

Online ISSN 2317-4404

BJSCR

16(2)

September/ November 2016

2016



Título / Title:	Brazilian Journal of Surgery and Clinical Research
Título abreviado/ Short title:	Braz. J. Surg. Clin. Res.
Sigla/Acronym:	BJSCR
Editora / Publisher:	Master Editora
Periodicidade / Periodicity:	Trimestral / Quarterly
Indexação / Indexed:	Latindex, Google Acadêmico, Bibliomed, DRJI, Periódicos CAPES e EBSCO host.

Início / Start: Dezembro, 2012/ December, 2012

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DENTAL EROSION: PREVALENCE IN PRIMARY DENTITION OF AMAZON CHILDREN

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Received: 04/14/2016; Accepted: 06/03/2016

ABSTRACT

Today tooth erosion is the situation commonly found in patients of pediatric dentistry offices. Facilities consumption of foods that promote erosion became frequent, bringing with it a percentage rather worrying about the appearance of the problem. The amount of citrus drinks and high-frequency soft drinks consumed is awakening to new information that is part of the set of methodologies for preservation of otherwise healthy dentures. This work, according to its results, is of great importance when you want to avoid dental erosion in children and maintain healthy teeth. AIM: Dental erosion used to be considered an oral finding in adult patients, but nowadays it is known that deciduous teeth are more susceptible to it because of their thinner tissues found in primary dentition. The aims of the present study were to find the prevalence of dental erosion in patients with primary dentition undergoing treatment at the UFPA Dental School and evaluate the influence of acidic beverage on the development of such a disease. STUDY DESIGN: The present study is a standardized clinical report. METHODS: Fifty children who were attending the Children's Clinic of the Dental School of the Federal University of Para were included in the research, with mean age of 4.6 years old among boys and girls. RESULTS: 42% of the children studied had dental erosion and according to the BEWE index, all of them had lower or no risk of disease development. STATISTICS: A chi-square statistic test ($p > 0.05$) showed that the intake of carbonated drinks ($p = 0.025$) and acidic juices ($p = 0.01$) were related to dental erosion in this population. MAIN CONCLUSION: It was found to be necessary to investigate dental erosion and intake of acidic beverages in early ages to prevent possible damage to the permanent dentition.

KEYWORDS: Erosion, prevalence, acid, beverages, children.

1. INTRODUCTION

The acid dental erosion is considered a clinical finding which is related to a progressive and irreversible loss of dental tissues, not involving bacteria but associated with acid presence. The mineral loss in dental hard tissues

could cause painful sensation, micro-hardness and structural defects in the teeth^{1,2,3}.

Citric acid represents the main acid found in fruits and vegetables, showing pH > 5 and > 3 for lemon, for example. The phosphoric acid, which is present in "Cola" drinks and carbonated waters, also has a pH > 3 , on average^{4,5}.

Both *in situ* and *in vitro* studies have shown that the intake of acidic beverages, mostly carbonated drinks and fruit juices, were related to the development of dental erosion^{6,8}. Other factors could influence the loss of dental hard tissues, such as saliva rates, emotional disturbances, systemic conditions and patterns of acidic beverage intake^{2,5,7,9}.

However, it is necessary to evaluate how these factors influence dental erosion. Clinically, the first signs of dental erosion are the loss of enamel brightness and translucency, and micro-hardness³.

In fact, the dentin could be affected by the development of dental erosion. However, it is necessary a histological study to evaluate which tissue was affected. Thus, to identify dental erosion lesions it is necessary to use an accurate index^{10,12,13}.

Studies have shown that mineral loss is greater in primary than in permanent dentition. This finding could be related to the thinner tissues in deciduous teeth^{3,5,13}.

The prevalence found in clinical studies ranges from 32 to 100%, and this great variability is explained by the different methodologies used and cultural variances as well. More studies on primary dentition seem to be necessary to define and compare the prevalence and etiologic factors involved^{7,14,15,16}.

The aims of the present study were to determine the prevalence of tooth wear caused by acid dental erosion in a group of 50 children undergoing treatment at the Federal University of Para Dental School and determine the relationship between erosion and acidic beverage and dietary constituents.

2. MATERIAL AND METHODS

Based on ethical principles set by the Declaration of Helsinki, this study was approved by the research ethical committee of the Federal University of Para. All caregivers signed an informed consent form allowing their children to participate in the present study.

The study involved 50 children who were attending the Children's Clinic of the Dental School of the Federal University of Para. They were given routine dental care or referred to specialists for other reasons, but they were not found to have any tooth tissue loss.

The children of both genders had a mean age of 4.6 years old, all presenting exclusively primary teeth and no dental loss. An informed consent form was signed by their parents or caregivers if they really wished to participate.

One examiner (CMLO) carried out the clinical examinations under the same conditions at Children's Clinic of the Dental School of the Federal University of Para. All the children were examined on a dental chair in a dorsal decubitus position, and only oral mirror and artificial light were used. The examiner is a paediatric dentist and was calibrated on the basic erosive wear examination index (BEWE) by using photographs.

The BEWE index is easy to use for ranking dental hard tissues according to a score ranging from 0 to 3 (Table 1) and the results can be compared to other indexes^{6,17,18}.

Table 1: Criteria for ranking erosive wear according to Bartlett D, Ganss C, Lussi A, 2008.

Score	
0	No erosive tooth wear
1	Initial loss of surface texture
2*	Distinct defect, hard tissue loss <50% of the surface area
3*	hard tissue loss > or = 50% of the surface area

*for scores 2 and 3, dentine is often involved.

Although all teeth were examined, only the highest score of each sextant was considered. Next, the scores were added up and the risk of dental erosion was obtained for each patient.

Because this index was set for permanent dentition, we suggested an adaption to the primary dentition. Table 2 lists the total scores, whereas Table 3 lists dental erosion risk and management required.

The data collected were recorded and then a question-

naire about drinking habits was applied to parents or caregivers of the children examined.

Table 2: Total scores to calculate the patient's risk of developing dental erosion. Adapted from Bartlett D, Ganss C, Lussi A, 2008.

Sextant examined	
A	Highest score between 55-53
B	Highest score between 52-62
C	Highest score between 63-65
D	Highest score between 75-73
E	Highest score between 72-82
F	Highest score between 83-85
Cumulative Score=Patient Dental Erosion Risk	
A+B+C+D+E+F	

Table 3: Risk levels as a guide for clinical management according to Bartlett D, Ganss C, Lussi A, 2008.

Risk Level	Cumulative scores of All sextants	Management
None	> or = 2	Routine maintenance and observation.
Low	Between 3 and 8	Dietary assessment, routine maintenance and observation.
Medium	Between 9 and 13	Identify the main aetiological factors for tissue loss and strategies to eliminate impacts.
High	= or < 14	Identify the main aetiological factors for tissue loss and strategies to eliminate impacts.

A descriptive method was used to evaluate the prevalence of dental erosion, whereas the relationship between presence of dental erosion and drinking habits was analyzed by using chi-square test at significance level set at $p < 0.05$.

According to the risk level found in each child and the data collected from the questionnaire, one can state that appropriate management was done in each case.

3. RESULTS

Of the 200 children seen between the second semester of 2009 and the first semester of 2010 at the Children's Clinic of the Dental School of the Federal University of Para, 50 met the inclusion criteria to participate in the present study. The prevalence found was 42% for this population.

According to the BEWE index, risk level among children with dental erosion was 71% compared to 29% among those with no dental erosion.

With regard to carbonated drinks, it was demonstrated that 100% of the children who drank such beverages less frequently (up to four times a week) did not have dental erosion, whereas those who frequently drink carbonated beverages had varied prevalences (Table 4). It was found a positive relationship between presence of dental erosion and intake of carbonated drinks ($p = 0.025$).

In the present study, the intake of juices by children involved lemon, acerola cherry, and orange. All children

who frequently drink lemon juice (4 times a week or more) presented dental erosion, whereas varied results were found regarding the other fruits (Table 5). According to the statistic analysis, it was found that the frequent intake of acidic juices is positively related to dental erosion in the children examined ($p = 0.01$).

Table 4: Percentage of carbonated beverages frequently drunk by children with dental erosion, Belém- PA, 2010.

Dental Erosion	Most frequently drunk carbonated drinks				
	None	Cola	Guaraná	Orange juice	Total
No	100.00	70.00	33.33	57.14	58.00
Yes	0	30.00	66.67	42.86	42.00
Total	100.00	100.00	100.00	100.00	100.00

Table 5: Percentage of acidic juices frequently drunk by children with dental erosion, Belem-Pa 2010.

Dental Erosion	Acid juices frequently drunk				
	None	Acerola	Orange	Lemon	Total
No	33.33	83.33	64.00	0.00	58.00
Yes	66.67	16.67	36.00	100.00	42.00
Total	100.00	100.00	100.00	100.00	100.00

When the intake of carbonated drinks and acid juices were compared in terms of dental erosion, it was observed that the intake of guarana carbonated drink was the only beverage, independent of the concomitant acidic juices intake, associated to dental erosion in this population.

To investigate whether abundant intake of water prevents erosive lesions, it was found no statistically significant difference between children who drink abundant amount of water and those who do not ($p=0.66$).

In Pará, there's a regional habit to drink a beverage based on açaí fruit. Thus, it was also decided to investigate whether this beverage affects the probability of dental erosion development. Even considering that people who drink açaí everyday have no dental erosion (Table 6), no statistic difference was found between intake of this beverage and dental erosion in the children examined.

Table 6: Dental erosion related to açaí drunk by children examined at the Federal University of Pará, Belém, PA, 2010.

Dental Erosion	Açaí daily consumption		
	No	Yes	Total
No	50.00	68.18	58.00
Yes	50.00	31.82	42.00
Total	100.00	100.00	100.00

4. DISCUSSION

Even with all the difficulty of conducting a clinical examination in children, it is necessary to encourage this kind of study because of the scarce literature on dental erosion in primary dentition. Although there are many *invitro* and *in situ* studies, they have limited results on the amount of oral cavity variables^{4,11}.

The present study has found a high prevalence of dental erosion (42%) in the children who participated in the research and were on dental treatment without any reference of dental erosion. Similarly, it was found a prevalence of 32% in children examined in Germany¹⁹.

Other prevalence studies relating age and diagnostic criteria are very different, with their results ranging from 30 to 100%^{7,14,15}. In some of these studies, the children were included specifically because of tooth wear problems, which explains the high prevalence found⁹.

Some cultural variances could influence this variability, for example, studies conducted in Saudi Arabia investigated only boys^{10,19}.

The prevalence of dental erosion in Europe is very high, probably because of the dietary constituents or the importance given to the theme, which is corroborated by most important and original studies^{2,6,14,16,18,19}.

Although the high prevalence found in the present study, the risk level in children with dental erosion lesions was none or low. This could be explained by the lower age and shorter time of dental exposure to the acid action. However, studies conducted in Saudi Arabia showed that 5-6-year-old children have moderate to extensive lesions, rates ranging from 34 to 51%^{15,20}. Moreover, in Europe such rates are virtually the same, ranging from 35 to 40% in children aged between 2-7 years old²⁰.

In Brazil, a cross-sectional study with children aged 12 years old reported a dental erosion prevalence of 13%, but the index used was different and the children were examined at schools without appropriate condition to conduct a good clinical exam¹⁶.

According to the literature, the intake of acid beverage is the most important etiological factor regarding dental erosion in children, and the present study confirms this^{1,2,3,7,21}. The frequent intake of carbonated drinks and acidic juices were statically related to the presence of dental erosion in the children examined.

In previous studies on carbonated drink, it was found a relationship between development of dental erosion and other acidic beverage^{18,22}. The drinking habits regarding these beverages appear to influence this relationship, however this was not the focus of the present study¹⁵. In other studies, the intake of carbonated drinks was not related to dental erosion^{9,19}.

The finding that guarana carbonated drink is more involved in the development of dental erosion diverges

from previous studies^{4,5,8}. However, this could be explained by the high consumption of this kind of carbonated drink because of its lower price compared to others. In addition, the present study was not aimed at investigating the economic situation of the population studied.

With regard to the acidic juices, it was observed that all of them have acid pH. But the prevalence of dental erosion corresponded to the acidic pattern of the beverage⁶. Lemon juice, when frequently drank, was fully associated with the presence of dental erosion in the children evaluated. This finding is corroborated by other studies available in the literature⁴.

Abundant water intake was not related to a decreased dental erosion in this population, which diverges from other studies reporting a negative association between these variables^{3,20}.

Despite the lack of previous knowledge in the literature about the characteristics of Açaí, a regional beverage in Pará. In fact, this fruit should be investigated to know whether it has any influence on dental erosion in the Amazon child population studied. The statistic test showed no relationship between both variables, although it was observed among the children drinking Açaí every day that there was no case of dental erosion. This finding could stimulate further research on the Açaí properties.

A good diagnosis of the disease and simple diet recommendations could help treat an initial lesion of dental erosion, avoiding complications in permanent dentition. Thus, it is necessary to stimulate further prevalence studies and etiological considerations about dental erosion, especially in Brazil, because the scarce amount of such studies on primary dentition.

5. CONCLUSION

This study shows high prevalence of dental erosion in the children examined (42%), with rate ranging from low (29%) and absent (71%).

Intake of soft drink and acidic fruit juice presented a statistically significant relationship with dental erosion ($p=0.025$ and $p=0.01$).

According to these results, early diagnoses should be stimulated as well as further clinical research on deciduous dental erosion treatment and prevention, including diet orientations.

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ENDODONTIC TREATMENT: FACTORS RELATED TO POSTOPERATIVE PAIN AND THE SUCCESS OF THERAPY

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Received: 07/04/2016; Accepted: 09/16/2016

ABSTRACT

The success of endodontic treatment is directly related to the infection control also depends on the instrumentation technique, the quality of their preparation, defined in degrees after the intervention in the root canal for treatment. Postoperative pain is characterized by pain manifestation of any degree and can occur soon after the beginning of root canal treatment, it is a condition that causes discomfort and pain due to infectious condition had as common in endodontic treatment, however, post-surgical pain does not influence the long-term success when the cause is diagnosed and opposed to time. In this sense, this study aims to verify the occurrence of postoperative pain in patients with endodontic treatments carried out in single or multiple session as well as to know the possible causes of postoperative pain and identify possible means to ensure the success of therapy endodontic. The success of this treatment depends on the fulfillment procedures following scientific, mechanical and biological principles, which are directly related to the successes and failures of endodontic treatment. Therefore, the procedures before and during endodontic treatment will provide a faster recovery of the patient, reducing infection rates and consequently the postoperative pain framework, contributing to the success of the intervention performed.

KEYWORDS: Endodontic treatment, postoperative pain, procedures, success.

1. INTRODUCTION

The endodontic treatment should be construed as a factor that can cause inflammatory reactions and thus causing postoperative pain. Regarding the treatment in a single session, even in the absence of microorganisms, resistance in vital pulp has a higher possibility of severe postoperative pain. In this perspective, a proper conduct of the preparation from the beginning, contributes to small trauma incidence, which leads to an inflammatory

occurrence of low intensity¹.

However, if the preparation is more traumatic, inflammatory greater possibilities occur, as well as the increase in painful symptomatology. Nevertheless, it should be noted that the post-operative pain has been conceptualized as a pain of any degree, whose manifestation occurs so start root canal treatment in as endodontic flare-up has been described as the continuation of pain and or swelling after endodontic treatment which ends up interfering with the patient's daily life, it feels obliged to return to the dentist's office in search of a new intervention to contain the pain that bothers you¹.

Thus, the success of endodontic treatment is directly related to the control of infection, which in turn causes the pain. But there are other factors that can have a negative influence, as the instrumentation technique and the quality of interventions at all stages after the intervention in the root canal for treatment.

The literature reports that the most common problems that can lead to endodontic failure, can have multiple etiologies, with reports Luvisotto (2007)²; the lack of aseptic control during treatment, improper access to the pulp cavity, undetected channels, fault instrumentation, inadequate and unsatisfactory fillings or missing coronal restoration after the endodontic treatment.

About this, Henriques *et al.* (2011)³ explains that the etiology of endodontic failures have higher incidence in cases of persistence or reintroduction of microorganisms in the root canal system because of an irregular mechanical-chemical preparation, which thus breaks with the aseptic chain, in cases of extra root infections that microorganisms runs through the body's defenses and can remain alive in the periradicular space and accidents or iatrogenic caused by the professional during the treatment at an early stage.

Nonetheless, what are the procedures to identify the possible causes of postoperative pain in patients under-

going endodontic treatment in single or multiple session? What is the most appropriate procedure to avoid postoperative pain in patients who carried out root endodontic treatments in both situations?

This study is justified by the importance of knowing the causes of postoperative pain, and these are related to the number of treatment sessions, allowing the adoption of targeted procedures aiming to reduce the incidence of postoperative pain in patients who underwent root canal treatments in a single session or multiple.

[Thus, this study aims to verify the occurrence of postoperative pain in patients with endodontic treatments carried out in single or multiple session as well as to know the possible causes of postoperative pain in patients undergoing endodontic treatments and identify possible means to ensure the success of endodontic therapy.

2. MATERIAL AND METHODS

To carry out this study we chose the proposal of Ganong (1987)⁴, according to the following steps: 1) identification of the research question, followed by a search of the descriptors or keywords; 2) determining the criteria for inclusion or exclusion of research in online databases; 3) categorization of studies, summarizing and organizing relevant information; 4) assessment of studies for critical analysis of the extracted data; 5) discussion and interpretation of the examination results, contextualizing theoretical knowledge and evaluating their applicability as; 6) presentation of the integrative review and synthesis of knowledge of each article reviewed briefly and systematic way.

In the present study the guiding question of the integrative review was: verify the occurrence of postoperative pain in patients with endodontic treatments carried out in single or multiple session as well as to know the possible causes of postoperative pain in patients undergoing endodontic treatments and identify possible means to ensure the success of endodontic therapy.

Bases (Latin American and Caribbean Literature on Health Sciences) LILACS, SciELO (Scientific Electronic Library on Line) and PubMed (- NCBI US National Library of Medicine National Center for Biotechnology Information) were consulted. Studies that have addressed the thematic, published from 2000 to 2015, regardless of the languages of publication were included. The following controlled for the search and also used as keywords descriptors were used: Endodontic treatment, postoperative pain, procedures, success.

3. LITERATURE REVIEW

Endodontic treatment

Endodontic treatment, as explained Waskiewicz *et al.* (2013)⁵, is to restore normalcy to the periapical tissues in abnormal state, minimizes the concentration of microor-

ganisms present on site that are causing infection, pulp necrosis and pain. In this way of intervention, the incidence of post-operative pain, after treatment, almost always as a result of acute inflammation response of periradicular tissue, and can take a few hours or days after endodontic treatment. Watanabe (2012)⁶ notes that the endodontic treatment usually consists of a combination of a chemical-mechanical working, at which the root canal space undergoes preparation for the introduction of a biocompatible material to seal the channel along its length. By this action, the microorganisms and organic matter rather than proliferate are removed by favoring the health of periradicular tissue. Following this line of reasoning, Petrini (2010)⁷, points out that the endodontic treatment is designed as a basic procedure in dentistry that can cause postoperative pain, regardless of the degree she started, or conceptualized flare-up as the moment the commencement or continuation of pain and/or swelling after endodontic treatment. The occurrence of postoperative pain is usually caused by acute inflammation, such as periradicular tissue response, beginning shortly after the endodontic procedure. It is noted that the endodontic treatment is a common intervention in dentistry that may lead to postoperative pain, regardless of the number of sessions. However, the prevalence of postoperative pain or flare-up are important factors when deciding the clinical procedure to be used, which requires attention and care.

Based on several studies, Luckmann *et al.* (2013)⁷ reported that the process of a proper root canal treatment requires at least the following: the correct handling of cases the method of treatment and filling, the expertise of the operator, the technical difficulties of the case, the resources of time of treatment, the complete knowledge of dental anatomy, obtaining quality radiographs for the study, the presence of calcifications and the slopes of the teeth in relation to the arcade, determining the length of work, disinfection between sessions and radiological quality control shutter. All these procedures should be done with caution, considering each case.

Pain incidence in endodontic treatment

The incidence of pain in endodontic treatment, has been a topic of discussion ever since there professionals who believe that the success and decreased pain, endodontic treatment, is the result of a single query for treatment. However, many professionals believe that when endodontic procedures are performed in multiple sessions, ensure proper cleaning of the root canal system, prioritizing the shutter, allowing enough time to have a thorough cleaning of the root canal system. The pathogenesis of complications, endodontic post-treatment indicate that the causes of flare-up include mechanical factors, chemical and microbial or affecting the periapical tissues¹.

Pinheiro *et al.* (2003)⁸ conducted a study with 60 teeth that had persistent apical periodontitis after endodontic treatment to evaluate the microbiota present. It was found microorganisms on teeth 51 and only one or two species per channel. The isolated microorganisms were 57.4% and 83.3% anaerobic facultative Gram-positive. The most commonly found species was *Enterococcus faecalis*. However, in teeth with clinical symptoms, it was noticed polymicrobial infections and strict anaerobes. It is noteworthy that the emergence of post-operative pain is often associated with acute inflammation as a response of the periradicular tissues. The research aimed to evaluate postoperative pain after endodontic treatment in dental school in Sudan. Where were selected 234 patients aged 18-62 years. Endodontic treatment was performed by undergraduate students in single and multiple sessions. The postoperative pain was reported by each patient using scale in two time intervals, 12 hours and 24 hours. It was found that there was no significant difference in postoperative pain between one visit and multi-session⁶.

Waskiewicz (2013)⁴ studied the severity of post-filling pain in adolescents undergoing endodontic treatment on one or both visits. They evaluated 121 patients aged from eleven to eighteen who had molars with pulp necrosis and were distributed between the two groups at random. For observation, all dental elements were prepared using a pre-cervical enlargement and a standard technique in instrumentation entire length of the root canal. In this study, postoperative pain was assessed on a visual analogue scale of 0-5. In the results it was observed that the rate of pain was 10.5% (6 of 57 cases) group in a single visit, and 23% (14 of 61 cases) in two visits group. It was found that there was no significant difference between groups. During the observations it was established that the intensity of pain was similar in both cases, particularly as the flare-ups, with a prevalence of 1.75% in single-visit group and 1.67% in the two group visits. It was concluded that the post-dental filling pain was more frequent in the group submitted to two sessions, but without significant differences.

The study conducted by Petrini (2010)⁶ aimed to compare the incidence of flare-up after filling of root canals procedures performed in single or multiple session in the one year period and establish the relationship between pre- and post-operative pain, setting the decrease in pain at intervals of 1, 7 and 30 days after root canal filling. To this end, there was existing clinical factors before, during and after the complete endodontic treatment of each tooth, including teeth with pulp vitality, presence or absence of preoperative pain, postoperative flare-up, and decreased pain postoperative. In patients requiring endodontic treatment in more than one tooth, the service had four weeks between each tooth. The pulp

vitality was determined by combined electrical test with the presence of bleeding pulp. Revisions attendances after root canal filling given in 1, 7 and 30 days. To give more authenticity to the study, effected up interviews with patients to determine whether or not painful symptoms between visits and when there was; how was the decrease in the range of queries. Pain was recorded as follows: no pain, negligible or moderate / severe. The flare-up endodontic been reported when patients have uncontrolled pain requiring medication and / or volume. Therefore, it treated endodontically 283 teeth in 255 patients; of these 56 were excluded for not attending the revaluation, 10 flare-up occurred in 21 multi-session and 19 in a single session. Of the 107 teeth that had the full treatment in one session, 67 had preoperative pain and 50 reported postoperative pain. Of the 40 teeth that had no preoperative pain, 8 had postoperative pain. In multi-session, 88 teeth presented with preoperative pain and 55 with postoperative pain. Of the 48 teeth that had no preoperative pain, only 6 had postoperative pain. Teeth with vitality had less postoperative pain than non-vital (not statistically significant). The single session of patients index showed that postoperative pain negligible on the 1st and 7th days were respectively 35.5% and 16.3%. In multiple session was 30.2% and 9.8%. Patients who had moderate / severe pain on the 7th day was higher in multi-session than in single session. On the 30th day there was no postoperative pain. It found a higher incidence of postoperative pain and flare-up after endodontic treatment in one query. However, the single session has been a more effective alternative to the multi-session, especially in communities where patients are often lacking after the first visit to attenuate the pain.

In this regard, Watanabe (2012)⁵ noted the clinical causes of failure and limitation of previous endodontic treatment by an inspection of the root apex and root surface dry with magnification of 26 times using a surgical microscope during endodontic microsurgery. Data were collected from patients between March 2001 to January 2011. Among the 493 teeth included in this study, we obtained the following results: infiltration by filling material (30.4%); channels not found and not treated (19.7%); insufficient dental fillings (14.2%); over-dental fillings (3.0%), anatomical complexity (8.7%), iatrogenic problems (2.8%); Apical calculations (1.8%) and cracks (1.2%). It is suggested that the clinical use of the microscope can make a more favorable prognosis.

4. DISCUSSION

Studies report that many factors influence the implementation and success of Endodontic treatment, among them, there is the perfect sterilization and disinfection of instruments and equipment to be used in the treatment, as if the residue remains in the files will be contamination leads producers to patient, which will

cause failure in endodontic treatment¹⁰.

In this view, the success rates and failure for root canal treatments, is a topic discussed and researched over the years, and today, with data collected at different times suffer a 53% increase to 95%, and research conducted in recent years indicate values close between 90% and 95%, due to the use of a modern, scientifically grounded best therapy that is constantly evolving. When performed endodontic retreatment the success rate approaches 80% thus becomes an effective alternative for cases of endodontic failure¹¹.

According to research conducted by Luckmann *et al.* (2013)⁷, endodontic treatment aims to maintain the dental element in function of the stomatognathic system, in which one of the main focuses is to keep special care to preserve the health of the patient. The success of this treatment depends on the fulfillment procedures following scientific, mechanical and biological principles, which are directly related to the successes and failures of endodontic treatment. The same researchers admit that the combination of the essential factors in endodontic success can be summarized in: clinical silence (absence of pain, swelling, fistula), average periapical bone structure (uniformity of lamina dura, regular periodontal space, absence or reduction of bone thinning, absence or interruption of root resorption), tooth in function and presence of coronary perfect sealing. Alert also to achieve a better result in teeth that are vital when compared to treatment with teeth that have had a necrotic pulp.

Other studies reported the successful endodontic treatment depends on the canal preparation, which comprises an intervention that could generate a tissue injury, and may thereby produce a postoperative framework sometimes with inflammation in tissues periradicular^{3,5,12,13}. It is important to emphasize that to get success rate in treatment of root canals of teeth with apical periodontitis, it is necessary to consider such factors as: the neutralization and removal of necrotic-toxic contents of the root canal system, a suitable mechanical preparation, employment a dressing used and the sealing of root canals by dental filling¹⁴.

The study conducted by Henriques *et al.* (2011)³, states that the success of an endodontic treatment is associated with the achievement of several factors, which begins with the selection and diagnosis of the case to be treated, including their therapy and prognosis, passing by careful execution of chemical mechanical preparation technique considering the complex morphology of the root canal system, three-dimensional shutter, as well as the maintenance of aseptic chain, among other operative steps by clinical monitoring and periodic patient over time of treatment. It is noteworthy that even with all the evolution of technology have endodontics, there are still many cases of failure related to microbiology, morphol-

ogy and technique used in endodontic treatment routine. In cases of failure of this type of treatment, the most suitable and used alternative is the endodontic retreatment.

Given these findings, recapitulate Strela (2004)¹⁴, writing that the American Association of Endodontics (1994) classifies the case as endodontic success based on the following criteria: a) Clinical: absence of periodontal disease or dental endodontics related to mobility; absence of fistula; absence of symptoms to percussion or palpation; tooth function; no edema or signs of infection; absence of other symptoms previously described by the patient. b) Radiographic: normal periodontal ligament space or lesser thickness than 1 mm; absence of pre-existing bone thinning; Normal hard blade in relation to the adjacent teeth; absence of resorption compared to the original x-ray; three-dimensional filling space channel, within the limits of its space to approximately 1 mm short of the radiographic apex.

According this line of reasoning, Henriques *et al.* (2011)³, mentions the European Society of Endodontics, which in 1994 listed out some essential clinical criteria for endodontic success, such as the absence of pain, infection, swelling, symptoms on palpation and percussion, fistula, periodontal disease associated with endodontium, tooth function in the arcade, the absence of subjective symptoms reported by the patient and X-ray as a space of normal periodontal ligament or insignificant thickness (less than 1 mm), elimination of a previous thinning periradicular, regular hard blade in relation to the adjacent bone, absence of resorption compared to the original three-dimensional X-ray and shutter the visible channel space within the limits of its space to approximately 0.5 mm short of the root apex are suggestive of success when considering a preservation period of approximately two years. Therefore, more important than the number of sessions, is how the professional proceeds to perform endodontic therapy, because success depends on the professional awareness, proper hygiene, disinfection and preparation of the channels, it is of decisive actions to percentage of successful therapy.

5. CONCLUSION

The positive result depends on the level of knowledge of pulp anatomy, obtaining good quality radiographs to study anatomical abnormalities or inclination of the dental element with the use of appropriate instruments as essential conditions to prevent operative accidents and achieve positive rates of procedures. The care before and during endodontic treatment will provide a faster recovery of the patient, reducing infection rates and consequently the postoperative pain framework, essential factors for successful treatment.

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PCR OF HEMOCULTURE AS STRATEGY TO IMPROVE THE DETECTION OF *Trypanosoma cruzi* IN THE CHRONIC PHASE OF CHAGAS DISEASE

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Received:08/19/2016; Accepted:10/22/2016

ABSTRACT

In order to associate the specificity of hemoculture and the sensitivity of PCR, we have proposed to perform the PCR from samples obtained from hemoculture (PCR-HC) of 79 patients in the chronic phase of Chagas disease. In addition to the PCR-HC, the methods of hemoculture and PCR of blood (PCR-BL) were performed. To carry out the PCR-HC, hemoculture and PCR-BL were used 2.4 mL of hemoculture sediment, 30 mL and 10 mL of blood, respectively. The DNA was extracted of each sample and the fragment of 330 pb of k-DNA from *Trypanosoma cruzi* minicircle was amplified. The PCR-HC ($p=0.00005$) and the PCR-BL ($p=0.00121$) were significantly more positive than the hemoculture. The positivity of the PCR-HC was also significantly higher ($p=0.00032$) than the PCR-BL. We have concluded that the PCR-HC is a valid alternative method to increase the detection of the parasite, improving the parasitological diagnosis and identification of etiologic treatment failure.

KEYWORDS: Diagnosis; Chagas disease; PCR of blood; PCR of hemoculture; hemoculture.

1. INTRODUCTION

Currently, 5 to 6 million people are infected with *Trypanosoma cruzi* all around the world, especially in Latin American countries, where Chagas disease is endemic¹. During the chronic phase of infection, which is characteristic of low parasitemia, methods of parasitological diagnosis become limited. Thus, the association between a highly specific method such as hemoculture (100%)^{2,3} and the high sensitivity of the PCR (76.4 to 100%)^{2,4} could be an interesting approach to increase the detection capability of *T. cruzi* in patients in the chronic phase of infection. In this context, a pilot study was proposed to perform PCR from hemoculture samples obtained from patients in the chronic phase of Chagas disease.

2. MATERIAL AND METHODS

Seventy-nine patients with reagent ELISA method Chagas Test Elisa III (Bioschile® Ingenieria Genética S.A, Chile), who were attended in 2012 in the Chagas Disease Laboratory of the Universidade Estadual de Maringá (UEM) and ambulatory of the Hospital Universitário de Londrina, were recruited for this study. The age of these patients ranged from 35 to 89 years, with an average of 60.5 ± 9.9 . Females were predominant, corresponding to 55.7% of the studied population. The patients signed a free and informed consent form approved by Permanent Committee of Ethics in Research Involving Human Beings (COPEP) of UEM, under protocol number 012/2010.

The hemoculture (HC) was performed with 30 mL of blood distributed in six heparinized conical tubes (15 mL). The tubes were centrifuged at 4 °C, 209.44 rad/s for 30 minutes to remove the plasma and add Liver Infusion Tryptose medium as previously described⁵, with modifications. Once a week, the tubes were homogenized and the pellet aliquot was analyzed every 30 days for a total of 180 days. In this period, 0.4 mL of the pellet of each one of the six tubes, totaling 2.4 mL, was added to an equal volume of Guanidine-HCl 6M / EDTA 0.2M for PCR analysis of hemoculture (PCR-HC). After one week at room temperature, these samples were boiled at 100 °C for 15 min and stored at 4 °C until use⁶. From this mixture, 2.4 mL of material was used for DNA extraction.

The PCR of blood (PCR-BL) was performed with 10 mL collected, in the same time of hemoculture, in a conical tube (50 mL) containing an equal volume of Guanidine-HCl 6M / EDTA 0.2M⁷ to extraction and amplification of DNA.

The DNA extraction and the conditions of PCR reaction and revelation of the amplified products were as

previously described⁸. The DNA was amplified in an automatic thermocycler (Techne® TC - 512 Staffordshire, England). The extraction and amplification of DNA were monitored using negative controls (non-infected individuals from non-endemic areas) and positive controls (individuals infected with *T. cruzi*). For amplification step, DNA of parasites obtained from culture was also used as positive control. The amplified products were visualized on 4% polyacrylamide gels, revealed by silver salts and digitally stored.

Data analysis was performed using *Software SAS 9.1*. The *Chi-square test* was used to investigate possible associations between variables. The significance level was 5% ($p < 0.05$).

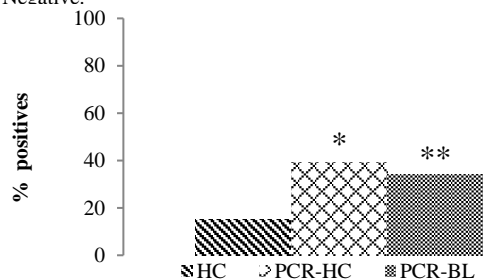
3. RESULTS

The hemoculture was positive in 15.2% (12/79) of the patients and the PCR-HC detected 39.2% (31/79), being the difference between these methods significant ($p=0.00005$). Twenty-seven (34.2%) patients were positive by PCR-BL, with an also significant difference ($p=0.00121$) in relation to hemoculture (Table 1, Figure 1).

Table 1. Comparison of the hemoculture (HC) results with the PCR of hemoculture (PCR-HC) and the PCR of blood (PCR-BL) in patients infected with *Trypanosoma cruzi* (n = 79).

Variables	HC				<i>p</i>
	P		N		
	n	%	n	%	
PCR-HC					
P	11	13.9	20	25.3	0.00005*
N	1	1.3	47	59.5	
PCR-BL					
P	9	11.4	18	22.8	0.00121**
N	3	3.8	49	62.0	

*Significant difference between HC and PCR-HC; **Significant difference between HC and PCR-BL; $p \leq 0.05$; P = Positive; N = Negative.



*Significant difference between HC and PCR-HC;

Figure 1. Positivity of the hemoculture (HC), PCR of hemoculture (PCR-HC) and PCR of blood (PCR-BL) in patients infected with *Trypanosoma cruzi* (n=79). Após o ";" No pé dessa figura falta a seguinte informação: **Significant difference between HC and PCR-BL; $p \leq 0.05$.

For 31 patients with PCR-HC positive, 18 (58.1%) were also positive by PCR-BL. For 48 patients with PCR-HC negative, 9 (18.7%) were positive by PCR-BL. Even so, the number of patients with PCR-HC positive (31/79) was significantly higher ($p=0.00032$) when compared to those detected by PCR-BL (27/79) (Table 2).

Table 2. Comparison of the PCR of hemoculture (PCR-HC) with the PCR of blood (PCR-BL) in samples from patients infected with *T. cruzi* (n = 79)

Variables	PCR-HC				<i>p</i>
	P		N		
	n	%	n	%	
	PCR-BL				
P	18	22.8	9	11.4	0.00032*
N	13	16.5	39	49.4	

*Significant difference between the methods; $p \leq 0.05$; P = Positive; N = Negative.

4. DISCUSSION

In order to increase the detection of *T. cruzi* in patients in the chronic phase of Chagas disease, the specificity of hemoculture was associated with the sensitivity of PCR and the PCR-HC was carried out. This method compared with the PCR-BL and with the hemoculture showed higher detection capability of the parasite.

The performance of the PCR-HC carried out in samples collected at 180 days (period of higher positivity of the hemoculture) was significantly better ($p < 0.05$), when compared with PCR-BL and the HC. The PCR-BL also showed a higher parasite detection capability than the HC, with a significant difference between these two methods, in both treated and untreated patients agreeing with other authors^{2,4,7,9,10,11}. The hemoculture was the method with the lowest detection capability of the parasite, however, it is important to consider its fundamental role for the diagnosis of infection by *T. cruzi*. It is also important to emphasize that the detection of the parasite was higher only when the HC was associated with the PCR (PCR-HC). It should be noted that even in the absence of the parasite multiplication, PCR-HC can detect parasites independent of Discrete Typing Unit (DTU) that belong, differing from HC that can select a DTU over another, depending on the time required for growth and development of the parasite. Other authors have also associated a molecular method (conventional PCR) with a parasitological method (xenodiagnosis - PCR-XD) and have observed an increase in *T. cruzi* detection capability when compared to the parasitological method carried out singly^{9,12}.

The association between the PCR and the HC (PCR-HC) was important to increase the parasite detection capability, because this method was also significantly better when compared to PCR-BL. This is the first study

reporting a significant difference when two methods with distinct approaches (HC and PCR) were associated. Another study⁵ that associated a molecular method (PCR) and another parasitological method (xenodiagnosis - XD) resulting in PCR-XD, showed no significant difference when compared to PCR-BL. The highest positivity of the PCR-HC can be explained by the larger volume of blood used in the hemoculture and by the parasite multiplication in the culture medium, which does not occur with the PCR-BL. Even the PCR-HC detecting the parasite in a significantly higher number of samples compared to PCR-BL, 9 samples were only positive by PCR-BL and 13 were only positive by PCR-HC. These results suggest that the combination of these two methods can be important to increase the number of individuals diagnosed. The DNA degradation or presence of inhibitors in the samples may explain this discrepancy in results.

5. CONCLUSION

We concluded that the PCR-HC is a valid alternative and choice to increase the detection of *T. cruzi* in patients in the chronic phase of infection. The association of this method with the PCR-BL increases the number of individuals with positive results and improves the diagnosis of the chronic phase and the identification of therapeutic failure. Despite the long time required for obtaining results by PCR-HC, this study opens possibilities for a more systematic analysis, in order to decrease the amount of time and verify if the association of real-time PCR with hemoculture can further increase the detection of *T. cruzi*.

Ethical Approval: This study was approved by Permanent Committee of Ethics in Research Involving Human Beings (COPEP) of UEM, under protocol number 012/2010.

Conflicts of interest: The authors declare that they have no conflicts of interest.

ACKNOWLEDGMENTS

We are grateful to Coordination for the Improvement of Higher Education Personnel (CAPES) for a study scholarship. We thank the patients who agreed to participate of this work.

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ZIKA VIRUS IN PREGNANT WOMEN: EPIDEMIOLOGICAL ANALYSIS FROM JANUARY TO MAY 2016 AT IPATINGA, MINAS GERAIS, BRAZIL

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Received: 08/19/2016; Accepted: 10/22/2016

ABSTRACT

The Zika fever is the most recent arbovirus in Brazil, declared in November 2015 by the Ministry of Health (MOH) as a public health emergency. Since then there has been transmission of ZIKA virus (ZIKAV) in several Brazilian states. The maternal-fetal transmission of ZIKAV can be demonstrated throughout pregnancy, there is still no complete knowledge about the clinical manifestations of infection caused by ZIKAV, data on pregnant women infected by ZIKAV are limited. This is a quantitative and qualitative exploratory study, through desk research and literature. The public used in the study were 32 pregnant women confirmed with ZIKAV infection in the period from January to May 2016 in the city of Ipatinga, Minas Gerais. Data were analyzed using the SPSS program. The ZIKAV infection during pregnancy is considered one of the causes of microcephaly, and congenital anomalies. The diagnosing by ZIKAV infection is performed by RT-PCR technique, available free exam for pregnant woman's through the Unified Health System (UHS). In the study period there were no cases of microcephaly related to ZIKAV infection despite the high number of confirmed cases in Ipatinga city. The most effective way of prevention is the combating *Aedes* mosquito outbreaks. The aim was to discuss the main features of ZIKAV, knowing the complications related to infection during pregnancy, present epidemiological data of pregnant women who were infected with the virus during pregnancy and which were notified and confirmed with the ZIKAV by UHS in the city of Ipatinga, Minas Gerais in the period of January to May 2016, and describes the measures to prevent and control vector.

KEYWORDS: Zika, gestation, transmission, microcephaly, epidemiology.

1. INTRODUCTION

The Zika fever is the latest arbovirus in Brazil. It is a disease caused by virus Zika (ZIKAV) originating from Africa, was first isolated in 1947 in Zika forest, Uganda from the serum sample from a monkey *Rhesus*^{1,2}.

The first transmission of ZIKAV on the American continent was identified in Brazil in May 2015. The virus circulation in Brazil's Northeast was confirmed from the viral isolation in suspected cases of Dengue^{1,2}.

The Ministry of Health declared public health emergency in November 2015; then the World Health Organization has identified the situation as an emergency of international concern in February 2016, encouraging initiatives for research and disease control in Brazil³.

Ever since there has been transmission of ZIKAV in several Brazilian states, expanding to countries in the Americas and Europe. On February 18, 2016 cases of the disease had been confirmed in more than 32 countries and territories around the world^{2,3}.

Regions infested by *Aedes aegypti*, the main vectors of the disease, can contribute to the circulation of ZIKAV and other arboviruses simultaneously as dengue (DENV) and Chikungunya (CHIKV)³.

The aim of this study was to discuss the main features of ZIKAV, knowing the complications related to infection during pregnancy, present epidemiological data of pregnant women who were infected with the virus during pregnancy and which were notified and confirmed by the National Health System (SUS) in the Ipatinga city – Minas Gerais State, Brazil, over the period January to May 2016, and also describes the measures for prevention and control.

2. MATERIAL AND METHODS

This is a quantitative and qualitative exploratory study, through document search and literature review. The study of the universe is composed of 32 pregnant women reported and confirmed with involvement by Zika virus in the period from January to May 2016 in the

Ipatinga city, Minas Gerais, Brazil. The collection of epidemiological data such as number of reported cases and confirmed cases during the period from January to May 2016 were held in June 2016 in Epidemiological Surveillance sector of Ipatinga and the Ministry of Health website. Data were separated by categories, then most relevant variables were selected through IBM SPSS, using simple statistics. The literature review was made selecting national and international articles available online in full text dated after the year 2013, using the databases Google Scholar, PubMed and SciELO. The keywords used were: Zika virus, pregnant women, microcephaly, mechanism of action, transmission, screening, diagnosis and treatment. an analytical reading and selective was made of the information contained in the sources of interest according to the quality and relevance of the content to the topic in order to achieve the goals in research.

3. LITERATURE REVIEW

Zika virus

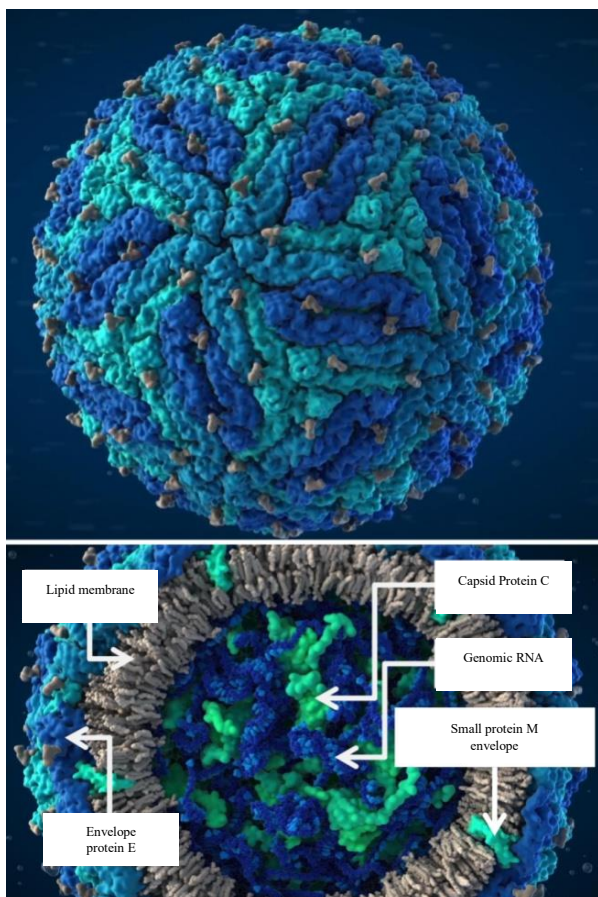


Figure 1. ZIKAV structural model in 3d. **Source:** <http://visual-science.com/projects/zika/3d-model/>.

The ZIKAV is an emerging arbovirus, belongs to the genus *Flavivirus*, family *Flaviviridae*^{1,4}. The ZIKAV is enveloped, it has icosahedral capsid; its genome is composed of single-stranded RNA and positive polarity with approximately 10.8 kilo-size bases. The genome contains a region 5' and 3' untranslated yielding a coding region that synthesizes the structural proteins the capsid (C), pre-membrane/ membrane (prM) and envelope (E) (Figure 1) and 7 non-protein structural (NS1, NS2a, NS2B, NS3, NS4A, NS4B and NS5). These are proteins that coordinate the viral replication and inhibit the response of the immune system^{5,6}.

To replicate, the ZIKAV binds to the cell membrane, the cell releases its RNA which happens to be processed by the cellular machinery for the production of viral proteins and new viruses (Figure 2)^{5,6}.

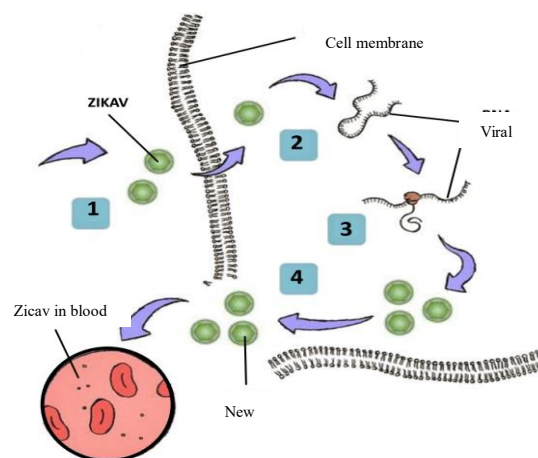


Figure 2. Simplified scheme of ZIKAV replication. (1) ZIKAV enters the bloodstream; (2) Within the cell, the virus releases its RNA, (3) that is processed by cellular machinery (4) generating new ZIKAV that disrupt the cell, being free in the circulation. **Source:** <http://www.nano-macro.com/2015/05/zika.html>.

Transmission

The ZIKAV is predominantly transmitted by blood-sucking mosquito bite of *Aedes* genus and may be of the species *Aedes aegypti* or *Aedes albopictus* (Figure 3)^{1,4,7}.

There are other forms of transmission and may be of mother to child (intrauterine); blood transfusions and through sexual intercourse. The ZIKAV has been detected in semen, blood, urine, amniotic fluid, saliva and bodily fluids found in the brain and spinal cord^{8,9}.

During the first weeks of ZIKAV infection, the virus can be isolated from the bloodstream. When the mosquito bites a human infected with viremia during this period, it acquires the virus can transmit to others¹⁰.

The *Aedes* mosquitoes that carry the ZIKAV feed on human blood preferably, and can sting multiple people in a single meal. They live in close proximity to human

habitations. The bites occur mainly during the day, and usually lay eggs both in and close to places with standing water; They can be found in most of the Americas^{10,11}.

The maternal-fetal transmission of ZIKAV was demonstrated throughout pregnancy. The full spectrum of results that may be associated with congenital infection is unknown¹².

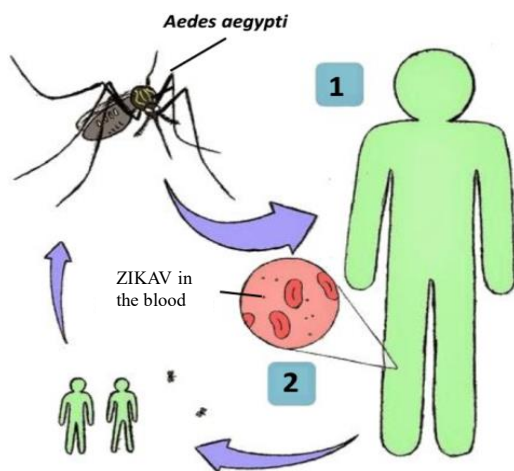


Figure 3. Simplified scheme of transmission ZIKAV by *Aedes aegypti*. (1) The *Aedes aegypti* mosquito bites an infected person and acquires the virus; (2) transmit the virus through bites another person. **Source:** <http://www.nano-macro.com/2015/05/zika.html>.

It is believed that the vertical transmission of ZIKAV occurs similarly to other TORCHS (transmitted infections fetus). The layer of syncytiotrophoblast cells (SYN) isolated from post-partum placenta showed extensive resistance to viral action, it seems likely that the fetus viruses go through pathways that do not involve replication in this cell layer SYN¹³.

As suggested by a recent study on the ZIKAV held in placental cells in the first and second trimester of pregnancy, the extravillous trophoblast (TEV) can be an entry portal for microorganisms, because it enables them to overcome the SYN barrier. However, it is unclear how pathogens reach these cells. As TEV are hidden in the decidua basalis, possibly, these cells are exposed to maternal blood and/ or infected immune cells during the vascularization^{12,13}.

Although SYNs isolated from postpartum placenta and SYNs exposed to the virus from the first quarter to be resistant to infection ZIKAV, it is possible that the ZIKAV to replicate in the early stages of pregnancy, or has other placental cells as a target. The ZIKAV RNA has been detected in placenta, fetal microcephaly^{10,13}.

The infection ZIKAV has an incubation period of three to twelve days¹⁴.

Zika virus during pregnancy

About the clinical manifestations of infection caused by ZIKAV, there is still no complete knowledge. It can be

considered that the infection is benign, but in Brazil have been registered many cases of Guillain-Barré Syndrome (GBS) days after development of clinical infection¹.

The data on pregnant women infected ZIKAV are limited. There is no evidence to suggest that pregnant women are more susceptible to infection, or are more severely affected¹².

Pregnant women can be infected ZIKAV in any trimester of pregnancy, and the reported symptoms during pregnancy are similar to non-pregnant individuals. May have mild fever (or absence of fever), rash, arthralgia (joint pain), itching in the body and conjunctivitis. The disease is self-limited and lasts up to a week. Severe cases are unusual^{12,14,15,16}.

The ZIKAV infection during pregnancy (Figure 5) is considered one of the causes of microcephaly, congenital defect wherein the head and the baby's brain are lower than expected in comparison with babies of the same age and sex¹⁷.

In addition to microcephaly, they were found other problems in fetuses infected ZIKAV before birth, such as eye defects, hearing loss and impaired growth^{12,17}.

There are reports that during pregnancy can occur placental insufficiency, intrauterine growth restriction, cerebellar atrophy, ventricular dilatation and intracranial calcifications. Congenital anomalies induced ZIKAV as neurological and eye disorders may present even without microcephaly^{12,13,17}.

Based on the available evidence it is believed that non-pregnant women who has infected ZIKAV, is not at risk of birth defects in future pregnancies¹⁷.

Scientists are collecting data to better understand the extensions and the impact of infection ZIKAV in pregnant women in order to clarify potential health problems that can cause infection^{12,17}.

Diagnosis

The diagnosis of infection ZIKAV can be performed by clinical examination; serology, through the identification of IgG and IgM antibodies; addition to the RT-PCR technique of molecular biology. Currently, only the RT-PCR technique is available^{14,18}.

The RT-PCR is a method used to identify the virus in early infection stage. Amplification of the genetic material (RNA) virus from patient samples allows to check the presence or absence of the individual ZIKAV^{12,14,18}.

It is necessary to perform a differential diagnosis of people infected with dengue virus due to similarities of symptoms, in order to establish appropriate measures for each case^{14,18}.

Flowchart diagnosis by the Unified Health System in Ipatinga, MG

The care of pregnant women with suspected infection ZIKAV the Unified Health System (SUS) begins with the

attendance of pregnant women to the Unit Basic Health (UBS) nearest your home; where health professionals will follow to diagnostic and monitoring measures^{19,20}.

Pregnant without rash is referred for prenatal care in primary health care; while pregnant with reporting or rash signs of infection, rash or fever without apparent cause is served by the health team of the unit which will evaluate other infectious disease; performing differential diagnosis for syphilis, HIV, toxoplasmosis, rubella. The rash onset date is noted in the pregnant woman's card, and the suspected case is reported in Zika form. It is filled also a mirror record of medical records, with the pregnant woman's data, which is attached to the notification form and forwarded to the Municipal Epidemiological Surveillance (VIEP)^{19,20}.

Then the mother is conducted to collect material for viral isolation (blood or urine) in the municipal laboratory. The samples collected are directed to clinical laboratory of Ezequiel Dias Foundation (FUNED). The embodiment of obstetric Ultrasound may be important for the morphological analysis of the fetal skull and displaying internal structures of the brain. If the ultrasound detect morphological or microcephaly change, the pregnant woman is sent to a referral service^{20,21}.

When the result of viral isolation for ZIKAV is negative, it is necessary to investigate other causes of symptoms through prenatal care in primary care. The positive results for ZIKAV are available by VIEP, and the mother is directed to consult with an infectious disease physician, who may order tests at its discretion; and consultation in prenatal high risk, which will follow up the monitoring of gestational development^{20,21}.

Before the release of the viral isolation result there is a connection with the team of psychologists, nutritionists, psychiatrists, gynecologists and obstetricians of the "Support Center for Health" (NASF) to support pregnant women awaiting the results, with priority focus in prevention and promotion of physical and mental health^{19,20}.

4. RESULTS AND DISCUSSION

In the period January-May 2016 were 13,973 reported cases of suspected infection of pregnant women ZIKAV in Brazil, with 5,925 laboratory confirmed cases (42.4%). There were 1,551 confirmed cases for microcephaly and/or changes in the central nervous system (CNS) suggestive of congenital infection²².

In Minas Gerais, in the same period, it was reported 877 suspected cases, corresponding to 6.27% of the cases in relation to Brazil. laboratory were confirmed 230 cases (26.23%), corresponding to 3.88% of the cases in relation to Brazil. Of the cases reported in Minas Gerais, 5.25% were discarded because they had negative results for infection ZIKAV and 68.52% were still under investigation. There have been three confirmed cases of micro-

cephaly and/ or changes in the CNS suggestive of congenital infection²².

According to Table 1 of frequencies you can see that in Minas Gerais most cities had 1 case; 85.4% had number of cases less than or equal to 7. As a city presented many cases greater than 40.

Table 1. Frequency of cases of pregnant women affected by ZIKAV by city in the state of Minas Gerais in the period from January to May 2016.

Number of cases	F	%	% accumulative
1	24	50	50
2	11	22,9	72,9
3	3	6,3	79,2
4	2	4,2	83,3
7	1	2,1	85,4
10	1	2,1	87,5
13	1	2,1	89,6
15	1	2,1	91,7
25	1	2,1	93,8
28	2	4,2	97,9
41	1	2,1	100
Total	48	100	

Source: The authors, 2016.

The municipalities of Minas Gerais State with the highest number of laboratory confirmed cases were Montes Claros (17.83%), Belo Horizonte (12.17%), Sete Lagoas (12.17%) and Ipatinga (10.87%)^{20,22}.

Physical and environmental characteristics may explain the epidemic observed in the study area. Ipatinga for example, it is located in a region of high average temperatures and humidity, conditions that can contribute to the reproduction of the Aedes mosquito in periods of rain, and consequently favor the transferability not only ZIKAV, but also of the Dengue virus (DENV) and Chikungunya virus (CHIKV)²³.

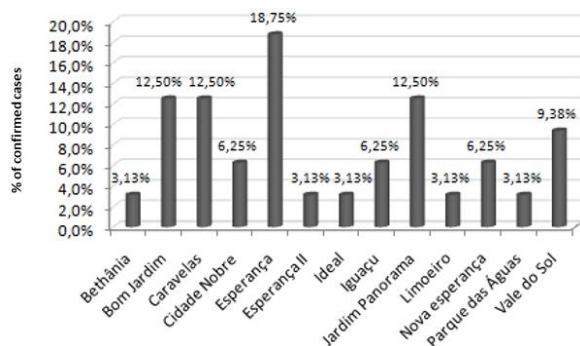
The standard deviation of Minas Gerais (Table 2) was higher compared to the Ipatinga, this means that the number of cases of pregnant women with confirmed ZIKAV in Ipatinga are closer to the average when compared to Minas Gerais. Analyzing the maximum and minimum value of cases it is noticed that in Ipatinga the number of cases per district varied from 1 to 6 cases, while in Minas Gerais the number of cases per town ranged from 1 to 41 cases.

In Ipatinga, between January and May 2016 were reported 141 cases of pregnant women suspected infection ZIKAV, with laboratory confirmed 32 cases (22.7%) distributed in 13 districts of the municipality, as shown in Figure 1; and there were no confirmed cases of microcephaly related to infection ZIKAV the study period.

Table 2. Statistical data of pregnant women affected by ZIKAV in Ipatinga (in neighborhoods) and Minas Gerais (for cities) in the period January to May 2016.

	Ipatinga	Minas Gerais
N	13	48
Mean	2,46	4,79
Median	2	1,5
Mode	1	1
Standard deviation	1,613	8,558
Interval	5	40
Minimum	1	1
Maximum	6	41
Percentile		
25	1	1
50	2	1,5
75	4	3

Source: The Authors, 2016.

**Figure 1.** Distribution neighborhood confirmed cases of ZIKAV in pregnant women in Ipatinga, Minas Gerais, in the period from January to May 2016. Fonte: The Authors, 2016.

The Esperança neighborhood had a higher number of confirmed pregnant women with laboratory infection ZIKAV, followed by the districts Bom Jardim, Caravelas and Jardim Panorama showed 4 cases each; and Sun Valley, with 3 cases. On Table 3 it was found that 92.3% of the number of cases neighborhoods have less than or equal to 4^{20,22}.

Table 3. Frequency of cases by pregnant neighborhood affected by ZIKAV in Ipatinga, in the period January-May 2016.

Number of cases	F	%	% accumulative
1	5	38,5	38,5
2	3	23,1	61,5
3	1	7,7	69,2
4	3	23,1	92,3
6	1	7,7	100
Total	13	100	

Source: The Authors, 2016.

The neighborhoods that had the highest number of cases are populated and are in areas with high rainfall, urban infrastructure deficiencies, low socioeconomic conditions, resulting in ideal places for mosquito vector populations of breeding, favoring their survival throughout the year and the rapid transmission of arboviruses²³.

Treatment, prevention and control

Treatment for ZIKAV is symptomatic. There are only treatment for relief of symptoms using analgesics and antipyretics, anti-inflammatories for reducing pain in joints and muscles, eye drops three to six times daily as lubricant, antiallergic drugs. It is important to rest and stand for seven days and eat foods rich in minerals and vitamins, drink plenty of fluids for fast recovery¹⁸.

The bloodsucking mosquitoes of the genus *Aedes* have shown extraordinary capacity for biological adaptation making it difficult to extinction. The fight against these mosquitoes is highly recommended to avoid the incidence of new cases of infection ZIKAV. The awareness and mobilization of the population are essential to avoid environments conducive to the development of vectors²⁴.

Measures to eliminate areas where mosquitoes develop is to cover water tanks with screens or covers, preventing access vector; musketeers use in windows and doors; regularly clean the roof gutters, do not leave bottles, gallons, tires and any objects that hold water exposed to rain^{24,25}.

Individual protection should be carried out through the use of repellent on exposed skin and clothing, particularly in pregnant women. The use of clothes that minimize skin exposure is recommended to provide protection against bites during the day when mosquitoes are most active^{24,25}.

There is no vaccine against the disease. The most effective form of prevention is to combat the mosquito *Aedes* outbreaks typical of urban areas of tropical and subtropical climate and use of repellents¹⁸.

It is essential the participation of society together with government agencies in the battle against the mosquito. A vector control program is able to reduce the probability of a viremic human blood serve as power supply for the *Aedes aegypti* and *Aedes albopictus*²⁵.

5. CONCLUSION

Through the research conducted, it was observed that the Zika fever is an arbovirus that recently arrived in the Americas, especially in areas infested by *Aedes aegypti*, one of the main vectors that can contribute to the circulation of ZIKAV and other arboviral diseases such as dengue and Chikungunya.

The media has widely reported the increasing number of suspected and confirmed cases of pregnant women

affected by ZIKAV in Brazil and the possible complications related to the virus to the fetus, such as microcephaly, eye abnormalities and neurological disorders. Studies are still under development to understand the real impact and the extent of infection, seeking to clarify potential health problems that can cause infection.

It is not known for sure what the means of transmission of ZIKAV, studies point to have isolated the virus in semen, blood, urine, amniotic fluid, saliva, and other bodily fluids. It is believed that the mother-to-child transmission can occur at any gestational period.

The consultation of the pregnant woman to the doctor is very important for the diagnosis, monitoring gestational development and fetal development, as well as guidance for prevention of other diseases.

Due to the high cost to perform the RT-PCR molecular biology, it is necessary to develop more affordable diagnostic methods in order to expand the investigation of cases in the general population; not only in pregnant women and fetuses.

In Brazil, laboratory were confirmed 5,925 cases of pregnant women affected by ZIKAV the study period, occurring 1,551 confirmed cases of microcephaly. In Minas Gerais it was confirmed only 3 cases of microcephaly associated with infection ZIKAV. However, in Ipatinga, despite the high number of confirmed cases of affected pregnant women, there were no cases of microcephaly related ZIKAV the study period, which does not rule out the possibility of the occurrence of cases in the future.

There are no vaccines against the disease, prevention should be held collectively and individually, limiting the spread of mosquito vectors, which are common in urban areas of tropical and subtropical climate, using repellents, covering water tanks, sanitation, do not leave water accumulated in tires, roofs, gutters.

Therefore, efforts are needed to further studies clarify the extent and impact of infection, in order to guide researchers in the development of vaccines and other medicines to contribute to the health of pregnant women, fetuses, and elsewhere, preventing the incidence of new neurological cases of microcephaly and involvements related to ZIKAV.

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