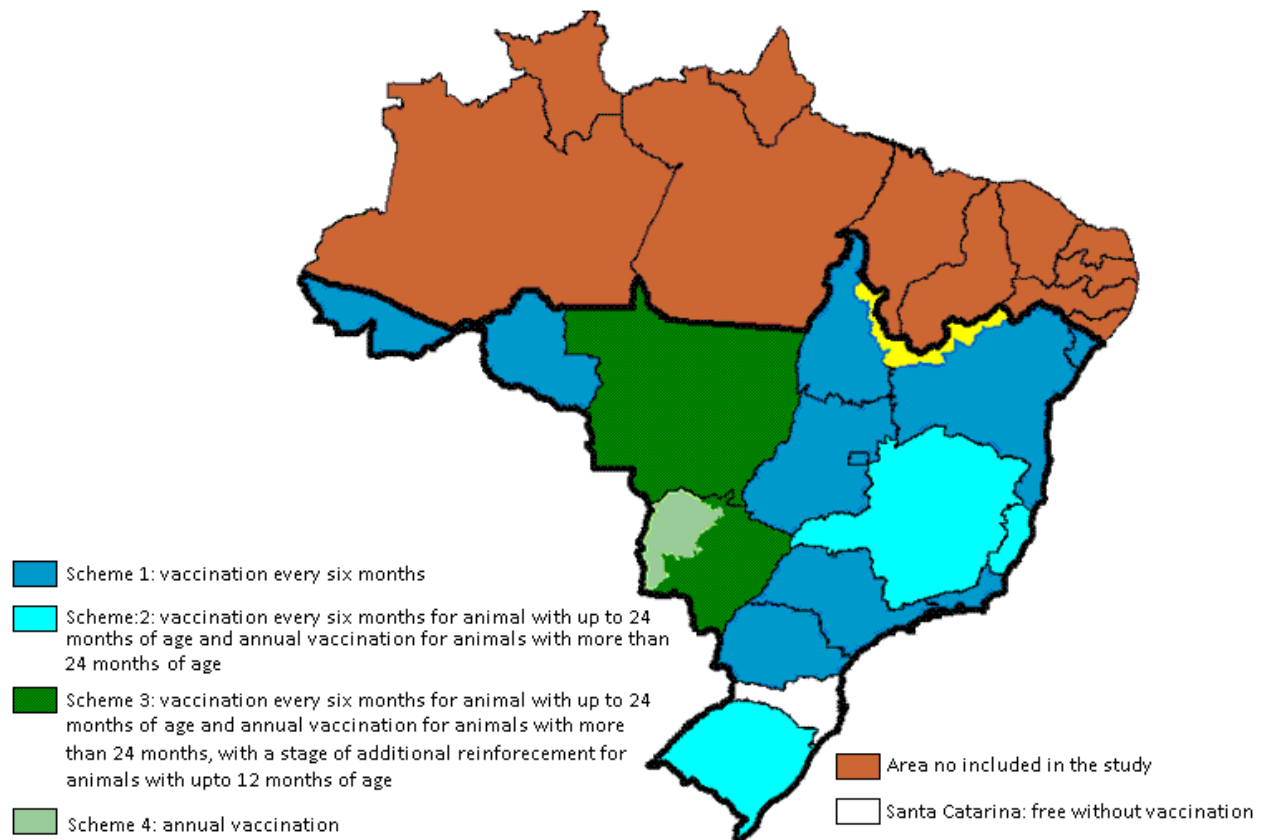
### 2.3. Schemes of vaccination

Vaccination is mandatory for bovines and bubalines, irrespective of the age of the animals, through the use of schemes adapted to the geographical and agro-productive realities predominant in each region of the country. These schemes may be summarised in four different types:

- Scheme 1: vaccination every six months of the entire bovine and bubaline herd in 30 days, adopted in most of the units of the Federation ;
- Scheme 2: vaccination every six months of bovines and bubalines with up to 24 months of age and annual vaccination for animals with more than 24 months of age, carried out in stages of 30 days;
- Scheme 3: vaccination every six months of bovines and bubalines with up to 24 months of age and annual vaccination for animals with more than 24 months of age, with a stage of additional reinforcement for animals with up to 12 months of age, in stages of 30 days; and
- Scheme 4: annual vaccination of all bovines and bubalines, in stages that vary from 45 to 60 days, carried out in regions where the geographical characteristics only allow for moving the animals during a limited period of the year.

The organisation of the units of the Federation of the zone free from foot and mouth disease with vaccination, according to the scheme of vaccination used, may be assessed by means of Picture 4.

It must be highlighted that vaccination against foot and mouth disease in the State of Santa Catarina was suspended in May, 2000. The interdiction involves both the inoculation of the vaccine and its commercialisation in the entire territory of that State. In May, 2007, the State was recognised by the OIE as a zone free from foot and mouth disease without vaccination.



**Picture 4.** Schemes of vaccination against foot and mouth disease used in the free zone

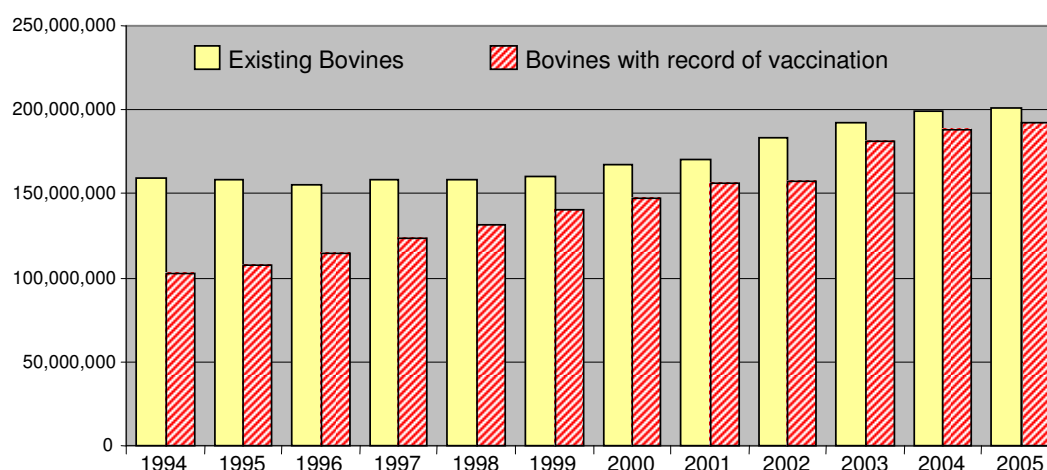
The months for carrying out the stages of vaccination vary according to each unit of the Federation, particularly considering the predominant climate conditions, the seasons of concentration of the birth of calves, the intensity and the seasonal character of the movement or of the commercialisation of animals. The standards of control establish the mandatory character of vaccination during the stages defined, and any vaccination out of the official calendar can only be carried out with the authorisation of the official veterinary service.

After each stage, the owner of the animals must record the vaccination stage with the local veterinary units, within the deadlines established by law. After his deadline, the official veterinary service must identify, in the registry, the producers in default, which are subjected to fines and prevented from moving the animals, and in these cases the herd may be vaccinated with the follow-up and the inspection of the official veterinary service. The procedures for the control of the stages of vaccination are in the Guide prepared by the DSA and mentioned in the previous item, involving specific activities to be conducted before, during and after each stage.

The historical series for the period from 1994 to 2005, concerning the recording of the vaccination against foot and mouth disease all over the country is presented in Table 1 and in Picture 5. Specifically for the units of the Federation of the zone free from foot and mouth disease with vaccination, in Table 2 we present the results for each stage of vaccination carried out between 2003 and 2005. In general there was an increment of the practice of vaccination in the period under analysis, with the maintenance of rates higher than 80% starting from 1998. In the specific case of the units of the Federation recognised as zone free from foot and mouth disease with vaccination, the results recorded indicate the consolidation of this practice in the region. In 2003, out of the 34 stages carried out, 21 (62%) featured rates of recording of vaccination equal to or higher than 95%, 9 (26%) showed values between 90 and 94%, and 4 (12%), values between 82 and 89%. In 2005, 24 (71%) stages featured results equal to or higher than 95%; 9 (26%), results between 90 and 94% and only 1 (3%) featured the rate of 89%, represented by the stage of March in the State of Rio de Janeiro. To check the compatibility of these rates of recording of the vaccination against foot and mouth disease with the levels of immune protection of the bovine population is one of the main purposes of this study.

**Table 1.** Bovine and bubaline population vaccinated against foot and mouth disease according to the declaration of the producer, Brazil, 1994 to 2005

Year	Vaccination campaigns against foot and mouth disease			
	Population of bovines and bubalines			Doses Inoculated
	Existing	Vaccinated	% Vaccinated	
1994	159.227.797	102.326.522	64%	198.816.883
1995	158.503.190	107.543.498	68%	207.733.516
1996	155.368.527	114.731.921	74%	218.312.698
1997	158.446.481	123.911.138	78%	228.809.106
1998	158.009.814	131.200.698	83%	243.562.873
1999	160.395.129	139.950.430	87%	236.903.765
2000	166.974.605	147.718.162	88%	232.017.381
2001	170.625.996	156.101.114	91%	277.505.686
2002	183.668.123	157.639.726	86%	292.629.840
2003	192.246.837	180.948.940	94%	313.502.481
2004	198.941.557	188.653.738	95%	332.788.563
2005	201.246.878	192.659.465	96%	343.289.451



Picture 5. Existing bovine population and with records of vaccination, Brazil, period from 1994 to 2005

Table 2. Rates of recording of the vaccination against foot and mouth disease, according to stage of vaccination in the units of the Federation recognised as zones free from foot and mouth disease until September, 2005.

UF	Month	Herd involved	2003			2004			2005		
			Existing	Vaccinated	%	Existing	Vaccinated	%	Existing	Vaccinated	%
AC	May	All herd	1,857,989	1,563,358	84.14	1,844,164	1,688,316	91.55	2,087,015	1,919,414	91.97
	Nov	All herd	1,764,051	1,590,809	90.18	1,984,975	1,878,160	94.62	2,298,511	2,177,691	94.74
BA	Mar	All herd	9,418,842	8,782,046	93.24	9,801,320	9,056,588	92.40	9,850,254	9,409,476	95.53
	Sep	All herd	9,705,273	8,976,042	92.49	9,607,397	8,867,774	92.30	10,137,958	9,695,934	95.64
DF	May	All herd	102,002	98,493	96.56	123,215	114,498	92.93	106,341	98,576	92.70
	Nov	All herd	102,002	98,146	96.22	104,601	99,446	95.07	114,484	104,026	90.87
ES	Mar	< 24 months	674,624	673,926	99.90	720,240	697,187	96.80	690,836	666,226	96.44
	Sep	All herd	1,837,988	1,802,888	98.09	1,901,693	1,871,076	98.39	2,012,998	1,977,725	98.25
GO	May	All herd	20,196,578	19,888,039	98.47	20,090,613	19,562,049	97.37	20,045,632	19,740,057	98.48
	Nov	All herd	20,011,223	19,762,755	98.76	20,034,169	19,690,815	98.29	20,549,589	20,308,758	98.83
MT	Fev	< 12 months	4,847,717	4,207,451	86.79	5,303,092	4,490,901	84.68	5,551,458	5,196,328	93.60
	May	< 24 months	9,839,486	9,677,105	98.35	10,371,977	10,111,744	97.49	10,433,986	10,278,015	98.51
MS	Nov	All herd	24,715,876	24,337,705	98.47	26,004,415	25,685,465	98.77	26,844,149	26,695,439	99.45
	Fev	< 12 months (Plateau)	5,268,766	5,173,999	98.20	5,333,397	5,235,807	98.17	5,129,300	5,074,356	98.93
MG	May	< 24 months (Plateau) + part of the flooded plain	11,714,507	11,487,182	98.06	12,166,668	12,002,772	98.65	12,249,002	12,144,732	99.15
	Nov	All herd (Plateau) + part of the flooded plain	22,646,993	22,337,394	98.63	22,215,689	22,022,049	99.13	21,501,644	21,399,883	99.53
PR	Mar	All herd - East Livestock Circuit	8,475,438	8,103,251	95.61	9,024,259	8,600,717	95.31	9,387,577	9,080,959	96.73
	May	All herd - Centre-West Livestock Circuit	11,332,649	11,101,195	97.96	11,619,972	11,319,396	97.41	10,721,378	10,325,554	96.31
RJ	Sep	< 24 months - East Livestock Circuit	3,826,411	3,145,932	82.22	3,826,411	3,556,346	92.94	3,956,721	3,792,504	95.85
	Nov	< 24 months - Centre-West Livestock Circuit	4,990,837	4,806,176	96.30	5,011,127	4,710,459	94.00	5,222,123	5,098,357	97.63
RS	May	All herd	10,158,271	9,299,469	91.55	10,393,122	10,226,866	98.40	10,098,076	9,968,618	98.72
	Nov	All herd	10,406,809	10,278,876	98.77	10,240,260	10,093,344	98.57	10,251,971	10,004,306	97.58
RO	Mar	All herd	1,959,264	1,819,380	92.86	2,008,106	1,832,964	91.28	2,138,765	1,901,338	88.90
	Sep	All herd	1,957,722	1,826,786	93.31	2,045,424	1,846,384	90.27	1,939,903	1,787,241	92.13
SP	Jaf - Feb	All herd	14,040,019	12,916,817	92.00	14,040,019	12,964,678	92.34	13,342,351	12,368,357	92.70
	Jul - Aug	< 24 months	4,757,983	4,282,184	90.00	5,413,071	4,914,022	90.78	4,389,936	3,981,677	90.70
TO	May	All herd	8,847,872	8,846,043	99.98	9,824,171	9,820,708	99.96	10,751,368	10,748,117	99.97
	Nov	All herd	9,621,225	9,620,271	99.99	10,676,093	10,675,146	99.99	11,349,452	11,348,828	99.99
SE	May	All herd	14,208,583	14,123,264	99.40	14,245,824	14,166,047	99.44	13,650,423	13,569,420	99.41
	Nov	All herd	14,514,884	14,426,343	99.39	13,993,218	13,902,301	99.35	13,713,694	13,659,478	99.60
SC	May	All herd	822,367	745,281	90.63	846,374	764,698	90.35	861,859	802,252	93.08
	Nov	All herd	824,569	725,538	87.99	872,382	785,466	90.04	937,857	846,300	90.24
MS	May	All herd	7,330,961	7,135,550	97.33	7,740,483	7,557,613	97.64	7,760,299	7,639,634	98.45
	Nov	All herd	7,638,468	7,502,122	98.22	7,893,071	7,739,732	98.06	7,917,145	7,771,591	98.16

Source: state organisations of animal sanitary defence

### 3. Material and methods

#### 3.1. Geographical scope and populations under study

The study was conducted in the zone free from foot and mouth disease with vaccination made up of the States of Acre (plus two municipalities of the State of Amazonas), Bahia, Espírito Santo, Goiás, Mato Grosso, Mato Grosso do Sul, Minas Gerais, Paraná, Rio de Janeiro, Rio Grande do Sul, Rondônia, São Paulo, Sergipe and Tocantins, in addition to the Federal District (Picture 1, item 2).

In Santa Catarina, where vaccination is not practised, in 2006 an independent study was carried out with the purpose of verifying the absence of vaccinated bovines, within the project for its international recognition as a zone free from foot and mouth disease without vaccination. Information on the study and the results obtained may be found in Annex 1 to this document.

The 16 units of the Federation involved were organised in 18 independent sub-populations, according to the schemes of vaccination described in item 2.3 and represented in Picture 3. In general, each unit of the Federation made up a sub-population for the sampled survey, except for the States of Mato Grosso do Sul and Minas Gerais. In these states co-exist sub-regions with different schemes of vaccination, which were divided, respectively, in three and two sub-populations under sampling.

### **3.2. Distribution and characterisation of the target population**

For the outlining of the study, the state organisations of animal sanitary defence transmitted to the DSA their electronic databases, and a central base was created containing the total of existing bovines per age group, per rural property and per municipality, concerning 2005. This information was organised according to the independent sub-populations, considering the groupings of specific interest for the study. Thus, among the different variables related to the target population of the study, which may directly or indirectly interfere with the expectation of immune protection for foot and mouth disease, it was decided to use two of them: i) type of rural property according to the number of existing bovines; and ii) age group of the bovines. These variables were considered for the determination and the allocation of the sample.

Regarding the type of rural property, three categories were considered as to the size of the herd: herds with up to 20 bovines; with 21 to 50 bovines and with more than 50 bovines. These categories were established with a view at characterising the motivation or the capacity of the owners of the animals to vaccinate their entire herds and are related to the socioeconomic condition of rural producers who face difficulties in moving the animals for vaccination. The expectation is that herds with up to 20 bovines correspond to owners with worse socioeconomic condition and lesser costs involved in the vaccination of their animals. Herds having between 21 and 50 bovines would correspond to owners in an intermediate condition and herds of more than 50 bovines would correspond to owners with more interest, better socioeconomic condition and, possibly, higher stewardship costs for the practice of vaccination. Table 3 presents the distribution of the number of properties with bovines in the geographical area under study, according to the units of the Federation and to the number of bovines in the herds. In global terms, one notices the existence of 1.7 million rural properties, 51% belonging to the category of herds with up to 20 bovines, 22% to the category of 21 to 50 bovines and 27% to the category with more than 50 bovines, with an important variation among the sub-populations defined.

Taking into account the fact that the expectation of protection is directly related to the number of vaccinations carried out in the animals, the following age groups were established for classifying the bovine population of the area under study: bovines of 6 to 12 months, of 13 to 24 months and older than 24 months. This division per age groups allowed for reducing the population variation by considering the different levels of protection expected for each group.

Tables 4 and 5 present information on the total of bovines per age group, obtained from the state organisations of animal sanitary defence. Regarding the age group between 6 and 12 months, the information is presented as bovines of up to 12 months, as a function of the availability of this information in the registry of the local veterinary units. However, the sample collection only involved animals with ages above 6 months, trying to mitigate possible interferences of passive immunity induced by colostrum.

In Table 4, the total of bovines per age group is presented per sub-population, and one notices a small variation vis-à-vis the global values of 22% of bovines with up to 12 months of age, 21% with 13 to 24 months and 57% with ages above 24 months.

In Table 5, the total of bovines is also grouped according to the categories of rural properties considered in the study, and one notices a significant variation in their distribution among the sub-populations. Small properties are mostly concentrated in Bahia, the Federal District, Paraná, Rio Grande do Sul and Sergipe, with percentages varying from 10.7% to 17.7% vis-à-vis the total of properties with bovines of each sub-population. Goiás, Mato Grosso, Mato Grosso do Sul, Rondônia and Tocantins record the smallest percentages of rural properties with up to 20 bovines, with values below of 3% vis-à-vis the total of bovines of the sub-population.

**Table 3.** Existing total of properties with bovines, according to the size of herds considered, 2005

UF and regions (sub-populations)	Existing total of properties with bovines according to the size of the herds						Total
	Up to 20 bovines		21 to 50 bovines		More than 50 bovines		
Acre and two municipalities of the State of Amazonas	7.506	39%	4.889	25%	6.990	36%	19.385
Bahia	147.917	66%	45.139	20%	30.279	14%	223.335
Federal District	1.832	64%	577	20%	447	16%	2.856
Espírito Santo	10.854	44%	6.733	27%	7.221	29%	24.808
Goiás	22.082	21%	27.709	26%	57.128	53%	106.919
Mato Grosso	28.580	25%	26.303	23%	59.636	52%	114.519
Mato Grosso do Sul (Pantanal, May)	211	13%	185	11%	1.256	76%	1.652
Mato Grosso do Sul (Pantanal, November)	427	28%	71	5%	1.003	67%	1.501
Mato Grosso do Sul (Plateau)	6.473	14%	8.572	18%	31.542	68%	46.587
Minas Gerais (Centre-West Circuit)	95.450	50%	46.359	24%	50.703	26%	192.512
Minas Gerais (East Circuit)	111.928	58%	42.457	22%	39.127	20%	193.512
Paraná	140.028	65%	40.061	19%	33.737	16%	213.826
Rio de Janeiro	44.419	74%	7.960	13%	7.641	13%	60.020
Rio Grande do Sul	151.205	69%	42.041	19%	27.186	12%	220.432
Rondônia	20.970	27%	20.765	26%	37.367	47%	79.102
São Paulo	58.960	39%	40.611	27%	51.841	34%	151.412
Sergipe	15.635	65%	4.781	20%	3.812	16%	24.228
Tocantins	12.461	25%	13.499	27%	24.121	48%	50.081
<b>Total</b>	<b>876.938</b>	<b>51%</b>	<b>378.712</b>	<b>22%</b>	<b>471.037</b>	<b>27%</b>	<b>1.726.687</b>

Source: state organisations of animal sanitary defence

**Table 4.** Existing bovine population, according to age groups considered, 2005

UF and regions (sub-populations)	Bovines by age group						Total of bovines
	< 12 months		13 to 24 months		> 24 months		
Acre and two municipalities of the State of Amazonas	516.345	22%	459.398	20%	1.354.325	58%	2.330.068
Bahia	1.709.627	20%	1.947.514	22%	5.014.785	58%	8.671.926
Federal District	23.319	22%	21.125	20%	61.837	58%	106.281
Espírito Santo	250.883	15%	292.823	17%	1.166.427	68%	1.710.133
Goiás	4.034.678	22%	4.210.716	23%	10.068.226	55%	18.313.620
Mato Grosso	6.247.263	23%	5.841.451	21%	15.637.524	56%	27.726.238
Mato Grosso do Sul (Pantanal, May)	487.023	22%	324.587	15%	1.362.379	63%	2.173.989
Mato Grosso do Sul (Pantanal, November)	241.694	21%	201.692	17%	720.402	62%	1.163.788
Mato Grosso do Sul (Plateau)	4.350.722	22%	4.052.897	21%	11.352.198	57%	19.755.817
Minas Gerais (Centre-West Circuit)	2.376.031	21%	2.418.876	22%	6.418.198	57%	11.213.105
Minas Gerais (East Circuit)	1.899.158	21%	1.962.355	21%	5.337.336	58%	9.198.849
Paraná	2.062.806	22%	2.322.256	25%	5.093.201	54%	9.478.263
Rio de Janeiro	412.217	23%	325.748	18%	1.063.124	59%	1.801.089
Rio Grande do Sul	1.755.487	18%	1.863.181	19%	6.318.666	64%	9.937.334
Rondônia	2.272.851	25%	1.653.626	18%	5.302.598	57%	9.229.075
São Paulo	2.883.399	21%	3.024.157	22%	7.578.881	56%	13.486.437
Sergipe	192.873	22%	180.744	21%	498.027	57%	871.644
Tocantins	1.778.081	24%	1.440.704	19%	4.346.867	57%	7.565.652
<b>Total</b>	<b>33.494.457</b>	<b>22%</b>	<b>32.543.850</b>	<b>21%</b>	<b>88.695.001</b>	<b>57%</b>	<b>154.733.308</b>



**Table 5.** Existing bovine population, according to sub-populations and to the size of the herds, 2005

UF and regions (sub-populations)	Total of bovines according to the size of the herds						Total
	Up to 20 bovines		21 to 50 bovines		More than 50 bovines		
Acre and two municipalities of the State of Amazonas	86.496	3,7%	172.130	7,4%	2.071.442	88,9%	2.330.068
Bahia	1.534.899	17,7%	1.506.804	17,4%	5.630.223	64,9%	8.671.926
Federal District	17.480	16,4%	19.614	18,5%	69.187	65,1%	106.281
Espírito Santo	113.714	6,6%	224.169	13,1%	1.372.250	80,2%	1.710.133
Goiás	310.229	1,7%	1.019.843	5,6%	16.983.548	92,7%	18.313.620
Mato Grosso	324.926	1,2%	901.278	3,3%	26.500.034	95,6%	27.726.238
Mato Grosso do Sul (Pantanal, May)	2.316	0,1%	6.268	0,3%	2.165.405	99,6%	2.173.989
Mato Grosso do Sul (Pantanal, November)	870	0,1%	2.603	0,2%	1.160.315	99,7%	1.163.788
Mato Grosso do Sul (Plateau)	76.639	0,4%	298.243	1,5%	19.380.935	98,1%	19.755.817
Minas Gerais (Centre-West Circuit)	722.174	6,4%	1.578.990	14,1%	8.911.941	79,5%	11.213.105
Minas Gerais (East Circuit)	903.384	9,8%	1.418.168	15,4%	6.877.297	74,8%	9.198.849
Paraná	1.013.635	10,7%	1.340.102	14,1%	7.124.526	75,2%	9.478.263
Rio de Janeiro	129.976	7,2%	274.735	15,3%	1.396.378	77,5%	1.801.089
Rio Grande do Sul	1.387.922	14,0%	1.350.758	13,6%	7.198.654	72,4%	9.937.334
Rondônia	232.272	2,5%	711.804	7,7%	8.284.999	89,8%	9.229.075
São Paulo	668.713	5,0%	1.405.294	10,4%	11.412.430	84,6%	13.486.437
Sergipe	153.102	17,6%	151.281	17,4%	567.261	65,1%	871.644
Tocantins	152.258	2,0%	469.590	6,2%	6.943.804	91,8%	7.565.652
	<b>7.831.005</b>	<b>5,1%</b>	<b>12.851.674</b>	<b>8,3%</b>	<b>134.050.629</b>	<b>86,6%</b>	<b>154.733.308</b>

### 3.3. Method of diagnosis

This study used as analytical method for assessing the immunity level of the bovine population the essay of competition enzymatic immune-absorption at the fluid stage (ELISA-CFL) standardised by PANAFTOSA for detecting specific antibodies against proteins of the viral capsid. The essay was developed in 1985 by Mc Cullough *et al*, in the Reference Laboratory of the OIE for Foot and Mouth Disease in Pirbright, UK (WRL). The technique was initially applied to characterise epitopes of the foot and mouth disease virus. Next year, also at the WRL, Hamblin *et al* (1986) adapted the essay to measure post-infection or vaccinal antibodies. Later on, several laboratories adopted the methodology, and at PANAFTOSA it was adapted for the study of vaccinal antibodies with South-American strains (Vianna Filho *et al*, 1993).

The behaviour of the ELISA-CFL developed at PANAFTOSA was assessed in the sub-project “*Correlación de técnicas de control de vacuna antiaftosa*” carried out in cooperation among the countries of the Southern Cone (participants in the sub-project of the Basin of the Riverplate for the eradication of the disease), PANAFTOSA and the European Economic Community (EEC).

The project studied the response, as to the level of circulating antibodies, determined by the ELISA-CFL technique and other *in vitro* essays, vis-à-vis the response of vaccinated bovines and challenged via intra-dermo-lingual with 10.000 infectious doses per bovine 50% (DIB 50%) of foot and mouth disease virus in the direct PGP essay (Foot Generalisation Essay). This direct essay measures the protecting immunity, which means, *inter alia*, a complex interaction of antibodies, which varies in terms of affinity and isotypes, and of phagocyte cells with viral antigens, making up the antigen-antibody complexes. The ability to form the aforementioned complexes will limit or prevent the generalisation of the disease and the emergence of clinical foot lesions.

The study was developed starting from three serum collections, obtained 28 to 30 days after vaccination, from bovines vaccinated in official essays of power control, with trivalent vaccines against foot and mouth disease (O1 Campos, A24 Cruzeiro and C3 Indaial) of oily formulation, and submitted to the direct essay of PGP challenge with the official strains of production O1 Campos, A24 Cruzeiro and C3 Indaial. The serum collections from bovines vaccinated and challenged to the viruses O1 Campos, A24 Cruzeiro and C3 Indaial, were defined, by mutual agreement, by a group of consultants of Argentina, Brazil, Uruguay, PANAFTOSA and the EEC. A fourth collection made up of serums of zero days after vaccination was also included in the study.

The serums were analysed by titling in the different indirect essays vis-à-vis the virus used for the challenge in the direct PGP essay. The titles obtained for each individual were recorded together with the response of the same individual to the PGP (Protected or Non-Protected from the challenge with 10.000 (DIB 50%) of foot and mouth disease virus). The statistical analysis between the indirect response (level of antibodies) and a direct response (result of the PGP) showed the existence of a significant co-relation between titles of circulating antibodies and protection to the PGP, which allowed for the establishment of a function of regression of the logistic type. This model can be used both as a function of regression or as a discriminating function. In the first case it is possible to estimate the expectation of protection (the likelihood of being protected) of a bovine based upon the knowledge of its title of antibodies and, in the second case, starting from the establishment of a cutting or discriminating value, to classify, based upon the knowledge of the title of antibodies of a bovine, whether it belongs to the population of PROTECTED OR NON-PROTECTED bovines when exposed to 10.000 (DIB 50%) of foot and mouth disease virus.

The ELISA essay is deemed easy to apply, low cost and it presents replicable results and uses non-activated reagents, which guarantees bio-security.

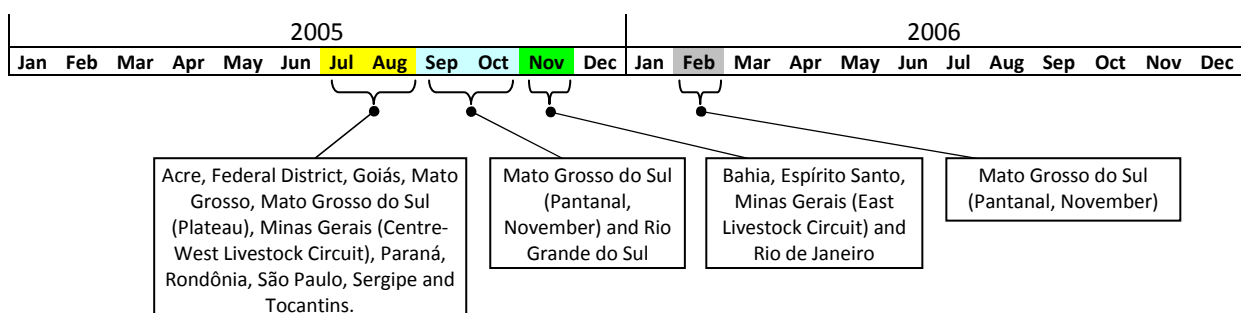
### 3.4. Sampling outline

The object of this study by sampling was to provide estimations on the immunity status of the bovine population of the zone free from foot and mouth disease with vaccination starting from the assessment of the number of bovines that would respond as protected if exposed to the foot and mouth disease virus. The study also allowed for the gathering of important information for the eradication programme, and for the assessing of the exposure to the risk of viral circulation in the free zone with vaccination, as well as for proving, even if globally, possible existing structure or conjuncture-related weaknesses.

On the other hand, the estimations of the prevalence of protected animals in the zone free from foot and mouth disease with vaccination will be used as a counterpoint for verifying the rates of vaccine coverage assessed by the programme starting from the recording of the declaration of vaccination carried out by the owners of animals with the local veterinary units.

#### Period of collection

Bearing in mind the objective of assessing the immunity status of the bovine population as a response to the systematic implementation of vaccination campaigns, and considering that the period of time between the inoculation of the vaccine and the collection of the blood directly interferes in the levels of humour response of vaccinated animals, the collections of blood were programmed to occur between 30 and 90 days after vaccination, period in which one expects the best responses as to the level of antibodies. Since the months in which the stages of vaccination occur vary among the sub-populations under study, four periods of collection, were defined, as it can be seen in Picture 6. Most of the sample collection was forecast for the months of July and August, 2005.



Picture 6. Periods forecast for sample collection, according to the sub-populations considered

Size of the sample and strategy of diagnosis

The size of the sample, for each sub-population considered, depends on the level of confidence required, on the maximum acceptable sampling error, on the proportion of protected animals one expects to find in the population and, in this case, on the characteristics of the laboratory essay used. For its calculation we used the formula below, according to Rahme & Joseph (1998).

$$n = \frac{Z_{\alpha/2}^2 p (1 - p)}{w^2 (Se + Sp - 1)^2}$$

Where:

- n = number of samples (adjusted according to Se and Sp of the laboratory test)
- $Z_{\alpha/2}$  = abscissa of the normal curve for  $(1-\alpha)$  of confidence
- p = proportion expected of protected animals in the population (NPP)
- w = amplitude of the interval of  $(1-\alpha)$  of confidence
- Se = sensitiveness of the laboratory test
- Sp = specificity of the laboratory test

The level of confidence desired was defined at 95% and the acceptable sampling error (w), at 15%. Vis-à-vis the proportion expected of protected animals (p), it is necessary to recognise that the higher the value expected, the lower will be the size of the sample necessary. Thus, considering the vaccination coverage recorded starting from the records presented to the official veterinary service, as made available in item 2.3 of this document (values higher than 90%), one could expect a significant level of population protection. On the other hand, it is known that the level of protection depends on the age composition of the population and on the number of vaccinations received by the animals in the context of a programme of vaccination systematically carried out for over a decade, with important rates of vaccination coverage, both for herds and for animals. Thus, it was decided to conduct the study considering the need for estimations independent from the level of protection according to age groups (sub-populations as to age) in the framework of the previously defined sampling parameters.

It is expected that the level of population protection will be smaller than the rate of vaccination coverage of the population, since it is impossible to assume that every vaccinated animal is protected against exposure to the agent. As it was mentioned before, the activity of vaccination is meant to provide levels of population immunity that are sufficient to prevent the spreading of the agent. It varies, in the case of foot and mouth disease, according to the number of doses given to each bovine, which, in the case of the Brazilian zone free from foot and mouth disease, also has to do with the origin of the animal (whether or not born in the property), since animals that have been move receive, in some cases, reinforcement doses of the vaccine. Thus, and with the concern of not establishing an insufficient sample for complying with the objectives of the study, we used the following values for the proportion expected of protected animals (p), according to the age groups defined in the study: **bovines between 6 and 12 months, 65%; bovines between 13 and 24 months, 75%; and bovines with more than 24 months of age, 85%.**

The sensitiveness and specificity of the laboratory test depend on the cutting value to be used to classify, based upon the title of antibodies measured by the ELISA-CFL, bovines as belonging to the population of PROTECTED or NON-PROTECTED animals. Bovines whose serums present title lower than or equal to that used as cutting discriminating value are classified as NON-PROTECTED, while those with higher titles are considered PROTECTED. As it was informed before, the sub-project of *“Correlación de Técnicas de Control de Vacunas Antiaftosa”*

determined a logistic function of regression for each of the vaccinal strains, O1 Campos, A24 Cruzeiro and C3 Indaial of foot and mouth disease. In this study said functions were used as discriminating functions and the determination of the respective cutting values was carried out with the support of PANAFTOSA, using the technique of two-graph-receiver operating characteristic (TG-ROC), by means of the programme Computer Methods for Diagnosis Tests (CMDT)\*, favouring the capacity of the essay in identifying NON-PROTECTED bovines (specificity). In the determination of the cutting values for the viruses O1 Campos and A24 Cruzeiro we used sets of data from the official power control of vaccines against foot and mouth disease, by direct essay (PGP), of Instituto Colombiano Agropecuario (LANIP/ICA – Colombia). These data do not show a linkage with the set of data that produced the discriminating functions established in the sub-project of “*Correlación de Técnicas de Control de Vacunas Antiaftosa*”. For the virus C3 Indaial the cutting title was determined starting from the data of the aforementioned sub-project, since we did not count on another source of data. The cutting values for each type of virus as well as the sensitiveness and specificity of the laboratory test can be found in Table 6.

Table 6 also presents the total forecast of samples, according to the type of virus and age groups, defined according to the parameters of sampling established. The largest sample was the one for virus C, mostly due to the lesser sensitiveness of the laboratory test, which led to the forecast of collection of 1.962 samples. Viruses A and O, according to the aforementioned works, feature behaviour similar to the correspondence between the challenges of the PGP essay and the humour titles. The small differences between the values used for sensitiveness and specificity led to the determination of different sizes for the number of samples, respectively, 778 and 932. However, for the two viral types we used in the laboratory the same cutting title: 2.10, and for the determination of the true prevalence we used values of sensitiveness and specificity of 0.8333 and 0.8571, respectively.

**Table 6.** Forecast number of samples, according to the type of virus, age groups and parameters used in the calculation of the size of the samples

Type of virus	Age Group (months)	p	Level of confidence	w	Se of the laboratory test	Sp of the laboratory test	Cutting Title	Cutting Title corrected *	Number of samples
A	6 to 12	0.65							326
	13 a 24	0.75	0.95	0.15	0.8333	0.8571	2.083	2.10	269
	More than 24	0.85							183
	Total of samples								778
O	6 to 12	0.65							391
	13 a 24	0.75	0.95	0.15	0.7158	0.9149	2.095	2.10	322
	More than 24	0.85							219
	Total of samples								932
C	6 to 12	0.65							823
	13 a 24	0.75	0.95	0.15	0.5179	0.9167	2.355	2.40	678
	More than 24	0.85							461
	Total of samples								1,962

\* defined for the implementation of essays in the laboratory.

As a result of the application of the laboratory test, we obtained the apparent proportion of bovines protected against foot and mouth disease called  $\theta_{AP}$  and understood as:

$$\theta_{AP} = \frac{\text{Number of bovines classified as protected}}{\text{Total of bovines in the sample}}$$

The apparent proportion was corrected as a function of the sensitiveness and specificity of the test, providing a punctual estimate of the true proportion of protected animals in the population (true prevalence, defined as  $\theta_{VE}$ ). For this correction we used the following formula, according to Klein and Costa (1987):

\* CMDT version 1.0  $\beta$ . Designed by Mathias Greiner (FU-Berlin) and developed by Jens Briesofsky.

$$\Theta_{VE} = \frac{(\Theta_{AP} + Sp - 1)}{(Se + Sp - 1)}$$

Where:

$\Theta_{VE}$  = true prevalence

$\Theta_{AP}$  = apparent prevalence

Se = sensitiveness of the laboratory test

Sp = specificity of the laboratory test

When necessary, in the cases in which the calculation of the true prevalence surpassed the limit of 100%, we used the Bayes' method, according to Lew & Levy (1989), replacing the value of the apparent prevalence in the previous formula for an estimator of the *a priori* prevalence, calculated by means of the following formula:

$$\text{Bayes' estimator} = \int_{1-Sp}^{Se} P^{x+1}(1-P)^{n-x} dP + \int_{1-Sp}^{Se} P^x(1-P)^{n-x} dP$$

Where:

P = apparent prevalence

x = positive samples

n = total of samples

d = derived

For the solution of the integral calculations necessary for calculating the Bayes' estimator we used the programme X(PLORE) developed by David Meredith, of the Department of Mathematics of the University of São Francisco, using the following lines of command:

“numerator = ln (P^(positive samples + 1) \* (1-P)^negative samples), P= 1-Sp to Se)”

“denominator = ln (P^(positive samples) \* (1-P)^negative samples), P= 1-Sp to Se)”

#### Distribution and allocation of the sample

The elementary sampling units are the bovines that, and since they are grouped in rural properties, the latter become the primary sampling units (UPAs) and define the need for implementing a sampling plan in two stages. For each sub-population established by the crossing of the units of the Federation with strategies of vaccination, 100 rural properties were selected as starting point for the collection of the samples. When it was impossible to find a sufficient number of bovines in the age group required in the property selected, the sample was complemented with animals from one or more neighbouring properties, belonging to the same category of herd size. Should the coverage of properties vaccinated be of 90%, the number of 100 properties for selection would lead to the likelihood of 99.99% that the sample, in any sub-population, would include at least one property where there has been no vaccination. Or else, the likelihood of 58.31% of the inclusion of up to 10 properties; of 52.56%, between 5 and 10 properties; and of 13.20%, of the inclusion of exactly 10 properties where vaccination was not carried out. The value of 90% for the coverage of properties vaccinated is deemed conservative when compared to those presented in Table 2 and to the fact that the properties without record of vaccination are investigated by the official veterinary service after completion of the stages.

The categorisation as to the size of the herds (up to 20 animals, between 21 and 50 and more than 50 animals) was dealt with as domains of sampling in each of the sub-populations and the number of UPAs in each of them was allocated proportionally to the bovine population in each domain. With a view at minimising the cost of access to the elementary sampling units, in each property selected samples of bovines were collected, according to the age groups considered, starting from a random process that ensured the maintenance of the global sampling fraction.

The properties were randomly selected by the DSA starting from the databases sent by the state organisations of animal sanitary defence. The database concerning each sub-population (unit of the Federation x strategy of vaccination) was initially subdivided according to the domains (properties with up to 20 bovines, with 21 to 50 bovines and with more than 50 bovines) and the sample concerning each of the age groups proportionally allocated to the bovine population of these categories. Table 7 shows the distribution of the samples, according to the categories of herds and age groups considered. The selection of the elementary units in each property was carried out by simple random sampling.

**Table 7.** Forecast of samples, according to sub-populations, age groups and size of the herds

Age Group	UF and regions (sub-populations)	Total of samples according to the size of the herds			Total
		Up to 20 bovines	21 to 50 bovines	More than 50 bovines	
Bovines Up to 12 months	Acre and two municipalities of the State of Amazonas	22	32	302	356
	Bahia	83	68	267	418
	Federal District	13	14	536	814
	Espírito Santo	3	5		
	Goiás	37	46	272	355
	Minas Gerais (Centre-West Circuit)	10	22	313	345
	Minas Gerais (East Circuit)	67	11	655	840
	Mato Grosso do Sul (Pantanal, May)	8			
	Mato Grosso do Sul (Pantanal, November)	49	65	297	411
	Mato Grosso do Sul (Plateau)	5	5	351	361
	Mato Grosso	6	2	391	399
	Paraná	7	10	400	417
	Rio de Janeiro	23	13	330	366
	Rondônia	52	53	295	400
	Rio Grande do Sul	59	13	628	822
	Sergipe	5			
	São Paulo	14	33	296	343
	Tocantins	59	47	237	343
Bovines of 13 to 24 months	Acre and two municipalities of the State of Amazonas	12	17	531	831
	Bahia	6	4		
	Federal District	32	45	337	414
	Espírito Santo	10	22	298	330
	Goiás	17	26	249	292
	Minas Gerais (Centre-West Circuit)	68	58	217	343
	Minas Gerais (East Circuit)	11	12	409	644
	Mato Grosso do Sul (Pantanal, May)	3	2		
	Mato Grosso do Sul (Pantanal, November)	28	35	224	287
	Mato Grosso do Sul (Plateau)	8	18	260	286
	Mato Grosso	56	95	539	690
	Paraná	40	53	244	337
	Rio de Janeiro	4	4	289	297
	Rondônia	1	3	326	330
	Rio Grande do Sul	5	6	329	340
	Sergipe	19	10	272	301
	São Paulo	43	44	244	331
	Tocantins	51	11	518	680
Bovines with more than 24 months	Acre and two municipalities of the State of Amazonas	1	1		
	Bahia	11	27	244	282
	Federal District	48	40	194	282
	Espírito Santo	10	14	417	666
	Goiás	5	4		
	Minas Gerais (Centre-West Circuit)	27	39	285	351
	Minas Gerais (East Circuit)	8	21	249	278
	Mato Grosso do Sul (Pantanal, May)	13	18	169	200
	Mato Grosso do Sul (Pantanal, November)	44	38	146	228
	Mato Grosso do Sul (Plateau)	76	83	306	465
	Mato Grosso	19	26	151	196
	Paraná	6	12	177	195
	Rio de Janeiro	36	67	367	470
	Rondônia	27	36	167	230
	Rio Grande do Sul	3	3	196	202
	Sergipe	2	2	222	226
	São Paulo	3	4	223	230
	Tocantins	13	8	186	207
Bovines with more than 24 months	Acre and two municipalities of the State of Amazonas	32	29	165	226
	Bahia	36	83	342	461
	Federal District	11	19	165	195
	Espírito Santo	31	27	133	191
	Goiás	70	10	319	490
	Minas Gerais (Centre-West Circuit)	1			
	Minas Gerais (East Circuit)	17	26	193	236
	Mato Grosso do Sul (Pantanal, May)	6	12	175	193
	Mato Grosso do Sul (Pantanal, November)				
	Mato Grosso do Sul (Plateau)				
	Mato Grosso				
	Paraná				
	Rio de Janeiro				
	Rondônia				
	Rio Grande do Sul				
	Sergipe				
	São Paulo				
	Tocantins				

### 3.5. Assessment of the levels of immunity

Taking into account the fact that every lot of vaccine produced in the country is approved for the three types of virus, we chose to carry out the laboratory test for one type of virus in each sub-population, reducing time and cost in terms of laboratory procedures. The type of virus assessed in each sub-population was randomly chosen, with greater likelihood for the types A and O, leading to the following distribution:

Type of virus	Sub-population
O	Acre and two municipalities of the State of Amazonas
	Espírito Santo
	Goiás
	Mato Grosso
	Mato Grosso do Sul (Pantanal, May)
	Rio Grande do Sul
	Rondônia
	Tocantins
A	Bahia
	Mato Grosso do Sul (Pantanal, November)
	Mato Grosso do Sul (Plateau)
	Minas Gerais (East Circuit)
	Paraná
C	São Paulo
	Federal District
	Minas Gerais (Centre-West Circuit)
	Rio de Janeiro
	Sergipe

### 3.6. Implementation of the activities of collection and of information recording

For the conduction of the work there were meetings aimed at the standardisation of the actions involving the DSA and the state organisations of animal sanitary defence, which were appointed responsible for the performance of the activities of collection, survey and recording of the information. For the control of the database generated by the study an application in Microsoft Office Access was developed, which was put in place at the central units of the state organisations of animal sanitary defence, at the laboratory of the MAPA in charge of the application of diagnosis tests and in the DSA. A manual for guidance and standardisation of the activities of collection, survey and recording of the information was also prepared and made available to all representatives of technical field teams.

For the recording of the information concerning the rural properties that participated in the study and for conducting the interview with the people responsible for the animals we used a single form, according to the model presented in Annex 2.

The information on each rural property involved identification, location, the structure of the existing bovine herd and data on the last vaccination against foot and mouth disease.

For the control and recording of the information on the samples collected we used also a single form, as per model presented in Annex 3. Each sample received a single identification and was accompanied by information on the animal (sex, age, origin and number of vaccinations received).

Once the stage of collection and interviews was completed, the state organisations of animal sanitary defence sent the samples, accompanied by the respective interview and collection forms, to LANAGRO, located in Pedro Leopoldo, MG, where the diagnosis tests were carried out. The database with the results of the diagnosis carried out, the copies of the interview and collection forms were referred to the DSA, where the final analyses were carried out together with PANAFTOSA.

## **4. Information on the sample established**

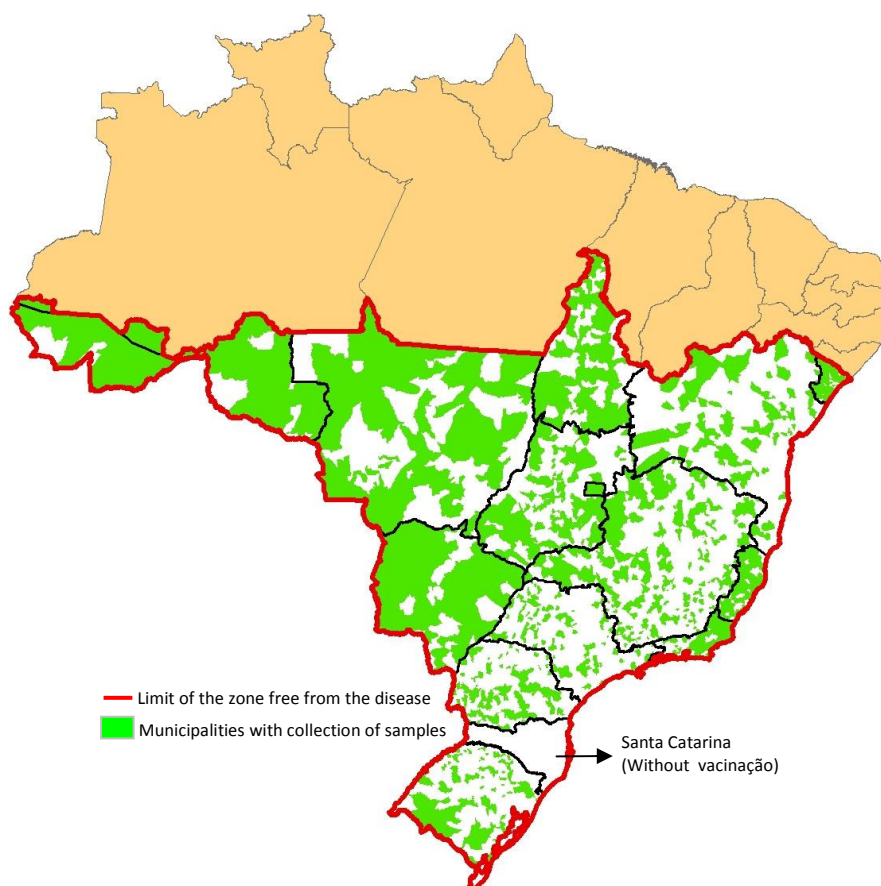
### ***4.1. Implementation and profile of the sample***

Picture 6 presents the space distribution of the samples according to the municipalities with at least one rural property where blood collections interviews were carried with those in charge of the animals. With the exception of Santa Catarina, the entire zone free from foot and mouth disease was subjected to study, and the geographical distribution of the sample was considered appropriate.

The sample initially selected of 100 rural properties was expanded in all the sub-populations considered. The increments recorded were necessary to reach the total forecast of elementary sampling units (bovines). The sub-population the featured greater increment of properties was represented by the sub-population of the Federal District, in the categories of size of herds “Up to 20 bovines” and “more than 50 bovines”. In the other sub-populations, this increment varied from 2 to 13 properties (Table 8).

The Information on the number of samples collected per sub-population, age group and size of herds, are made available in Table 9. Considering all the sub-populations, 1.272 samples were collected, in addition to what had been forecast, totalling 20.423 bovines sampled and assessed as to the level of antibodies. Out of these samples, 1.898 (9%) were obtained in herds with up to 20 bovines; 2.477 (12%) in herds between 21 and 50 bovines; and 16.048 (79%) in herds with more than 50 bovines. Bearing in mind the fact that for each group of age and type of virus an independent sample was obtained in each of the sub-populations, we point out that within the study they were distributed as follows: 8.565 samples (42%) of bovines between 6 and 12 months, 7.017 (34%), of bovines between 13 and 24 months, and 4.841 (24%), of bovines with more than 24 months of age.





**Picture 7.** Geographical distribution of the sample according to municipalities where interviews and sample collection were carried out

**Table 8.** Number of rural properties with sample collection, according to sub-populations and size of herds

UF and regions (Sub-populations)	Total of properties according to the size of the herd									
	Up to 20 bovines			21 to 50 bovines			More than 50 bovines			Total carried out
	Forecast	Carried out	Difference	Forecast	Carried out	Difference	Forecast	Carried out	Difference	
Acre and two municipalities of the State of Amazonas	39	39	0	25	28	3	36	37	1	104
Bahia	66	69	3	20	21	1	14	16	2	106
Federal District	64	89	25	20	30	10	16	31	15	150
Espírito Santo	44	44	0	27	28	1	29	30	1	102
Goiás	21	24	3	26	26	0	53	54	1	104
Mato Grosso	25	30	5	23	23	0	52	57	5	110
Mato Grosso do Sul (Pantanal, May)	13	12	-1	11	11	0	76	79	3	102
Mato Grosso do Sul (Pantanal, November)	28	9	-19	5	6	1	67	88	21	103
Mato Grosso do Sul (Plateau)	14	14	0	18	18	0	68	71	3	103
Minas Gerais (Centre-West Circuit)	50	51	1	24	26	2	26	33	7	110
Minas Gerais (East Circuit)	58	59	1	22	25	3	20	20	0	104
Paraná	65	71	6	19	20	1	16	22	6	113
Rio de Janeiro	74	72	-2	13	20	7	13	19	6	111
Rio Grande do Sul	69	75	6	19	18	-1	12	17	5	110
Rondônia	27	27	0	26	28	2	47	49	2	104
São Paulo	39	39	0	27	29	2	34	38	4	106
Sergipe	65	60	-5	20	31	11	16	19	3	110
Tocantins	25	22	-3	27	28	1	48	54	6	104

**Table 9.** Comparison between samples collected and forecast, according to sub-populations, age group and size of herds

Age Group	UF and regions (sub-populations)	Total of bovines sampled according to the size of the herds								
		Up to 20 bovines			21 to 50 bovines			More than 50 bovines		
		Forecast	Carried out	Difference	Forecast	Carried out	Difference	Forecast	Carried out	Difference
Bovines Up to 12 months	Acre and two municipalities of the State of Amazonas	13	22	9	28	32	4	287	302	15
	Bahia	64	83	19	69	68	-1	260	267	7
	Federal District	152	133	-19	174	145	-29	498	536	38
	Espírito Santo	17	37	20	38	46	8	272	272	0
	Goiás	7	10	3	22	22	0	299	313	14
	Mato Grosso	5	24	19	14	12	-2	308	330	22
	Mato Grosso do Sul (Pantanal, May)	1	5	4	2	5	3	325	351	26
	Mato Grosso do Sul (Pantanal, November)	1	6	5	1	2	1	391	391	0
	Mato Grosso do Sul (Plateau)	2	7	5	7	10	3	383	400	17
	Minas Gerais (Centre-West Circuit)	50	67	17	121	118	-3	654	655	1
	Minas Gerais (East Circuit)	37	49	12	60	65	5	296	297	1
	Paraná	46	52	6	60	53	-7	287	295	8
	Rio de Janeiro	24	59	35	48	135	87	213	628	415
	Rio Grande do Sul	51	61	10	47	45	-2	230	237	7
	Rondônia	8	14	6	26	33	7	293	296	3
	São Paulo	20	32	12	44	45	1	329	337	8
Sergipe	152	126	-26	153	174	21	519	531	12	
Tocantins	7	10	3	22	22	0	298	298	0	
Bovines of 13 to 24 months	Acre and two municipalities of the State of Amazonas	9	17	8	18	26	8	244	249	5
	Bahia	53	68	15	53	58	5	218	217	-1
	Federal District	110	113	3	127	122	-5	443	409	-34
	Espírito Santo	12	28	16	29	35	6	229	224	-5
	Goiás	3	8	5	11	18	7	256	260	4
	Mato Grosso	3	19	16	8	10	2	260	272	12
	Mato Grosso do Sul (Pantanal, May)	1	4	3	1	4	3	268	289	21
	Mato Grosso do Sul (Pantanal, November)	1	1	0	1	3	2	322	326	4
	Mato Grosso do Sul (Plateau)	2	5	3	5	6	1	317	329	12
	Minas Gerais (Centre-West Circuit)	34	56	22	81	95	14	565	539	-26
	Minas Gerais (East Circuit)	25	40	15	44	53	9	254	244	-10
	Paraná	31	43	12	44	44	0	249	244	-5
	Rio de Janeiro	45	51	6	99	111	12	536	518	-18
	Rio Grande do Sul	33	50	17	35	38	3	202	194	-8
	Rondônia	6	11	5	19	27	8	245	244	-1
	São Paulo	16	27	11	34	39	5	274	285	11
Sergipe	113	105	-8	116	144	28	450	417	-33	
Tocantins	4	8	4	13	21	8	253	249	-4	
Bovines with more than 24 months	Acre and two municipalities of the State of Amazonas	8	13	5	14	18	4	163	169	6
	Bahia	42	44	2	39	38	-1	139	146	7
	Federal District	73	76	3	81	83	2	308	306	-2
	Espírito Santo	14	19	5	26	26	0	144	151	7
	Goiás	4	6	2	11	12	1	169	177	8
	Mato Grosso	3	13	10	6	8	2	176	186	10
	Mato Grosso do Sul (Pantanal, May)	1	3	2	1	3	2	183	196	13
	Mato Grosso do Sul (Pantanal, November)	1	2	1	1	1	0	219	223	4
	Mato Grosso do Sul (Plateau)	1	3	2	4	4	0	215	223	8
	Minas Gerais (Centre-West Circuit)	34	36	2	68	67	-1	361	367	6
	Minas Gerais (East Circuit)	24	27	3	36	36	0	160	167	7
	Paraná	24	32	8	31	29	-2	165	165	0
	Rio de Janeiro	33	36	3	69	83	14	360	342	-18
	Rio Grande do Sul	26	33	7	25	25	0	133	133	0
	Rondônia	5	11	6	15	19	4	164	165	1
	São Paulo	12	17	5	23	26	3	186	193	7
Sergipe	82	70	-12	79	101	22	302	319	17	
Tocantins	4	6	2	12	12	0	168	175	7	

#### 4.2. History of vaccination according to information received from those in charge of the animals

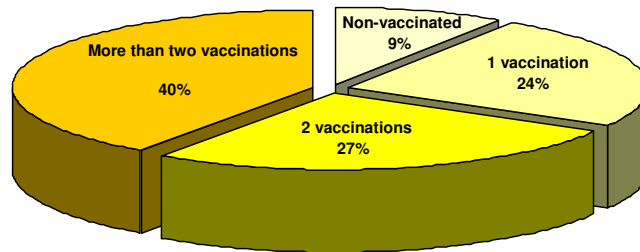
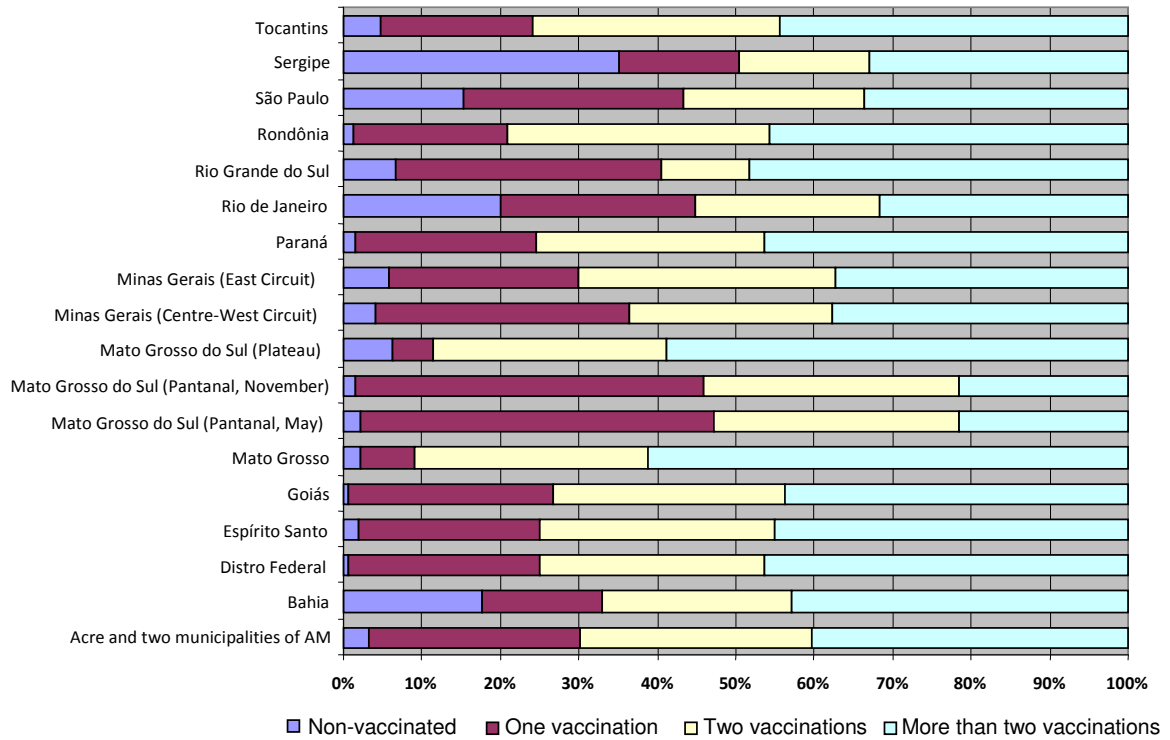
Within the study, the distribution of the history of vaccinations, according to the declarations of those responsible for the animals selected to make up the samples studied, indicated that 9% of the bovines sampled did not have a history of vaccination, 24% had undergone one vaccination, 27% two vaccinations and 40% more than two vaccinations (Table 10 and Picture 8).

The highest frequency of bovines reported as non-vaccinated was recorded in the sub-populations represented by Sergipe (35%), Rio de Janeiro (20%), Bahia (18%) and São Paulo (15%). In the other sub-populations, the participation of non-vaccinated bovines was lesser than 7%. In the other extreme of the sequence, bovines with more than two vaccinations, the sub-populations that highlighted with the highest percentage participation of vaccinated bovines were those represented by Mato Grosso (61%) and the Plateau of Mato Grosso do Sul (59%), where the strategy of vaccination includes the inoculation of a reinforcement dose in bovines under the age of 12 months. Regarding the frequency of bovines with a history of vaccination in the central groupings of one and two vaccinations, highlight the sub-populations identified as Mato Grosso and Mato Grosso do Sul (Plateau), with a strong predominance of the group of two vaccinations, and Rio Grande do Sul, in the opposite sense, the group of one vaccination. One can also noticed, though not so strongly, a trend towards the group of two vaccinations in the sub-populations concerning Rondônia and Tocantins.

Regarding animals with a history of non-vaccination, supplementary information related to the origin of the animals will be presented ahead, trying to help understand the consistency of this information obtained from the people in charge of the animals sampled.

**Table 10.** Composition of the samples, according to history of vaccination and sub-populations

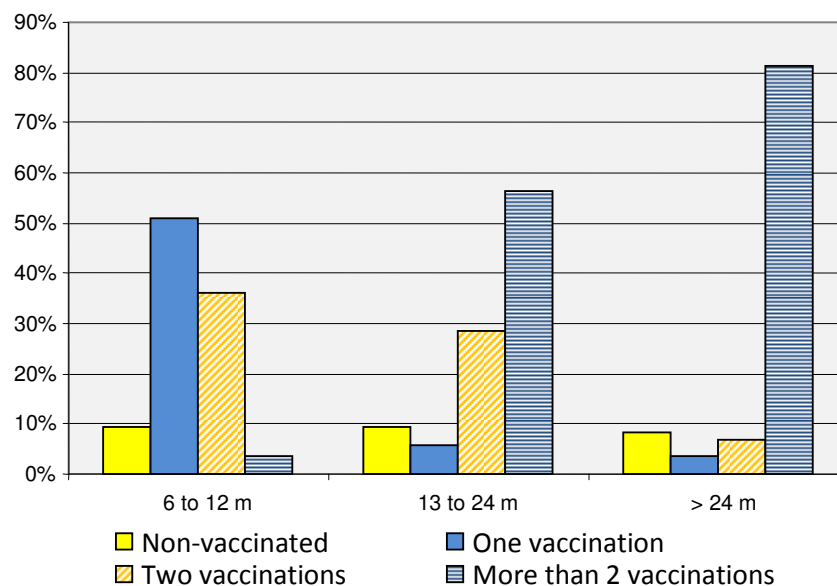
Sub-populations	Non-vaccinated		One vaccination		Two vaccinations		More than two vaccinations		Total
Acre and two municipalities of the State of Amazonas	28	3%	228	27%	250	29%	342	40%	848
Bahia	174	18%	153	15%	237	24%	425	43%	989
Federal District	14	1%	466	24%	552	29%	891	46%	1.923
Espírito Santo	16	2%	194	23%	251	30%	377	45%	838
Goiás	6	1%	215	26%	243	29%	362	44%	826
Mato Grosso	19	2%	60	7%	260	30%	535	61%	874
Mato Grosso do Sul (Pantanal, May)	18	2%	388	45%	268	31%	186	22%	860
Mato Grosso do Sul (Pantanal, November)	14	1%	424	44%	312	33%	205	21%	955
Mato Grosso do Sul (Plateau)	61	6%	52	5%	294	30%	580	59%	987
Minas Gerais (Centre-West Circuit)	81	4%	649	32%	514	26%	756	38%	2.000
Minas Gerais (East Circuit)	56	6%	237	24%	321	33%	364	37%	978
Paraná	15	2%	220	23%	278	29%	444	46%	957
Rio de Janeiro	392	20%	486	25%	465	24%	620	32%	1.963
Rio Grande do Sul	55	7%	275	34%	92	11%	394	48%	816
Rondônia	10	1%	162	20%	274	33%	374	46%	820
São Paulo	154	15%	279	28%	231	23%	337	34%	1.001
Sergipe	696	35%	306	15%	330	17%	655	33%	1.987
Tocantins	38	5%	156	19%	252	31%	355	44%	801
<b>Total</b>	<b>1.847</b>	<b>9%</b>	<b>4.950</b>	<b>24%</b>	<b>5.424</b>	<b>27%</b>	<b>8.202</b>	<b>40%</b>	<b>20.423</b>



**Picture 8.** Graphic representations of the distribution of the history of vaccinations, per sub-population and globally

Picture 9 presents the distribution of the history of vaccinations according to the age groups defined in the study. One notices, for bovines declared as without any vaccination, the absence of a trend towards the increment or decrease vis-à-vis the age groups, the percentage of animals in this condition in each of the age groups remaining at approximately 9%. This might indicate a systematic failure in the vaccination coverage, since what was expected was that the number of non-vaccinated animals would diminish as age increases or, on the other hand, represent the difficulty in classifying per age the animals belonging to the borderline between the two first age groups. Regarding this last point, there is the additional difficulty faced by those responsible for the animals in presenting the history of vaccinations of animals coming from other properties.

Also with regard to the history of vaccinations and the age of the animals sampled, one notices a trend towards the decrease of the number of bovines with one vaccination and towards the increment of the number of bovines with more than two vaccinations. It is also worth highlighting that approximately 85% of the bovines between 13 and 24 months showed two or more vaccinations, this being the age group to present the bigger movement for the completion of the animal-husbandry cycle and the one that plays a relevant role in the epidemiology of foot and mouth disease, with more than two vaccinations.



**Picture 9.** Graphic representation of the history of vaccination of the animals sampled, according to age groups considered

Next we highlight some pieces of information on the history of vaccination reported by the people in charge of the animals sampled, according to the age groups used in the study and according to the sub-populations considered. This information is compiled in Table 11 and in Picture 10.

For the age group of bovines between 6 and 12 months, a-typical frequencies were recorded vis-à-vis the number declared of non-vaccinated bovines in the sub-populations identified as Sergipe (33%), Rio de Janeiro and Bahia (20%), São Paulo (14%) and the East Circuit of Minas Gerais (10%). These sub-populations featured, respectively, a history of one or two vaccinations for 65%, 81%, 78% and 89% of the bovines with ages between 6 and 12 months. Also for this age group, in the sub-populations of Mato Grosso do Sul (Pantanal, May) and Mato Grosso Sul (Pantanal, November) featured a history of one vaccination for 95% and 92% of the bovines sampled, in the sub-populations represented by Mato Grosso do Sul (Plateau) and Mato Grosso histories of two or more vaccinations were recorded for 87% of the bovines and in those identified as Rio Grande do Sul and Minas Gerais (Centre-West Circuit) there were, respectively, histories of one vaccination for 79% and 66% of the bovines. In the other sub-populations, the history of vaccination was concentrated, in a reasonably balanced manner, in one or two vaccinations, varying between 93% and 98%.

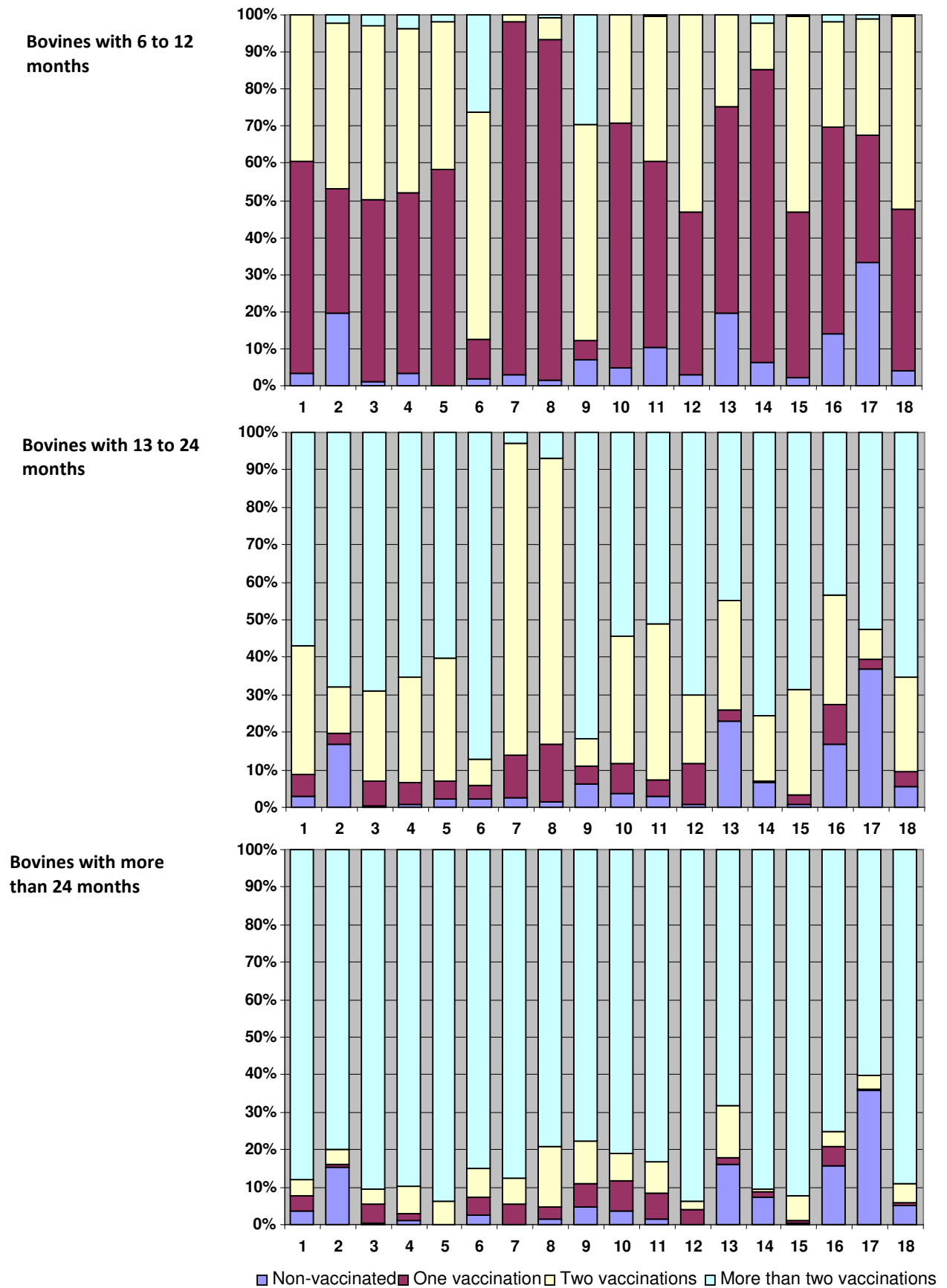
For the sample concerning the age group of bovines between 13 and 24 months of age, in the same sub-populations identified for the group of age of 6 to 13 months, except for the one related to Minas Gerais (East Circuit), a-typical frequencies vis-à-vis the number declared of non-vaccinated bovines were also recorded, representing 37% in Sergipe, 23% in Rio de Janeiro and 17% in São Paulo and in Bahia. In these same sub-populations, the history of bovines sampled with more than two vaccinations was of 53% in Sergipe, 74% in Rio de Janeiro and São Paulo and 68% in Bahia. The sub-populations identified as Mato Grosso and Mato Grosso do Sul

(Plateau) featured, respectively, 87% and 82% of the bovines of the sample with more than two vaccinations. Those identified as Pantanal, May and Pantanal, November of Mato Grosso do Sul featured, respectively, 83% and 76% of the bovines sampled with a history of two vaccinations. The samples of the sub-populations identified as Paraná and Rio Grande do Sul featured, respectively, 70% and 76% of the bovines with a history of more than two vaccinations, indicating, in both, a ratio of approximately one bovine with a history of two vaccinations for a little less than four bovines with histories of more than two vaccinations. For the sample of bovines obtained from the sub-population identified as Federal District, this same ratio was of approximately one for three, 68% of bovines with a history of more than two vaccinations. Regarding the samples obtained in the sub-populations identified as Rondônia, Espírito Santo and Tocantins, the record was of respectively 68%, 66% and 65% of bovines with a history of more than two vaccinations, representing the ratio of one bovine with two vaccinations for a little more than two bovines with histories of more than two vaccinations. The ratio of approximately one bovine with a history of two vaccinations for a little less than two bovines with histories of more than two vaccinations corresponds to the sub-populations of Goiás, Minas Gerais (Centre-West Circuit) and Acre, plus two municipalities of the State of Amazonas, with, respectively, 60%, 54% and 57% of the bovines with more than two vaccinations. The sub-population identified as Minas Gerais (East Circuit) featured a history of two or more vaccinations for 51% of the bovines included in the sample and the ratio of one bovine with a history of two vaccinations for a little more than one bovine with a history of more than two vaccinations.

The distribution of the history of vaccination for the bovines with more than 24 months showed the same profile identified in the sub-populations that featured atypical frequencies of bovines without records of vaccination for the other age groups. Thus, in the sub-population of Sergipe we identified 35% of the bovines with more than 24 months of age without records of vaccination, in Rio de Janeiro and in São Paulo 16%, and in Bahia, 15%. In Sergipe, the history of vaccinations of the bovines with at least one vaccination represented 60% of the bovines with more than two vaccinations and 4% of the bovines with two vaccinations. For the sub-population identified as São Paulo, the history of vaccination declared indicated that 75% of the bovines of the sample featured more than two vaccinations and 4%, two vaccinations. In the sub-population called Rio de Janeiro it was observed that 68% of the bovines of the sample had more than two vaccinations and 14%, two vaccinations. In the sub-population Bahia it was noticed that 80% of the bovines had had more than two vaccinations and 4%, two vaccinations. In those identified as Mato Grosso do Sul (Pantanal, November) and Mato Grosso do Sul (Plateau), there were records, respectively, of more than two vaccinations in 79% and 78% of the bovines of the sample and of two vaccinations in 16% and 11% of the bovines sampled. Histories of more than two vaccinations between 81% and 88% of the bovines were found for the sub-populations identified as Mato Grosso, Minas Gerais (Centre-West) and Minas Gerais (East Circuit). In these sub-populations percentages of the bovines with histories of two vaccinations between 7% and 9% were recorded. In the other sub-populations, the history of vaccinations indicated percentages higher than 88% of the bovines of the samples with more than two vaccinations and of at most 7% for bovines with two vaccinations.

**Table 11.** Composition of the sample according to age group, sub-population and number of vaccinations

Age	ID	Sub-population	Number of vaccinations							Total	
			0	1	2	> 2	0	1	2		> 2
6 to 12 months	1	Acre and two municipalities of the AM	12	3.4%	203	57.0%	141	39.6%	0	0.0%	356
	2	Bahia	81	19.4%	141	33.7%	186	44.5%	10	2.4%	418
	3	Federal District	10	1.2%	400	49.1%	379	46.6%	25	3.1%	814
	4	Espírito Santo	12	3.4%	173	48.7%	157	44.2%	13	3.7%	355
	5	Goiás	0	0.0%	201	58.3%	137	39.7%	7	2.0%	345
	6	Mato Grosso	7	1.9%	39	10.7%	224	61.2%	96	26.2%	366
	7	Mato Grosso do Sul (Pantanal, May)	10	2.8%	344	95.3%	7	1.9%	0	0.0%	361
	8	Mato Grosso do Sul (Pantanal, November)	6	1.5%	366	91.7%	24	6.0%	3	0.8%	399
	9	Mato Grosso do Sul (Plateau)	29	7.0%	22	5.3%	243	58.3%	123	29.5%	417
	10	Minas Gerais (Centre-West Circuit)	40	4.8%	554	66.0%	246	29.3%	0	0.0%	840
	11	Minas Gerais (East Circuit)	43	10.5%	206	50.1%	161	39.2%	1	0.2%	411
	12	Paraná	12	3.0%	175	43.8%	213	53.3%	0	0.0%	400
	13	Rio de Janeiro	162	19.7%	458	55.7%	202	24.6%	0	0.0%	822
	14	Rio Grande do Sul	22	6.4%	271	79.0%	42	12.2%	8	2.3%	343
	15	Rondônia	7	2.0%	154	44.9%	181	52.8%	1	0.3%	343
	16	São Paulo	58	14.0%	230	55.6%	118	28.5%	8	1.9%	414
	17	Sergipe	276	33.2%	286	34.4%	259	31.2%	10	1.2%	831
	18	Tocantins	13	3.9%	144	43.6%	172	52.1%	1	0.3%	330
13 to 24 months	1	Acre and two municipalities of the AM	9	3.1%	17	5.8%	100	34.2%	166	56.8%	292
	2	Bahia	58	16.9%	10	2.9%	42	12.2%	233	67.9%	343
	3	Federal District	3	0.5%	42	6.5%	154	23.9%	445	69.1%	644
	4	Espírito Santo	2	0.7%	17	5.9%	80	27.9%	188	65.5%	287
	5	Goiás	6	2.1%	14	4.9%	94	32.9%	172	60.1%	286
	6	Mato Grosso	7	2.3%	11	3.7%	20	6.6%	263	87.4%	301
	7	Mato Grosso do Sul (Pantanal, May)	8	2.7%	33	11.1%	247	83.2%	9	3.0%	297
	8	Mato Grosso do Sul (Pantanal, November)	5	1.5%	50	15.2%	252	76.4%	23	7.0%	330
	9	Mato Grosso do Sul (Plateau)	21	6.2%	16	4.7%	25	7.4%	278	81.8%	340
	10	Minas Gerais (Centre-West Circuit)	24	3.5%	57	8.3%	233	33.8%	376	54.5%	690
	11	Minas Gerais (East Circuit)	10	3.0%	15	4.5%	140	41.5%	172	51.0%	337
	12	Paraná	3	0.9%	36	10.9%	60	18.1%	232	70.1%	331
	13	Rio de Janeiro	156	22.9%	19	2.8%	200	29.4%	305	44.9%	680
	14	Rio Grande do Sul	19	6.7%	1	0.4%	49	17.4%	213	75.5%	282
	15	Rondônia	2	0.7%	7	2.5%	80	28.4%	193	68.4%	282
	16	São Paulo	59	16.8%	37	10.5%	103	29.3%	152	43.3%	351
	17	Sergipe	245	36.8%	18	2.7%	53	8.0%	350	52.6%	666
	18	Tocantins	15	5.4%	11	4.0%	70	25.2%	182	65.5%	278
More than 24 months	1	Acre and two municipalities of the AM	7	3.5%	8	4.0%	9	4.5%	176	88.0%	200
	2	Bahia	35	15.4%	2	0.9%	9	3.9%	182	79.8%	228
	3	Federal District	1	0.2%	24	5.2%	19	4.1%	421	90.5%	465
	4	Espírito Santo	2	1.0%	4	2.0%	14	7.1%	176	89.8%	196
	5	Goiás	0	0.0%	0	0.0%	12	6.2%	183	93.8%	195
	6	Mato Grosso	5	2.4%	10	4.8%	16	7.7%	176	85.0%	207
	7	Mato Grosso do Sul (Pantanal, May)	0	0.0%	11	5.4%	14	6.9%	177	87.6%	202
	8	Mato Grosso do Sul (Pantanal, November)	3	1.3%	8	3.5%	36	15.9%	179	79.2%	226
	9	Mato Grosso do Sul (Plateau)	11	4.8%	14	6.1%	26	11.3%	179	77.8%	230
	10	Minas Gerais (Centre-West Circuit)	17	3.6%	38	8.1%	35	7.4%	380	80.9%	470
	11	Minas Gerais (East Circuit)	3	1.3%	16	7.0%	20	8.7%	191	83.0%	230
	12	Paraná	0	0.0%	9	4.0%	5	2.2%	212	93.8%	226
	13	Rio de Janeiro	74	16.1%	9	2.0%	63	13.7%	315	68.3%	461
	14	Rio Grande do Sul	14	7.3%	3	1.6%	1	0.5%	173	90.6%	191
	15	Rondônia	1	0.5%	1	0.5%	13	6.7%	180	92.3%	195
	16	São Paulo	37	15.7%	12	5.1%	10	4.2%	177	75.0%	236
	17	Sergipe	175	35.7%	2	0.4%	18	3.7%	295	60.2%	490
	18	Tocantins	10	5.2%	1	0.5%	10	5.2%	172	89.1%	193



Picture 10. Graphic representation of the distribution of the sample per sub-population, age group and number of vaccinations (the figures in axis X correspond to the identification of the sub-populations according to Table 11)



### 4.3. Origin of the animals

Another piece of information obtained by the study that may indirectly influence the levels of population immunity for foot and mouth disease concerns the origin of the animals sampled, whether or not born in the rural properties involved in the work. Animals commercialised or transported, per force of legal standards, are subjected to supplementary vaccinations. In Tables 12 and 13 we present information on the participation in the sample, of animals born in the rural properties visited, according to age group and category of herd, respectively. In global terms, 78% of the bovines sampled were born in the same rural property (native). The sub-populations with the highest percentage participation of native bovines were represented by Rio Grande do Sul (98%); Tocantins and the marsh region of Mato Grosso do Sul (91%). The smaller percentage participation was recorded in Sergipe (51%). Regarding the age groups considered, the percentage of native animals was of 84% for bovines with ages between 6 to 12 months; 78%, for bovines between 13 and 24 months; and 65% for bovines with more than 24 months. As to the categories of herds, no important differences were recorded.

**Table 12.** Origin of the bovines sampled, according to sub-population and age group

Sub-populations	Age groups considered in the study								
	6 to 12 m			13 to 24 m			> 24 m		
	Born in the properties	Total		Born in the properties	Total		Born in the properties	Total	
Acre and two municipalities of the State of Amazonas	326	92%	356	258	88%	292	151	76%	200
Bahia	300	72%	418	247	72%	343	147	64%	228
Federal District	623	77%	814	435	68%	644	223	48%	465
Espírito Santo	340	96%	355	267	93%	287	137	70%	196
Goiás	303	88%	345	226	79%	286	136	70%	195
Mato Grosso	347	95%	366	264	88%	301	149	72%	207
Mato Grosso do Sul (Pantanal, May)	351	97%	361	264	89%	297	148	73%	202
Mato Grosso do Sul (Pantanal, November)	375	94%	399	302	92%	330	189	84%	226
Mato Grosso do Sul (Plateau)	383	92%	417	298	88%	340	168	73%	230
Minas Gerais (Centre-West Circuit)	700	83%	840	510	74%	690	280	60%	470
Minas Gerais (East Circuit)	364	89%	411	276	82%	337	163	71%	230
Paraná	335	84%	400	267	81%	331	170	75%	226
Rio de Janeiro	706	86%	822	470	69%	680	238	52%	461
Rio Grande do Sul	337	98%	343	278	99%	282	185	97%	191
Rondônia	320	93%	343	247	88%	282	115	59%	195
São Paulo	307	74%	414	224	64%	351	118	50%	236
Sergipe	503	61%	831	370	56%	666	266	54%	490
Tocantins	316	96%	330	251	90%	278	165	85%	193
<b>Total</b>	<b>7.236</b>	<b>84%</b>	<b>8.565</b>	<b>5.454</b>	<b>78%</b>	<b>7.017</b>	<b>3.148</b>	<b>65%</b>	<b>4.841</b>

**Table 13.** Origin of the bovines sampled, according to sub-population and size of herd

Sub-populations	Categories of herds considered in the study								
	Up to 20 bovines			21 to 50 bovines			More than 50 bovines		
	Born in the properties	Total		Born in the properties	Total		Born in the properties	Total	
Acre and two municipalities of the State of Amazonas	40	77%	52	71	93%	76	624	87%	720
Bahia	143	73%	195	134	82%	164	417	66%	630
Federal District	171	53%	322	209	60%	350	901	72%	1.251
Espírito Santo	64	76%	84	80	75%	107	600	93%	647
Goiás	16	67%	24	43	83%	52	606	81%	750
Minas Gerais (Centre-West Circuit)	109	69%	159	207	74%	280	1.174	75%	1.561
Minas Gerais (East Circuit)	94	81%	116	119	77%	154	590	83%	708
Mato Grosso do Sul (Pantanal, May)	11	92%	12	10	83%	12	742	89%	836
Mato Grosso do Sul (Pantanal, November)	8	89%	9	7	100%	7	851	91%	939
Mato Grosso do Sul (Plateau)	13	87%	15	14	70%	20	822	86%	952
Mato Grosso	49	89%	55	26	84%	31	685	87%	788
Paraná	95	75%	127	101	80%	126	576	82%	704
Rio de Janeiro	123	84%	146	274	83%	329	1.017	68%	1.488
Rondônia	22	61%	36	59	75%	79	601	85%	705
Rio Grande do Sul	129	93%	138	108	95%	114	563	100%	564
Sergipe	196	65%	301	185	44%	419	758	60%	1.267
São Paulo	46	61%	76	83	75%	110	520	64%	815
Tocantins	20	83%	24	54	98%	55	658	91%	722
<b>Total</b>	<b>1.349</b>	<b>71%</b>	<b>1.891</b>	<b>1.784</b>	<b>72%</b>	<b>2.485</b>	<b>12.705</b>	<b>79%</b>	<b>16.047</b>

Specifically for the animals declared as non-vaccinated by the persons interviewed in the rural properties, Table 14 presents additional information related to the origin of the animals. One notices that, considering all sub-populations, 77% of the animals reported as non-vaccinated were acquired in other rural properties. As it has been previously informed, per force of the sanitary standards in force, animals must have at least one vaccination against foot and mouth disease before any movement. Therefore, it is likely that the person interviewed answered that the animals had not been vaccinated by them, which does not exclude the possibility that said animals had been vaccinated in the properties of origin. Considering this possibility, the participation of non-vaccinated animals in the sample would be of over 9% for circa 2%, with the highest concentrations recorded in the sub-populations of Rio de Janeiro (7%), of Rio Grande do Sul (6%), of Bahia, East Circuit of Minas Gerais and Plateau of Mato Grosso do Sul (4%).

**Table 14.** Origin of the animals with histories of non-vaccination, according to the sub-populations considered in the study

Sub-population	Native		Non-native		Total
Acre and two municipalities of the State of Amazonas	0	0%	28	100%	28
Bahia	40	23%	134	77%	174
Federal District	4	29%	10	71%	14
Espírito Santo	6	38%	10	63%	16
Goiás	0	0%	6	100%	6
Minas Gerais (Centre-West Circuit)	8	10%	73	90%	81
Minas Gerais (East Circuit)	40	71%	16	29%	56
Mato Grosso do Sul (Pantanal, May)	0	0%	18	100%	18
Mato Grosso do Sul (Pantanal, November)	0	0%	14	100%	14
Mato Grosso do Sul (Plateau)	40	66%	21	34%	61
Mato Grosso	16	84%	3	16%	19
Paraná	0	0%	15	100%	15
Rio de Janeiro	140	36%	252	64%	392
Rondônia	0	0%	10	100%	10
Rio Grande do Sul	47	85%	8	15%	55
Sergipe	61	9%	635	91%	696
São Paulo	1	1%	153	99%	154
Tocantins	25	66%	13	34%	38
<b>Total</b>	<b>428</b>	<b>23%</b>	<b>1.419</b>	<b>77%</b>	<b>1.847</b>

#### **4.4. Period of collection and interval between collection and the date of vaccination**

Finally, among the variables considered in the study that may directly or indirectly influence the levels of population immunity for foot and mouth disease, we present information on the period of collection and the time between collection and the date of the last vaccination, according to sub-populations (Table 15).

The period of collection varied between five days in Tocantins and 80 days in Rio de Janeiro, an average of 17 days. In addition to Rio de Janeiro, in the sub-populations represented by Rio Grande do Sul and by the East Livestock Circuit of Minas Gerais, the time of collection surpassed by far the period of 30 days. In the other sub-populations, the period of collection varied from 12 to 34 days.

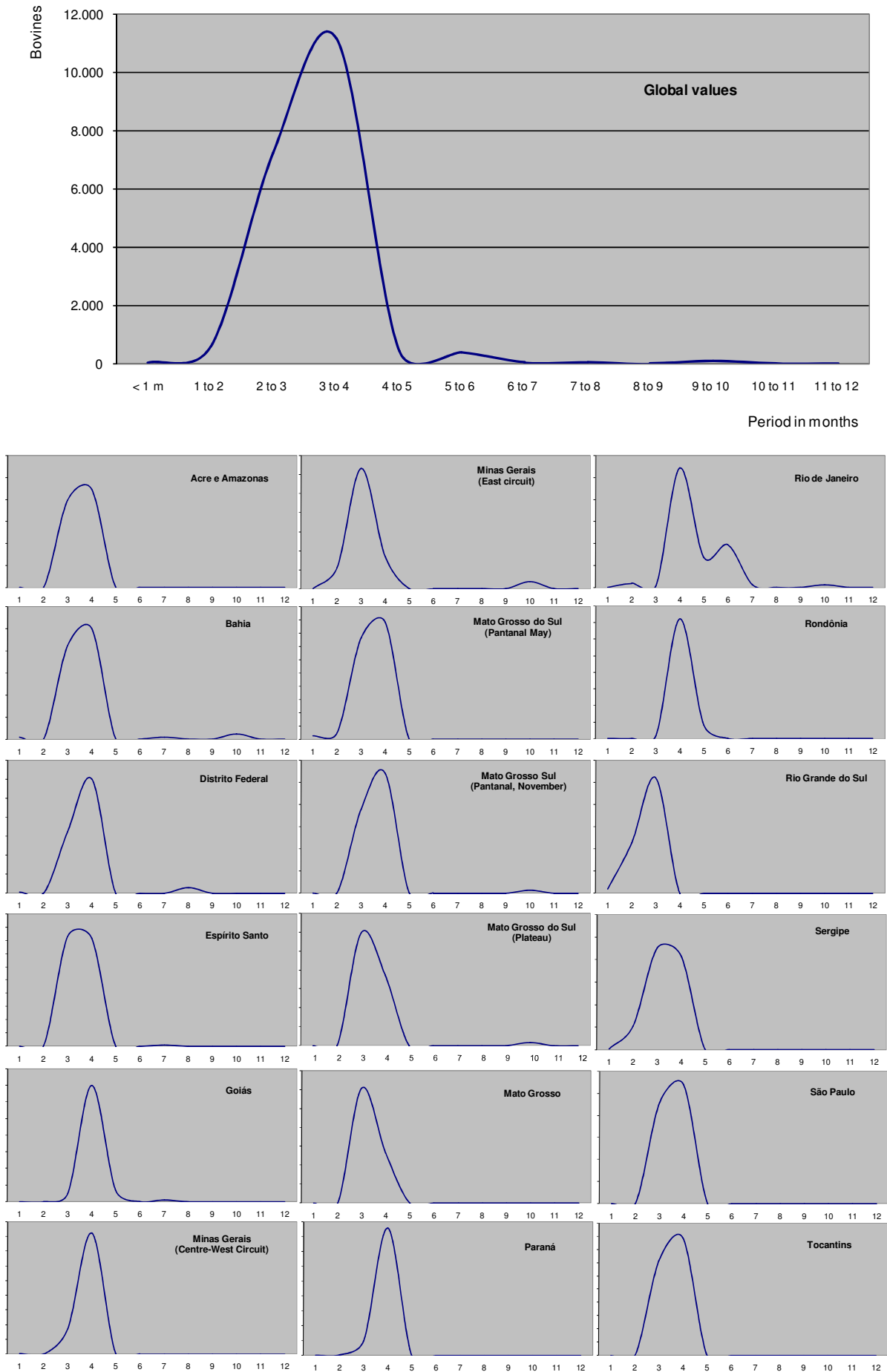
The period of collection of the samples was forecast to be carried out between 30 and 90 days after completion of the last stage of vaccination against foot and mouth disease. In Table 15 the information on the time interval between collection and the last vaccination was defined considering the date of vaccination reported by rural producers, which may vary between the first and the last day of the stage that, in most cases, is of 30 days. Thus, the average and mean values recorded are within the forecast in all sub-populations. However, in some of them one notices much dispersion in the average, highlighting the sub-populations represented by the East Circuit of Minas Gerais, the region of the Plateau of Mato Grosso do Sul, Bahia and Rio de Janeiro that featured a coefficient of variation equal to or higher than 40%.

Picture 11 shows graphic representations of the distribution of the bovines sampled according to the intervals of time between the date of vaccination and the collection, for the study in general and according to sub-population. The total of animals sampled, according to the intervals of time considered, is presented in Table 16. In global terms, 92% of the samples were collected in the period of 2 to 4 months of the last stage of vaccination. Out of the other samples, 56 (0.3%) were collected in an interval shorter than 30 days; 615 (3.1%) between 30 and 60 days, and 1.037 (5.2%) in an interval between 4 and 11 months. It is worth highlighting that, for 313 animals sampled (1.5% of the total) there is no information on the last date of vaccination or the interval between collection and vaccination surpassed 12 months, as it can be assessed in Table 17. The highest frequencies concerning the number of animals for which it was impossible to determine the time interval between the date of vaccination and the collection were recorded in the sub-populations identified as Rio de Janeiro (6.3% vis-à-vis the total of the sub-population), Rio Grande do Sul (5.8%), East Circuit of Minas Gerais (2.8%) and Bahia (2.5%).

**Table 15.** Information on the period of collection of the samples and interval between collection and the date of the last vaccination.

UF and regions (sub-populations)	Collection of samples			Days After last vaccination					
	Beginning	Completion	Days	Average	Mean	Min	Max	DP	CV
Acre and two municipalities of the State of Amazonas	12-Aug-05	31-Aug-05	19	96	93	63	283	33	35%
Bahia	20-Dec-05	2-Jan-06	13	104	92	28	288	50	48%
Federal District	17-Aug-05	29-Aug-05	12	98	95	7	233	32	32%
Espírito Santo	6-Dec-05	3-Jan-06	28	94	91	37	197	23	25%
Goiás	24-Feb-06	9-Mar-06	13	111	107	89	201	19	17%
Mato Grosso	15-Aug-05	5-Sep-05	21	91	88	68	112	12	13%
Mato Grosso do Sul (Pantanal, May)	17-Aug-05	30-Aug-05	13	82	93	4	119	22	26%
Mato Grosso do Sul (Pantanal, November)	14-Feb-06	17-Mar-06	31	95	97	31	293	34	36%
Mato Grosso do Sul (Plateau)	17-Aug-05	30-Aug-05	13	108	88	64	285	54	50%
Minas Gerais (Centre-West Circuit)	19-Aug-05	22-Sep-05	34	98	104	19	123	18	18%
Minas Gerais (East Circuit)	2-Dec-05	26-Jan-06	55	85	79	36	300	48	56%
Paraná	15-Aug-05	30-Aug-05	15	97	101	29	118	16	16%
Rio de Janeiro	3-Jan-06	24-Mar-06	80	128	116	34	314	52	40%
Rio Grande do Sul	10-Feb-06	24-Mar-06	42	52	65	12	79	17	33%
Rondônia	15-Aug-05	31-Aug-05	16	111	109	88	131	11	10%
São Paulo	12-Aug-05	24-Aug-05	12	91	92	60	111	11	12%
Sergipe	15-Aug-05	1-Sep-05	17	86	88	13	115	21	24%
Tocantins	22-Aug-05	27-Aug-05	5	97	93	81	116	10	10%

Min = minimum value; Max = maximum value; DP = standard deviation; CV = coefficient of variation



Picture 11. Graphic representation of the bovines sampled according to intervals between collection and the date of the last vaccination

**Table 16.** Total of bovines sampled according to sub-population and the time interval between collection and vaccination

Sub-populations	Interval between collection and the last vaccination (in months)											Total
	< 1	1 a 2	2 a 3	3 a 4	4 a 5	5 a 6	6 a 7	7 a 8	8 a 9	9 a 10	10 a 11	
Acre and two municipalities of the State of AM	0	0	398	446	0	0	0	0	0	1	0	845
Bahia	8	0	425	501	0	0	8	0	0	22	0	964
Federal District	8	0	649	1.207	0	0	0	59	0	0	0	1.923
Espírito Santo	0	2	414	407	0	0	4	0	0	0	0	827
Goiás	0	0	47	701	68	0	10	0	0	0	0	826
Mato Grosso	0	0	608	262	0	0	0	0	0	0	0	870
Mato Grosso do Sul (Pantanal, May)	12	23	383	442	0	0	0	0	0	0	0	860
Mato Grosso do Sul (Pantanal, November)	0	2	380	543	1	0	0	0	0	14	0	940
Mato Grosso do Sul (Plateau)	0	0	594	372	0	0	1	0	1	15	0	983
Minas Gerais (Centre-West Circuit)	3	0	343	1.645	3	0	0	0	0	0	0	1.994
Minas Gerais (East Circuit)	0	111	631	170	2	0	0	0	0	37	0	951
Paraná	0	2	98	857	0	0	0	0	0	0	0	957
Rio de Janeiro	0	37	10	1.084	272	386	24	0	0	25	2	1.840
Rio Grande do Sul	20	228	521	0	0	0	0	0	0	0	0	769
Rondônia	0	0	15	723	82	0	0	0	0	0	0	820
São Paulo	0	2	456	537	0	0	0	0	0	0	0	995
Sergipe	5	208	901	831	0	0	0	0	0	0	0	1.945
Tocantins	0	0	358	443	0	0	0	0	0	0	0	801
<b>Total</b>	<b>56</b>	<b>615</b>	<b>7.231</b>	<b>11.171</b>	<b>428</b>	<b>386</b>	<b>47</b>	<b>59</b>	<b>1</b>	<b>114</b>	<b>2</b>	<b>20.110</b>

**Table 17.** Total of bovines sampled without information on the date of vaccination or with time interval between collection and vaccination longer than 12 months

Sub-populations	Samples		
	Total	Irregular*	
Acre and two municipalities of the State of Amazonas	848	3	0.4%
Bahia	989	25	2.5%
Federal District	1.923	0	0.0%
Espírito Santo	838	11	1.3%
Goiás	826	0	0.0%
Mato Grosso	874	4	0.5%
Mato Grosso do Sul (Pantanal, May)	860	0	0.0%
Mato Grosso do Sul (Pantanal, November)	955	15	1.6%
Mato Grosso do Sul (Plateau)	987	4	0.4%
Minas Gerais (Centre-West Circuit)	2.000	6	0.3%
Minas Gerais (East Circuit)	978	27	2.8%
Paraná	957	0	0.0%
Rio de Janeiro	1.963	123	6.3%
Rio Grande do Sul	816	47	5.8%
Rondônia	820	0	0.0%
São Paulo	1.001	6	0.6%
Sergipe	1.987	42	2.1%
Tocantins	801	0	0.0%
<b>Total</b>	<b>20.423</b>	<b>313</b>	<b>1.5%</b>

\* Without date of recording of the last vaccination or with more than 12 months between collection and the date of recording of the last vaccination.

## 5. Results and discussion

Considering the objective of assessing the levels of population immunity for foot and mouth disease, we decided to present the results related to the sub-populations, to the schemes of vaccination, to the history of vaccination, to the size of the herd and to the origin of the animals (native or not) always according to the type of virus and to the age groups. However, since the database is available at the DSA/SDA, it's possible, according to specific interests, that supplementary analyses are carried out.

### According to the type of virus, age groups and sub-populations

Table 18 presents the percentages of bovines protected for each sub-population and Tables 19 to 21 the results per age groups.

Table 18, shows that the lower limit of the proportion of bovines protected was of 98% for the virus "C", 87% for the virus "O" and 68% for the virus "A". Notice that for virus "A" the lower value was restricted to the subpopulation from East Circuit of Minas Gerais. For the other subpopulations the value was 98%, similar to the results of virus type C.

The joint analysis of the age groups shows that, out of the sub-populations assessed, in 14 (74%) true prevalence of immunised bovines of 99% was recorded, in three the true prevalence was between 90% and 92%, and only in the sub-population represented by the East Circuit of Minas Gerais the prevalence was under 80%, with an interval of confidence of 68% to 75%.

The results, according to the age groups, indicated for the sub-populations challenged with virus "O" that the lower limits of the intervals of 95% of confidence for the proportion of bovines protected in the age group of 6 to 12 months were equal to or higher than 90% for the sub-populations identified as Rondônia, Tocantins, Mato Grosso and Goiás; 82% for Acre and Amazonas; and under 80% for the subpopulations from Rio Grande do Sul, Mato Grosso do Sul (Pantanal, May) and Espírito Santo. These last three subpopulations featured limits higher than 95% of confidence between 78% and 84%. The subpopulation of Espírito Santo featured the lowest limit, 64%, equal to the value assumed for the proportion of protected bovines for the age group of bovines at issue (between 6 and 12 months). For virus "A", the only subpopulation with true prevalence under 80% was represented by Minas Gerais Centre-West Circuit (56%), for the others the limits were between 86% and 99%. Considering virus "C", the population from Minas Gerais Centre-West Circuit also featured the lowest limit for true prevalence (74%). The other populations tested for this type featured limits ranging from 88% to 99%.

As it was expected, the age group where the lowest prevalence of immunised bovines was recorded was represented by the animals between 6 and 12 months of age. In spite of the expectation of low values for this group (65%), in more than half the sub-populations assessed (72%) prevalence higher than 85% was recorded, a rate expected for the animals between 13 and 24 months of age, and in eight (44%) the prevalence was higher than 95%. Out of the sub-populations with the lowest rates of immunity for populations of bovines with ages between 6 and 12 months, only in the East Circuit of Minas Gerais the levels of protection were below the estimated value of 65%.

For the age group of bovines between 13 and 24 months, 17 sub-populations (94%) featured prevalence higher than 95%, with a lower limit of confidence of 93%. The lesser value was also recorded in the East Circuit of Minas Gerais, with intervals of confidence varying from 72% to 83%. Finally, for the age group of bovines with more than 24 months, in the sub-population of the East Circuit of Minas Gerais true prevalence of 89% was recorded and, in the other sub-populations, 99%.

The analysis per sub-population shows, for nearly all the units of the Federation, an excellent immune coverage of the bovine population, irrespective of the age group considered. The values obtained surpassed by far the initial expectations of the study, with the only exception of the East Circuit of Minas Gerais, where, clearly, one notices an immune coverage lower than that of the other sub-populations considered. In the East Circuit of Minas Gerais, the higher limit of the interval of confidence is of 95%; considering the age groups together, it is of 75%, a percentage lower than what is expected in eradication programmes, highlighting the fact that the problem is concentrated in younger animals.

Comparison between the percentages of record of vaccination obtained in the stages immediately prior to the periods of sample collection and the prevalence obtained in the study, for all bovines sampled, may be done by means of Table 22. In eight units of the Federation (44% of the total) the percentages of records of vaccination dropped within the intervals of confidence at 95% obtained in the study and in another seven (39% of the total), these percentages were smaller than the lower limit. In only three sub-populations the percentages of records of vaccination were bigger than the higher limit of the interval of confidence at 95%: East Circuit of Minas Gerais, Espírito Santo and the region of the Pantanal, May, in Mato Grosso do Sul.

**Table 18.** Laboratory results, according to type of virus and sub-population

Type of virus	Sub-populations	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Acre and Amazonas	703	848	83%	99%	99%	100%
	Goiás	735	826	89%	99%	98%	100%
	Rondônia	746	820	91%	99%	98%	100%
	Tocantins	736	801	92%	99%	98%	100%
	Espírito Santo	639	838	76%	90%	87%	92%
	Rio Grande do Sul	676	816	83%	98%	97%	100%
	Mato Grosso	808	874	92%	99%	98%	100%
	Mato Grosso do Sul (Pantanal, May)	672	860	78%	92%	90%	95%
O	Bahia	826	989	84%	99%	98%	100%
	Paraná	861	957	90%	99%	98%	100%
	São Paulo	838	1.001	84%	99%	98%	100%
	Minas Gerais (East Circuit)	621	978	63%	71%	68%	75%
	Mato Grosso do Sul (Plateau)	864	987	88%	99%	98%	100%
	Mato Grosso do Sul (Pantanal, November)	786	955	82%	99%	98%	99%
C	Federal District	1.314	1.923	68%	99%	98%	100%
	Rio de Janeiro	1.405	1.963	72%	99%	98%	100%
	Sergipe	1.352	1.987	68%	99%	98%	100%
	Minas Gerais (Centre-West Circuit)	1.179	2.000	59%	99%	98%	100%

**Table 19.** Laboratory results for bovines of 6 to 12 months, according to type of virus and sub-population

Type of virus	Sub-populations	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Acre and Amazonas	264	356	74%	87%	82%	91%
	Goiás	279	345	81%	96%	94%	99%
	Rondônia	287	343	84%	99%	98%	100%
	Tocantins	283	330	86%	99%	98%	100%
	Espírito Santo	227	355	64%	72%	66%	78%
	Rio Grande do Sul	236	343	69%	79%	74%	85%
	Mato Grosso	327	366	89%	99%	98%	100%
	Mato Grosso do Sul (Pantanal, May)	241	361	67%	76%	71%	81%
O	Bahia	316	418	76%	89%	85%	92%
	Paraná	342	400	86%	99%	98%	100%
	São Paulo	304	414	73%	86%	82%	90%
	Minas Gerais (East Circuit)	219	411	53%	56%	51%	62%
	Mato Grosso do Sul (Plateau)	334	417	80%	95%	93%	98%
	Mato Grosso do Sul (Pantanal, November)	294	399	74%	86%	82%	90%
C	Federal District	421	814	52%	88%	85%	91%
	Rio de Janeiro	468	822	57%	99%	98%	100%
	Sergipe	477	831	57%	99%	98%	100%
	Minas Gerais (Centre-West Circuit)	384	840	46%	74%	69%	78%

**Table 20.** Laboratory results for bovines of 13 to 24 months, according to type of virus and sub-population

Type of virus	Sub-populations	Samples		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Acre and Amazonas	248	292	85%	98%	96%	100%
	Goiás	264	286	92%	99%	98%	100%
	Rondônia	264	282	94%	99%	98%	100%
	Tocantins	262	278	94%	99%	98%	100%
	Espírito Santo	231	287	80%	96%	93%	99%
	Rio Grande do Sul	253	282	90%	99%	97%	100%
	Mato Grosso	283	301	94%	98%	97%	100%
	Mato Grosso do Sul (Pantanal, May)	244	297	82%	99%	98%	100%
O	Bahia	299	343	87%	99%	98%	100%
	Paraná	303	331	92%	99%	98%	100%
	São Paulo	308	351	88%	99%	98%	100%
	Minas Gerais (East Circuit)	228	337	68%	77%	72%	83%
	Mato Grosso do Sul (Plateau)	309	340	91%	99%	98%	100%
	Mato Grosso do Sul (Pantanal, November)	282	330	85%	99%	98%	100%
C	Federal District	465	644	72%	99%	98%	100%
	Rio de Janeiro	523	680	77%	99%	98%	100%
	Sergipe	465	666	70%	99%	98%	100%
	Minas Gerais (Centre-West Circuit)	436	690	63%	99%	98%	100%

**Table 21.** Laboratory results for bovines with more than 24 months, according to type of virus and sub-population

Type of virus	Sub-populations	Samples		Prevalence		Interval of confidence	
		Protected	Total	Apparent	True	Lower	Higher
A	Acre and Amazonas	191	200	96%	99%	97%	100%
	Goiás	192	195	98%	99%	97%	100%
	Rondônia	195	195	100%	99%	98%	100%
	Tocantins	191	193	99%	99%	98%	100%
	Espírito Santo	181	196	92%	99%	97%	100%
	Rio Grande do Sul	187	191	98%	99%	97%	100%
	Mato Grosso	198	207	96%	99%	98%	100%
	Mato Grosso do Sul (Pantanal, May)	187	202	93%	99%	97%	100%
O	Bahia	211	228	93%	99%	97%	100%
	Paraná	216	226	96%	99%	97%	100%
	São Paulo	226	236	96%	99%	98%	100%
	Minas Gerais (East Circuit)	174	230	76%	89%	84%	95%
	Mato Grosso do Sul (Plateau)	221	230	96%	99%	97%	100%
	Mato Grosso do Sul (Pantanal, November)	210	226	93%	99%	97%	100%
C	Federal District	428	465	92%	99%	98%	100%
	Rio de Janeiro	414	461	90%	99%	98%	100%
	Sergipe	410	490	84%	99%	98%	100%
	Minas Gerais (Centre-West Circuit)	359	470	76%	99%	98%	100%

**Table 22.** Comparison between the percentages of recording of the stage prior to vaccination and the prevalence obtained in the study

Sub-populations	% of record of vaccination*	Prevalence		Interval of confidence	
		Apparent	Adjusted	Lower	Higher
Acre and Amazonas	92.00%	83%	99%	97%	100%
Bahia	95.00%	84%	99%	98%	100%
Federal District	92.70%	68%	99%	98%	100%
Espírito Santo	98.25%	76%	90%	87%	92%
Goiás	98.48%	89%	99%	98%	100%
Mato Grosso	98.51%	92%	99%	98%	100%
Mato Grosso do Sul (Pantanal, May)	97.65%	78%	92%	90%	95%
Mato Grosso do Sul (Pantanal, November)	98.84%	82%	99%	97%	99%
Mato Grosso do Sul (Plateau)	99.48%	88%	99%	98%	100%
Minas Gerais (Centre-West Circuit)	96.31%	59%	99%	98%	100%
Minas Gerais (East Circuit)	95.85%	63%	71%	68%	75%
Paraná	98.72%	90%	99%	98%	100%
Rio de Janeiro	92.13%	72%	99%	98%	100%
Rio Grande do Sul	90.70%	83%	98%	97%	100%
Rondônia	99.97%	91%	99%	98%	100%
São Paulo	99.41%	84%	99%	98%	100%
Sergipe	93.08%	68%	99%	98%	100%
Tocantins	98.45%	92%	99%	98%	100%

\* stage immediately anterior to the collection of the samples



According to the type of virus, age groups and schemes of vaccination

The study carried out was not designed in a way that allows for detailed comparisons between the schemes of vaccination used in the country, bearing in mind the fact that the strategy of collection of the samples only considered the period expected of higher immunity response of the bovine population. However, for an initial assessment, results grouped per scheme of vaccination are made available in Tables 23 to 26. In the first table the results concern the total of bovines sampled and in the others they concern the age groups considered in the study.

More important differences were only recorded in the age group of 6 to 12 months, and it was noticed that schemes 1 (every six months) and 3 (every six months for animals under the age of 24 months and annual animals for older than 24 months, with reinforcement for animals under the age of 12 months) allowed for the highest levels of population immunity. Specifically for Scheme 2 (every six months for animals under the age of 24 months and annual for animals older than 24 months), results recorded in the regions where the virus tested was of the type O, were lower than those recorded in the regions assessed with virus of the types A and C, which shows that the difference was influenced by the low results obtained in the East Circuit of Minas Gerais.

**Table 23.** Laboratory results for the total of bovines sampled, according to type of virus and schemes of vaccination

Type of virus	Schemes of vaccination against foot and mouth disease	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	1. Every six months	2.920	3.295	89%	99%	99%	100%
	2. Every six months for <24 months and annual for > 24 months	1.315	1.654	80%	94%	93%	96%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	808	874	92%	99%	98%	100%
	4. Annual	672	860	78%	92%	90%	95%
O	1. Every six months	2.525	2.947	86%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	621	978	63%	71%	67%	75%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	864	987	88%	99%	98%	100%
	4. Annual	786	955	82%	99%	97%	100%
C	1. Every six months	4.071	5.873	69%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	1.179	2.000	59%	99%	98%	100%

**Table 24.** Laboratory results for bovines with ages between 6 and 12 months, according to type of virus and schemes of vaccination

Type of virus	Schemes of vaccination against foot and mouth disease	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	1. Every six months	1.113	1.374	81%	97%	95%	98%
	2. Every six months for <24 months and annual for > 24 months	463	698	66%	75%	71%	80%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	327	366	89%	99%	98%	100%
	4. Annual	241	361	67%	76%	70%	82%
O	1. Every six months	962	1.232	78%	92%	90%	95%
	2. Every six months for <24 months and annual for > 24 months	219	411	53%	56%	50%	63%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	334	417	80%	95%	92%	98%
	4. Annual	294	399	74%	86%	81%	91%
C	1. Every six months	1.366	2.467	55%	96%	95%	98%
	2. Every six months for <24 months and annual for > 24 months	384	840	46%	74%	67%	81%

**Table 25.** Laboratory results for bovines between 13 and 24 months of age, according to type of virus and schemes of vaccination

Type of virus	Schemes of vaccination against foot and mouth disease	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	1. Every six months	1.038	1.138	91%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	484	569	85%	99%	98%	100%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	283	301	94%	99%	98%	100%
	4. Annual	244	297	82%	98%	96%	100%
O	1. Every six months	910	1.025	89%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	228	337	68%	77%	71%	84%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	309	340	91%	99%	98%	100%
	4. Annual	282	330	85%	98%	96%	100%
C	1. Every six months	1.453	1.990	73%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	436	690	63%	99%	97%	100%

**Table 26.** Laboratory results for bovines with ages above 24 months, according to type of virus and schemes of vaccination

Type of virus	Schemes of vaccination against foot and mouth disease	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	1. Every six months	769	783	98%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	368	387	95%	99%	98%	100%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	198	207	96%	99%	98%	100%
	4. Annual	187	202	93%	99%	97%	100%
O	1. Every six months	653	690	95%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	174	230	76%	89%	83%	95%
	3. Every six months for <24 months and annual for > 24 months, + reinforcement for < 12 months	221	230	96%	99%	98%	100%
	4. Annual	210	226	93%	99%	97%	100%
C	1. Every six months	1.252	1.416	88%	99%	98%	100%
	2. Every six months for <24 months and annual for > 24 months	359	470	76%	99%	98%	100%

#### According to the type of virus, age groups and history of vaccination

During the activity of collection of the samples the persons in charge of the animals were asked what the estimate was for the number of vaccinations inoculated in each of the bovines sampled. The results are presented in Tables 27 to 29, respectively for the age groups of 6 to 12 months, 13 to 24 months and more than 24 months. The information on the history of vaccination was grouped in four categories: without vaccination or without information; one vaccination; two vaccinations; and more than two vaccinations.

The immunisation levels of the age groups of the bovines between 13 and 24 months as well as of the bovines older than 24 months presented no variation according to the number of doses declared by the persons in charge of the animals, as it can be assessed in Picture 12.

Bovines between 6 and 12 months, in turn, featured some immunity variation according to the number of vaccinations declared, even because the information is more recent, which probably makes it more accurate. Bovines with two or more vaccinations, irrespective of the type of virus assessed, always featured rates of population immunity higher than those of other categories.

For the category of animals without vaccination, an important number of bovines with immunity protection were observed, indicating the inaccuracy of the information both vis-à-vis the history of vaccination and the age. In the latter case, when it is about the age group of 6 to 12 months, there may have been interference of colostrum, particularly in animals with ages close to six months, born from cows with histories of many vaccinations.

**Table 27.** Laboratory results for bovines with ages between 6 and 12 months, according to type of virus and history of vaccination

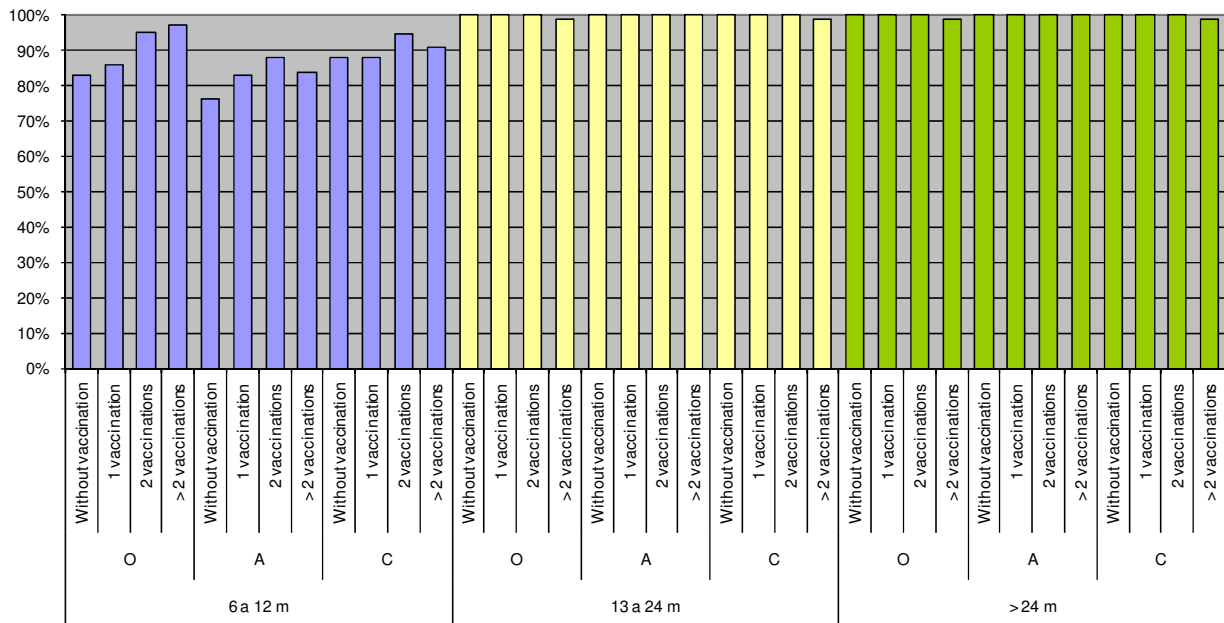
Type of Virus	History reported of nº of vaccinations	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Without vaccination / information	60	83	72%	84%	73%	95%
	One vaccination	1.126	1.529	74%	86%	83%	88%
	Two vaccinations	848	1.061	80%	95%	93%	97%
	More than two vaccinations	110	126	87%	97%	93%	100%
O	Without vaccination / information	154	229	67%	77%	69%	85%
	One vaccination	819	1.140	72%	83%	80%	86%
	Two vaccinations	719	945	76%	90%	87%	92%
	More than two vaccinations	117	145	81%	96%	92%	100%
C	Without vaccination / information	253	488	52%	88%	82%	95%
	One vaccination	880	1.698	52%	88%	85%	92%
	Two vaccinations	593	1.086	55%	95%	91%	98%
	More than two vaccinations	24	35	69%	91%	69%	100%

**Table 28.** Laboratory results for bovines with ages between 13 and 24 months, according to type of virus and history of vaccination

Type of Virus	History reported of nº of vaccinations	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Without vaccination / information	62	68	91%	99%	99%	100%
	One vaccination	95	111	86%	99%	99%	100%
	Two vaccinations	627	740	85%	99%	99%	100%
	More than two vaccinations	1.265	1.386	91%	99%	99%	100%
O	Without vaccination / information	130	156	83%	99%	99%	100%
	One vaccination	138	164	84%	99%	99%	100%
	Two vaccinations	494	622	79%	99%	99%	100%
	More than two vaccinations	967	1.090	89%	99%	99%	100%
C	Without vaccination / information	271	428	63%	99%	99%	100%
	One vaccination	93	136	68%	99%	99%	100%
	Two vaccinations	433	640	68%	99%	99%	100%
	More than two vaccinations	1.092	1.476	74%	99%	99%	100%

**Table 29.** Laboratory results for bovines with ages above 24 months, according to type of virus and history of vaccination

Type of Virus	History reported of nº of vaccinations	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Without vaccination / information	36	39	92%	99%	99%	100%
	One vaccination	35	38	92%	99%	99%	100%
	Two vaccinations	86	89	97%	99%	99%	100%
	More than two vaccinations	1.365	1.413	97%	99%	99%	100%
O	Without vaccination / information	81	89	91%	99%	99%	100%
	One vaccination	53	61	87%	99%	99%	100%
	Two vaccinations	97	106	92%	99%	99%	100%
	More than two vaccinations	1.027	1.120	92%	99%	99%	100%
C	Without vaccination / information	194	267	73%	99%	99%	100%
	One vaccination	58	73	79%	99%	99%	100%
	Two vaccinations	124	135	92%	99%	99%	100%
	More than two vaccinations	1.235	1.411	88%	99%	99%	100%



**Picture 12.** Graphic representation of the immunisation levels according to number of vaccinations, type of virus and age group

#### According to the type of virus, age groups and size of the herd

In Tables 30 to 33 we present the results by size of herd. The joint analysis of the age groups (Table 30) indicates an immune coverage comparatively lower for the category up to 20 bovines in the regions where were assessed the types of virus A and O, which was not observed for the virus C. For the category of 21 to 50 bovines the lowest true prevalence (87%) was recorded only in the region assessed for the type of virus O. For the category above 50 bovines no differences were recorded among the types of virus assessed, showing an immune coverage of 99%.

The biggest differences among categories of herd, similarly to the other variables assessed, were recorded in the age group of 6 to 12 months (Table 31). For this age group, in the category of herds with more than 50 bovines, the immune coverage always remained equal to or higher than 90%, irrespective of the type of virus assessed. In the category up to 20 bovines the lowest rates of population immunity were recorded, irrespective of the type of virus assessed, and a true prevalence of 60% was found for the virus O; 64% for the virus C and 72% for the virus A. In the category between 21 and 50 bovines the results showed little variation among the types of virus assessed, having remained between 73 and 76%.

For the age group above 24 months of age (Table 32), no important differences were recorded, and there was an immune coverage above 94%, irrespective of the type of virus and of the category of herd.

In spite of the variations recorded among the types of virus assessed, one notices a trend towards greater stability, with higher levels of immunity, for the category of herds with more than 50 bovines. On the other hand, for the other categories, particularly when one assessed bovines with ages between 6 and 12 months, the lowest prevalence of protection was recorded. These results corresponded to the forecasts of the study, considering that the owners of herds with more than 50 bovines have greater interest and better conditions to carry out the vaccination against foot and mouth disease.

**Table 30.** Results for all bovines, according to the size of the herds and type of virus

Type of virus	Size of the herds	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Up to 20 bovines	316	425	74%	87%	82%	100%
	21 to 50 bovines	425	526	81%	96%	94%	100%
	More than 50 bovines	4.974	5.732	87%	99%	99%	100%
O	Up to 20 bovines	359	538	67%	76%	71%	81%
	21 to 50 bovines	430	581	74%	87%	82%	100%
	More than 50 bovines	4.007	4.748	84%	99%	99%	100%
C	Up to 20 bovines	527	928	57%	99%	98%	100%
	21 to 50 bovines	863	1.378	63%	99%	98%	100%
	More than 50 bovines	3.860	5.567	69%	99%	98%	100%

**Table 31.** Results for bovines of 6 to 12 months of age, according to the size of the herds and type of virus

Type of virus	Size of the herds	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Up to 20 bovines	115	180	64%	72%	62%	81%
	21 to 50 bovines	146	220	66%	75%	67%	83%
	More than 50 bovines	1.883	2.399	78%	93%	92%	94%
O	Up to 20 bovines	127	229	55%	60%	50%	69%
	21 to 50 bovines	157	243	65%	73%	65%	81%
	More than 50 bovines	1.525	1.987	77%	90%	88%	92%
C	Up to 20 bovines	159	385	41%	64%	53%	75%
	21 to 50 bovines	267	572	47%	76%	68%	84%
	More than 50 bovines	1.324	2.350	56%	99%	98%	100%

**Table 32.** Results for bovines of 13 to 24 months, according to the size of the herds and type of virus

Type of virus	Size of the herds	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Up to 20 bovines	107	143	75%	88%	80%	96%
	21 to 50 bovines	160	181	88%	98%	96%	100%
	More than 50 bovines	1.782	1.981	90%	99%	98%	100%
O	Up to 20 bovines	131	184	71%	82%	74%	90%
	21 to 50 bovines	155	203	76%	89%	83%	96%
	More than 50 bovines	1.443	1.645	88%	99%	98%	100%
C	Up to 20 bovines	182	325	56%	94%	88%	100%
	21 to 50 bovines	314	472	67%	99%	97%	100%
	More than 50 bovines	1.393	1.883	74%	99%	98%	100%

**Table 33.** Results for bovines with more than 24 months of age, according to the size of the herds and type of virus

Type of virus	Size of the herds	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
A	Up to 20 bovines	94	102	92%	98%	94%	100%
	21 to 50 bovines	119	125	95%	99%	96%	100%
	More than 50 bovines	1.309	1.352	97%	99%	98%	100%
O	Up to 20 bovines	101	125	81%	94%	88%	100%
	21 to 50 bovines	118	135	87%	98%	94%	100%
	More than 50 bovines	1.039	1.116	93%	99%	98%	100%
C	Up to 20 bovines	186	218	85%	99%	96%	100%
	21 to 50 bovines	282	334	84%	99%	97%	100%
	More than 50 bovines	1.143	1.334	86%	99%	98%	100%

According to the type of virus, age groups and origin of the animals

Results are presented in Tables 34 a 36, according to age group considered. Only for bovines between 6 to 12 months of age differences for population immunity between native and non-native animals were recorded (Table 34). In this age group, irrespective of the type of virus assessed, native bovines always featured lower rates of population immunity when compared to bovines coming from other properties, with values for true prevalence varying between 84% and 90%, while for the category of non-native bovines the values varied between 94% and 98%. The results are according to the forecast, bearing in mind the fact that bovines that have been moved, particularly young animals, are subjected to additional vaccinations provided for in legal standards in some units of the Federation. This difference is eliminated with the increase of the age of animals that is indirectly related to the bigger number of vaccinations received.

Specifically for animals reported by the persons interviewed as non-vaccinated, it was recorded that among non-native animals, in global terms, 70% were classified as protected, reinforcing the possibility of their having been vaccinated in the properties of origin.

**Table 34.** Results for bovines of 6 to 12 months of age, according to type of virus and origin of the animals

Type of virus	Native	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
O	No	315	395	80%	94%	91%	98%
	Yes	1.494	2.064	72%	84%	82%	86%
C	No	451	775	58%	98%	95%	100%
	Yes	1.299	2.532	51%	87%	84%	90%
A	No	133	159	84%	96%	92%	100%
	Yes	2.011	2.640	76%	90%	88%	92%

**Table 35.** Results for bovines of 13 to 24 months of age, according to type of virus and origin of the animals

Type of virus	Native	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
O	No	363	418	87%	99%	98%	100%
	Yes	1.366	1.614	85%	99%	99%	100%
C	No	614	895	69%	99%	98%	100%
	Yes	1.275	1.785	71%	99%	98%	100%
A	No	233	250	93%	99%	98%	100%
	Yes	1.816	2.055	88%	99%	98%	100%

**Table 36.** Results for bovines with more than 24 months of age, according to type of virus and origin of the animals

Type of virus	Native	Bovines		Prevalence		Interval of confidence	
		Protected	Total	Apparent	Adjusted	Lower	Higher
O	No	388	421	92%	99%	98%	100%
	Yes	870	955	91%	99%	98%	100%
C	No	767	879	87%	99%	97%	100%
	Yes	844	1.007	84%	99%	98%	100%
A	No	379	393	96%	99%	98%	100%
	Yes	1.143	1.186	96%	99%	98%	100%

Inoculation and recording of the vaccination against foot and mouth disease

Initially, the persons interviewed were asked if they had carried out the vaccination against foot and mouth disease in the stage immediately prior to the collection of the samples (question number 7). Among the persons interviewed, 1.925 (98% of the total) informed having carried out the vaccination, and the percentages per unit of the Federation mostly varied between 95% and 100%. Only in the State of Rio de Janeiro a percentage below 90% was observed.

The official veterinary service of each unit of the Federation checked the recording of the vaccination in the local veterinary units. Among the persons interviewed who answered having vaccinated the animals, 1.885 (98%) recorded the vaccination, while 84 producers (4%) did not vaccinate or did not record the vaccination against foot and mouth disease. The biggest number of these producers was observed in Rio de Janeiro, representing 27% of the persons interviewed in said state. Except for this unit of the Federation, the percentages recorded in the other were close to the records of vaccination presented after the stages of vaccination against the disease, as it may be assessed by means of Table 38, as well as the rates of immune protection recorded in this study.

Considering the results according to the size of the herd, producers with up to 20 bovines featured the highest percentage of persons interviewed that did not vaccinate or did not record the vaccination, representing 65 producers, circa 8% of the total of the 820 persons interviewed in this category. In the category of producers with 21 to 50 bovines, the percentage of persons interviewed that did not vaccinate or did not record the vaccination against foot and mouth disease was of 3% and in the category of producers with more than 50 bovines the percentage was of 1%.

The questionnaire used also allowed for the recording of the producers interviewed that received the assistance of the official veterinary service in the vaccination against foot and mouth disease. In global terms, it was noticed that 169 producers, 9% of the total interviewed, were accompanied by the official veterinary service during vaccination, with the highest percentages recorded in Sergipe (34% of the total of persons interviewed in that state), Rio Grande do Sul (28%), Rio de Janeiro (17%) and Tocantins (11%).

**Table 38.** Comparison between the percentages of recording of the vaccination obtained in the population and in the sample

Sub-population	Records of vaccination against foot and mouth disease	
	% of record in the population	% of record in the sample
Acre plus two municipalities of the State of Amazonas	92%	97%
Bahia	95%	94%
Federal District	93%	99%
Espírito Santo	98%	95%
Goiás	98%	100%
Mato Grosso	99%	97%
Mato Grosso do Sul	99%	98%
Minas Gerais	96%	93%
Paraná	99%	100%
Rio de Janeiro	92%	73%
Rio Grande do Sul	91%	95%
Rondônia	100%	100%
São Paulo	99%	97%
Sergipe	93%	94%
Tocantins	98%	100%

## 6. Conclusions

Irrespective of all the activities and procedures involved in the preparation of the reports of the stages of vaccination, significant correspondence was recorded between the vaccination coverage assessed by means of the record of vaccination and the immunisation levels obtained starting from the results of the diagnosis tests. Somehow, the rates of immune protection recorded reflect the tradition of the country in the conduction of vaccination campaigns against foot and mouth disease, for over three decades used as one of the main strategies of PNEFA, and the good quality of the vaccine used, particularly starting from the 1990s.

According to the description of the profile of the sample and particularly as to the characteristic “number of vaccinations received by the animals” it must be said that the decision to deal with age groups as independent samples to control the effect of multiple vaccinations in the assessment of the levels of immunity featured little discriminating capacity. This fact is validated by the high levels of immunity assessed by the study for all age groups, which also proves the efficiency of Brazil’s programme of vaccination. An efficiency that is translated into the conclusion that in the age group of 6 to 12 months, only 10% of the animals were not vaccinated and that circa 40% featured two or more vaccinations, according to declarations of the persons interviewed. It must be added that circa 85% of the animals between 12 and 24 months, an extremely important age group in the epidemiological model of foot and mouth disease due to the characteristic of movement, featured three or more vaccinations. Other data and information related to the response to vaccination collected by the study may be the object of further analyses.

The study indicated that the bovine population of the zone free from foot and mouth disease with vaccination features sufficient immune coverage for the susceptibility of the population to the foot and mouth disease virus be drastically reduced. This, in turn, reduces the risk of occurrence of infection processes, in addition to inhibiting or reducing viral multiplication, should the animals eventually be exposed to the agent. In sum, this situation allows one to consider the unfeasibility of viral circulation in specific geographical spaces of reasonable dimensions in the zone free from foot and mouth disease with vaccination.

As a complement, based upon the analysis of the laboratory results obtained in this study, it is necessary to highlight that:

- in general, the levels of protection were higher than 90% for nearly all sub-populations considered, extrapolating some initial forecasts, particularly for the age groups concerning younger animals;
- the sub-populations that featured the smaller lower limits for the interval of 95% of confidence relating to the levels of protection were those represented by the State of Goiás (87%) and by the East Livestock Circuit of Minas Gerais (68%). In the latter, the estimated level of protection was below the value desired for eradication programmes (80%);
- the level of protection of 71% for the sub-population of the East Livestock Circuit of Minas Gerais is lower than the one observed for the other sub-populations assessed. On the other hand, this lower immunisation coverage, when associated to the absence of records of clinical disease and to the results of the studies of viral circulation, reinforces the hypothesis of non-existence of residual virus, since there would be in this region about 30% of bovines that would respond as non-protected if they were aggressed by the foot and mouth disease virus ;
- the lowest levels of immunisation coverage, as expected, were recorded for the bovines with ages between 6 and 12 months. However, even for this age group, out of the 18 sub-populations considered in the study, in eight we recorded values for the lower limit of the interval of 95% confidence higher than 90% and, in five, values between 80% and 90%. Only in five sub-populations we recorded values lower than 80%: the East Livestock Circuit of Minas Gerais (56%); Goiás (72%) and Rio de Janeiro (74%), Espírito Santo (77%) and Rio Grande do Sul (79%);



- considering that bovines with more than 12 months of age represent about 80% of the existing population in most of the sub-populations assessed, the rates of immunity recorded for animals with 13 to 24 months or with more than 24 months reinforce the high level of immune coverage in the bovine population of the zone free from foot and mouth disease with vaccination;
- the immunisation levels for non-native bovines, those that entered the properties in which the serum samples were obtained were equal to or higher than those obtained for native animals of the property. This represents important epidemiological information, bearing in mind the risk factor determined by the movement of animals in the epidemiological model of foot and mouth disease;
- in what regards the size of the herds, in global terms the study reinforced the need for special attention on the part of the official veterinary service to the owners of small herds during the stages of vaccination against foot and mouth disease.

It is necessary to consider that as a consequence of the record of outbreaks of foot and mouth disease starting from October, 2005, in Mato Grosso do Sul and in Paraná, great expectations emerged as to the results of this study. However, the interpretation of these results must consider the following aspects:

- the analysis must be limited to the sub-populations considered, avoiding extrapolations to smaller or more specific geographical regions, such as for example, municipalities, areas of regional or international border, such as the one where the outbreaks in Mato Grosso do Sul occurred;
- in most of the sub-populations, the collection of the samples was carried out prior to the notification of the outbreaks of foot and mouth disease. Even in the sub-populations in which the collection of the samples was made after the notification, with the exception of the sub-population represented by the bovine herd of the Pantanal of Southern Mato Grosso, with vaccination in November, the samples collected were related to stages of vaccination carried out prior to the recording of the occurrence of the disease. Thus, for these sub-populations there is no justification for any influence of the outbreaks of foot and mouth disease on the results obtained in this work;
- even in the sub-population represented by the bovines of the Pantanal of Southern Mato Grosso with vaccination in November, where the stage of vaccination and the collection of the samples occurred after the notification of the outbreaks of foot and mouth disease, there was no record of significant differences vis-à-vis the other sub-populations considered in the State of Mato Grosso do Sul;
- one may infer that the high rates of immune coverage recorded in the sub-populations considered in the State of Mato Grosso do Sul and in the State of Paraná contributed to prevent more spreading of the disease.

Finally, the results obtained show a high degree of accord with the elimination of the clinical presentation of the disease in the territory included in the study, with the exception of the municipalities affected by foot and mouth disease in Mato Grosso do Sul and Paraná. They are also consistent with the results of the studies of assessment of viral circulation carried out as part of the epidemiological assessments for obtaining the international recognition of the sanitary condition of free with vaccination. The conclusion is that the levels of population immunity attained were sufficient to break the epidemiological chain of circulation of the virus (reproduction rate lower than 1) and to reach the condition of free. In the case of the municipalities of Mato Grosso do Sul affected by the outbreaks of foot and mouth disease, the presence of infection is explained by local failures in the vaccination coverage (partial vaccination in bigger establishments and non-vaccination in small properties mostly located in rural settlements) and by the evident risk that exists on the international border. In Paraná, the outbreaks occurred as a consequence of the epidemiological link established by the transit of animals coming from properties containing sick animals. However, the high immune coverage recorded in the State prevented the dissemination of the disease.

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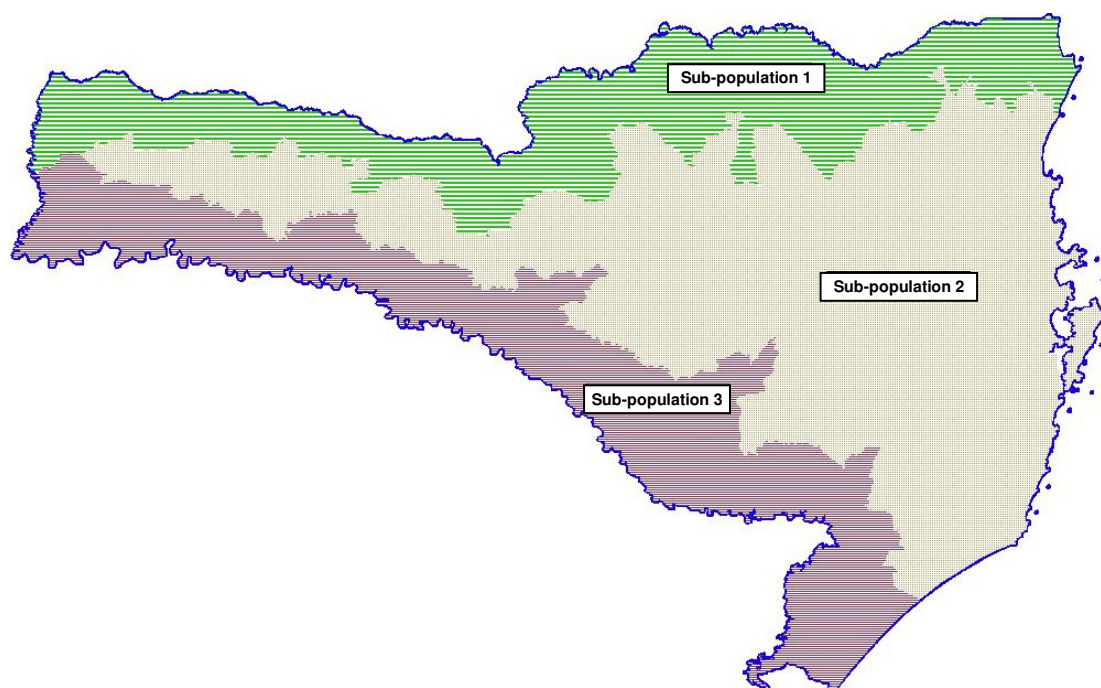
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**Annex 1** – Work carried out in Santa Catarina for the assessment of the presence of vaccinated bovines

During the 75<sup>th</sup> General Session of the OIE, held in the period of May 20<sup>th</sup> through 25<sup>th</sup>, 2007, the State of Santa Catarina was recognised as a zone free from foot and mouth disease without vaccination. The recognition was based upon a favourable opinion of the Scientific Commission, after analysis of the Brazilian report by the *Ad Hoc* group for foot and mouth disease.

In order to meet the conditions expressed in Article 2.2.10.4 of the Land Code of the OIE, in what regards the verification of the absence of circulation of the foot and mouth disease virus, a serum-epidemiological investigation was carried out in the area proposed for the establishment of a zone free from foot and mouth disease without vaccination. The study was conducted in the period from October, 2006 to January, 2007, in compliance with the guidelines and technical bases expressed in Annex 3.8.7 of the Land Code and in Chapters 1.1.1 and 2.1.1 of the Land Manual. The entire work was carried out by the Brazilian veterinary service.

The study was conducted on the entire territory of the State of Santa Catarina that, for the purpose of distribution of the samples, was divided in three sub-populations as shown in Picture 1. It was decided to use one geographical stratification, considering differentiated risks for herds located at approximately 20 km of the state borders. For each of the sub-populations defined an independent sample was calculated, according to the following statistical parameters: minimum prevalence of 2% of infected herds; prevalence, in each herd, of 5% of infected bovines; and 95% of confidence. The target population of the study was made up of bovines between 6 and 36 months of age, divided in the categories of 6 to 12 months; 13 to 24 months and 25 a 36 months. Table 1 presents information on the sampling carried out in each of the sub-populations considered.



Picture 1. Sub-populations considered in the serum-epidemiological investigation, Santa Catarina, 2006

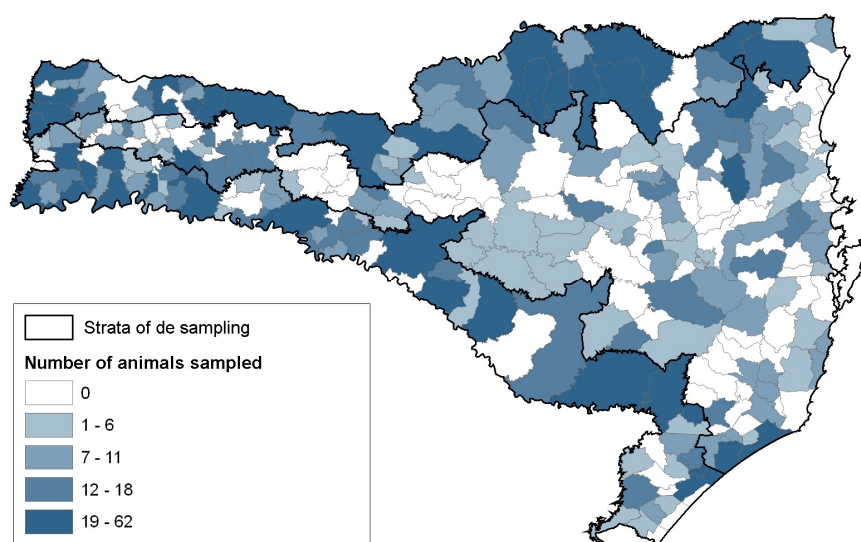
**Table 1.** Total of UPAs, properties, bovine herd and samples collected in the serum-epidemiological investigation, SC, 2006

	UPAs	Properties sampled	Bovine population in the UPAs				Total	Bovine population sampled			Total
			<6m	6-12m	12-24m	>24m		6-12m	12-24m	24-36m	
Sub-population 1	156	428	1.534	1.591	2.042	5.548	10.715	1.145	1.517	981	3.643
Sub-population 2	156	540	1.496	1.566	2.165	5.902	11.129	1.065	1.628	1.019	3.712
Sub-population 3	156	379	1.902	1.633	2.061	6.428	12.024	1.185	1.565	954	3.704
<b>Total</b>	<b>468</b>	<b>1.347</b>	<b>4.932</b>	<b>4.790</b>	<b>6.268</b>	<b>17.878</b>	<b>33.868</b>	<b>3.395</b>	<b>4.710</b>	<b>2.954</b>	<b>11.059</b>

The sampled investigation and the activities of surveillance carried out in Santa Catarina were sufficient to verify the absence of circulation of the foot and mouth disease virus in the State. The description and the full results of the study are available at the Department of Animal Health /SDA/MAPA.

With the purpose of assessing the immunity profile for foot and mouth disease in Santa Catarina, part of the samples collected in the study for the assessment of viral circulation was also tested with the laboratory essay ELISA CFL. In each sub-population, the number of serums to be submitted to titling was calculated, with 95% of confidence, to determine a level of prevalence equal to or lower than 2.5% of vaccination, with a level of accuracy of 1%. Once the total number of animals in each sub-population was calculated, they were randomly distributed as to the type of virus to be tested, since the vaccine includes structural proteins of the viruses type A, O and C. The study considered as being protecting titles those equal to or higher than 2.10, in the case of the samples tested for the types A and O, and to 2.40 in the case of the samples tested for the type C. The study tested 619 serums, 171 of bovines with ages between 6 and 12 months; 266, between 13 and 24 months, and 182, between 25 and 36 months. The distribution of the bovines tested may be assessed by means of Picture 2, and the results are available in Table 2.

Among the samples assessed, 613 (99%) featured results lower than to the titles of protection considered, allowing to characterise the bovine herd of Santa Catarina, in population terms, as non-protected for foot and mouth disease, with the safeguard of the parameters of confidence of the study. Out of the six bovines with protecting titles for foot and mouth disease, three were animals with ages of 6 to 12 months and another three animals had ages between 24 and 36 months. These six samples were tested for the other types of virus and the results indicated that there were remnants of vaccination against foot and mouth disease.





Picture 2. Distribution of the bovines tested for ELISA CFL, according to municipalities and sub-populations

**Table 2.** Results of the tests for ELISA CFL, according to sub-population and age group considered

Sub-population	Bovines of 6 to 12 m			Bovines of 13 to 24 m			Bovines of 25 to 36 m			Total of bovines		
	Total	Non-Protected		Total	Non-Protected		Total	Non-Protected		Total	Non-Protected	
1	69	68	99%	75	75	100%	64	62	97%	208	205	99%
2	42	40	95%	91	91	100%	56	56	100%	189	187	99%
3	60	60	100%	100	100	100%	62	61	98%	222	221	100%
<b>Total</b>	<b>171</b>	<b>168</b>	<b>98%</b>	<b>266</b>	<b>266</b>	<b>100%</b>	<b>182</b>	<b>179</b>	<b>98%</b>	<b>619</b>	<b>613</b>	<b>99%</b>

Annex 2 – Form for information recording on the property and on the results of the interview

 DSA/SDA/MAPA	<b>National Programme for the Eradication of Foot and Mouth Disease – PNEFA</b> Assessment of the Efficiency of Vaccination against Foot and Mouth Disease in Brazil's Free Zone	 Programa Nacional de Erradicação da Febre Afosa
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1. Identification of the property drafted and definition of the number of samples for collection

To be filled by UF Central Unit	a. UF	b. Municipality	c. Name of the Property			
	d. Name of the Owner				e. Code of the Local Unit	f. Code of the Investigation
	g. Category of the Property → <input type="checkbox"/> Up to 20 bovines <input type="checkbox"/> From 21 to 50 bovines <input type="checkbox"/> More than 50 bovines			h. Number of Samples for Collection <input type="checkbox"/> 6 to 12 months <input type="checkbox"/> 13 to 24 months <input type="checkbox"/> > 24 months		

2. Herd in existence in the property randomly chosen at the UF database:

< 6 months		6 to 12 months		13 to 24 months		25 to 36 months		More than 36 months		Total		Grand Total
Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	Male	Female	

3. Does the property fit the category established?  Yes  No → in this case replace, indicating the new property below:

a. Name of the Owner		b. Already registered?	c. Code of the Local Unit
d. Name of the Property			e. Code of the Investigation

4. Rebanho bovino existente na propriedade escolhida em substituição à propriedade inicial:

< 6 meses		6 a 12 meses		13 a 24 meses		25 a 36 meses		> de 36 meses		Total		Total geral
Macho	Fêmea	Macho	Fêmea	Macho	Fêmea	Macho	Fêmea	Macho	Fêmea	Macho	Fêmea	

→ For the collection of samples, comply with the following conditions:

- a specific form should be used, identifying in its heading the name of the owner and the code of the investigation;
- the collection should respect the number per age group established in item 1. h.; if the existing bovine population does not allow for the collection according to the specification, include another property of the same category to comply with the number of samples established (it is not allowed to replace samples among animals from different age groups);
- for each property included with the purpose of completing the number of samples it is necessary to fill specific forms, using the code of the investigation used for the initial property followed by the letters of the alphabet (a, b, c...)

5. Geographical location	a. Latitude				b. Longitude				6. Area (ha)	a. Total		b. Pasture	
	_____° _____' _____"				_____° _____' _____"								

7. Was there vaccination at the last stage?	<input type="checkbox"/> No <input type="checkbox"/> Yes	8. Was the vaccination recorded?	<input type="checkbox"/> Não <input type="checkbox"/> Sim	Day/month/year	Lab. Cod.	Codes for the identification of laboratories: Bayer BA Intervet IN Vallée VA Coopers CO Merial ME Pfizer PF	9. Was the vaccination assisted by the official veterinary service?	<input type="checkbox"/> Yes <input type="checkbox"/> No
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10. Mark with an "X" the option that best characterises the answers of the persons interviewed: C for correct, E for wrong (or for "doesn't know") and ± for partially correct (mark only one option)

Question	C	E	±
a. Is vaccination against foot and mouth disease mandatory?			
b. What are the penalties for those who do not vaccinate against foot and mouth disease?			
c. What is the schedule of vaccination in the state?			
d. What are the age groups that must be vaccinated in each stage?			
e. Should sheep and goats be vaccinated during the stage?			
f. What are the domestic animals susceptible to foot and mouth disease?			
g. What is the proper part of the animal's body to inoculate the vaccine?			
h. What is the volume of vaccine to be inoculated per animal?			
i. Where should the vaccination be recorded at the UF at issue?			
j. What is the period for recoding the vaccination at the UF?			
k. How must the vaccine be stored and transported?			
l. At what age should bovines be vaccinated?			
m. What are the critical signs that characterise foot and mouth disease?			
n. In the event of suspicion of occurrence of foot and mouth disease, is it mandatory to notify?			
o. Should the notification be made swiftly?			

11. Mark with an "X" or fill according to the answers of the persons interviewed:

a. Do you vaccinate all the existing bovines or part of them? All Part b. Do you vaccinate the animals you purchase? Yes No c. Do you vaccinate pregnant cows? Yes No d. Do you vaccinate oxen during the termination stage? Yes No e. Do you vaccinate calves that are nursing (just born)? Yes No f. Do you have a refrigerator? Yes No g. How do you learn about the vaccination stage? Television Notice letter Radio Meetings News paper Mass/Religious services Poster Neighbours Others: h. What do you do with the leftover vaccine? There are no leftovers (I buy the exact number) I give to neighbours or friends I vaccinate other species I keep in the refrigerator to: Re-vaccinate the animals in the property Vaccinate animals born after the stage Vaccinate animals I purchase Use in the next stage	Other ↓
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12. In the event of the presence of clinical signs in the animals in the property, what would you do?

Call a veterinarian you know  Immediately inform the veterinary service  Ask a neighbour or a producer you know for help  Try to solve the problem on your own  
 Doesn't know  Others:

13. How do you vaccinate the animals?  In your own enclosure or trunk  Borrowed enclosure or trunk  In the bow  Other:

14. How do you take care of the vaccination equipment?  Wash with water and soap  Boil  and apply disinfectant  None

15. Frequency with which you change the needle during vaccination:  Use only one  Every \_\_\_ animals  When it breaks/bends  When it gets dirty

16. In the last stage of vaccination, did you notice an intense inflammatory reaction (abscesses) in the animals?  No  In a few animals  In most or in all animals

17. Amount of labour used in vaccination: family members:  permanent employees:  temporary employees:  18. Do you use vaccinators trained by the official service?  Yes  No

19. DO you know the GTA?  No  Yes → In what type of transit do you use it:  within the municipality  to another municipality  to another UF  All

20. Person in charge of the collection of samples and of information:

Legible name	Signature
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First copy → Laboratory / Second copy → central coordination at the UF / Third copy → person in charge of the collection

Annex 3 – Form for recording the information concerning the bovines sampled

 DSA/SDA/MAPA	<b>National Programme for the Eradication of Foot and Mouth Disease – PNEFA</b> <b>Assessment of the Efficiency of Vaccination against Foot and Mouth Disease</b> in Brazil's Free Zone	 Programa Nacional de Erradicação da Febre Afosa
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1. Identification of the property for the collection of samples			2. Contador:	
a. UF	b. Municipality	c. Name of the owner		Sheet _____
d. Name of the property			e. Code of the Local Unit	f. Code of the investigation
				Of: _____

3. Identification of the samples and information on the animals sampled																	
Nº	Identification of the sample		Sex	Age (months)			Born in the property?	Nº of vaccinations in the property	Nº	Identification of the sample		Sex	Age (months)			Born in the property?	Nº of vaccinations in the property
	Code of the investigation	Sequential number		6 to 12	13 to 24	> 24				Code of the investigation	Sequential number		6 to 12	13 to 24	> 24		
1	-	-							46	-							
2	-	-							47	-							
3	-	-							48	-							
4	-	-							49	-							
5	-	-							50	-							
6	-	-							51	-							
7	-	-							52	-							
8	-	-							53	-							
9	-	-							54	-							
10	-	-							55	-							
11	-	-							56	-							
12	-	-							57	-							
13	-	-							58	-							
14	-	-							59	-							
15	-	-							60	-							
16	-	-							61	-							
17	-	-							62	-							
18	-	-							63	-							
19	-	-							64	-							
20	-	-							65	-							
21	-	-							66	-							
22	-	-							67	-							
23	-	-							68	-							
24	-	-							69	-							
25	-	-							70	-							
26	-	-							71	-							
27	-	-							72	-							
28	-	-							73	-							
29	-	-							74	-							
30	-	-							75	-							
31	-	-							76	-							
32	-	-							77	-							
33	-	-							78	-							
34	-	-							79	-							
35	-	-							80	-							
36	-	-							81	-							
37	-	-							82	-							
38	-	-							83	-							
39	-	-							84	-							
40	-	-							85	-							
41	-	-							86	-							
42	-	-							87	-							
43	-	-							88	-							
44	-	-							89	-							
45	-	-							90	-							

→ Attention: the columns Code of the investigation and Sequential number must be mandatorily filled for all samples. The identification in the labels should follow the model below:

Code of the investigation - Sequential	Example →	UF1 - 1	Note. →	Do not write zero before the numbers and separate with hyphen the code of the investigation from the sequential number
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4. Record of dates: collection: \_\_\_\_/\_\_\_\_/\_\_\_\_ → Remittance to the laboratory: \_\_\_\_/\_\_\_\_/\_\_\_\_ → Receipt at laboratory: \_\_\_\_/\_\_\_\_/\_\_\_\_

Legible name	Signature

First copy → Laboratory / Second copy → central coordination at the UF / Third copy → person in charge of the collection



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Secretariat for Agriculture and Livestock Defence*

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