INTRODUCTION

Isolated or nonsyndromic oral clefts, including cleft lip (CL), CL and/or palate (CL and/or P), and isolated cleft palate (CP), are one of the most common congenital malformations in humans. They have geographic and racial variations and seem to occur as a result of a complex interaction between genetic variations and environmental factors. Since environmental factors play a role of major importance in the etiology of oral clefts, the purpose of this study was to investigate its relation with the occurrence of nonsyndromic oral cleft patients in a Brazilian subpopulation.

MATERIALS AND METHODS

A cross-sectional study was conducted at the Craniofacial Rehabilitation Center in the General Hospital of the University of Cuiabá (HGU/UNIC), Cuiabá city, Mato Grosso, Brazil.

From the total of 776 oral cleft patients treated at the HGU/UNIC from 2004 to 2012, 200 patients were selected for this study by fulfilling the inclusion criteria that were: Being isolated or nonsyndromic oral cleft patients and younger than 6 years old. Patients were contacted by telephone and letter and attended to HGU/UNIC 116 volunteers.

The research project was approved by the Ethics Committee in Research of the University of Cuiabá (protocol of approval number: 2012-003) before its beginning. The aim of the study was carefully explained to the patients’ parents, and their formal consent was obtained. Procedures followed

ABSTRACT

Background: A cross-sectional study was conducted at the Craniofacial Rehabilitation Center in the General Hospital of the University of Cuiabá, Cuiabá city, Mato Grosso, Brazil.

Materials and Methods: Poisson regression model was used to analyze the relationship between antenatal factors and the occurrence of oral clefts in 116 patients.

Results: Oral clefts were more common in males (64.66%) and White race (46.02%). The mean age of the children was 21.91 months. The most common type of cleft was cleft lip and palate (CLP, 55.17%). Maternal and paternal smoking in the first trimester of pregnancy and parity were significantly associated with the occurrence of CLP. Parent’s age, educational level, and occupation did not interfere in the occurrence of oral clefts. There was also no significant association between maternal illness, medication use, alcohol consumption, and maternal exposure to chemicals in the first trimester of pregnancy and the occurrence of clefts in this population.

Conclusion: The analysis of the environmental factors present during the pregnancy of children with oral clefts revealed a significant association between parity (second onward), maternal smoking, and paternal smoking and the occurrence of CL and/or palate in this population.

Key words: Cleft lip, cleft palate, epidemiology, public health, risk factors

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were in accordance with the Helsinki declaration of 1975 as revised in 2000. Confidentiality of subjects was ensured by desisting from mentioning participants’ names, initials, or hospital numbers.

A questionnaire was developed and administered by two interviewers supervised by the principal investigator aiming to collect the following information: Sociodemographic profile of the patients and their parents; oral cleft type, age of the parents during pregnancy; number of pregnancies and prenatal events such as maternal health problems, drug use, alcohol use, maternal smoking, and paternal drug use, contact with chemicals and exposure to ionizing radiation.

Descriptive and inferential statistics were used for data analysis. In the descriptive part, absolute and relative frequencies were presented. In the inferential part, measures of association were determined between the dependent variables and the independent or explanatory variables using Chi-square, Fisher’s exact test, and likelihood ratio test with a significance level of 0.05. The prevalence ratio was also obtained with its respective confidence intervals of 95% and the association between variables. The variables with \( P < 0.20 \) were considered for the construction of a multivariate Poisson regression model, with confidence intervals of 95% and \( \text{P-value} \) selected variables by backward method. The variables with a significance level lower than 0.05 remained in the final model. Statistical analyses were performed with SPSS Version 15.0 (SPSS Inc., Chicago, IL, USA), Minitab Version 15.0 (Minitab Inc., State College, PA, USA), and STATA Version 10.0 (StataCorp LP, College Station, TX, USA).

### RESULTS

The cleft lip and palate (CLP) was the most common feature with 64 cases (55.17%), followed by CL with 30 (25.86%) and CP with 22 (18.97%) cases.

Distribution of patients according to sex and age is presented in Table 1. Patients were distributed according to their parent-reported race/color of the skin as White \( (n = 52, 46.02\%) \), Black \( (n = 22, 19.47\%) \), Brown \( (n = 39, 34.51\%) \), and Native American \( (n = 3, 2.60\%) \). According to the order of birth, 46 patients \( (39.66\%) \) were the fruit of the first pregnancy, 37 \( (31.90\%) \) the second pregnancy, and 33 \( (28.45\%) \) the third or other pregnancy.

The parental age during pregnancy is presented in Table 2 and parental schooling divided into illiterate, elementary school, middle school, and higher education is presented in Table 3.

Maternal occupation was divided into housekeeping, service, commerce, student, education, and other. Paternal occupation was divided into service, rural, commerce, industry, education, student, and other. Parental occupation is presented in Table 4.
Regarding changes in maternal health during the first trimester of pregnancy, it was found that 42 (36.21%) mothers of the 116 had infection, 14 (12.07%) had hypertension, and 4 (3.45%) had diabetes.

Concerning the use of medication during the first trimester of pregnancy, 39 mothers (33.62%) reported having used painkillers; 39 (33.62%) administered antibiotics; 4 (3.45%) used benzodiazepines; 3 (2.59%) used anticonvulsants; 3 (2.59%) used anti-inflammatory, and 6 (5.17%) said that they had used homemade medicines. One mother (0.86%) used corticosteroids during pregnancy. From the total mothers, 56 (48.28%) used folic acid and 64 (55.17%) used vitamins during pregnancy.

When asked about alcohol consumption during pregnancy, 22 mothers (17.24%) reported having done so. As for smoking, 9 mothers (7.76%) had this habit during pregnancy. It was also questioned about paternal smoking which was reported in 27 cases (23.28%). Illegal drug use was reported by 2 mothers (1.72%).

The contact of mothers with chemicals during pregnancy was also investigated and 32 (27.59%) said that they had contact with these products. The most frequently reported were cleaning products (n = 13, 11.30%), pesticides (n = 9, 7.83%), cosmetics (n = 7, 6.09%), and other (n = 3, 2.61%). Eight mothers (6.96%) were exposed to ionizing radiation during pregnancy.

CL and CLP were considered a single group for the inferential analysis (CL and/or P). Chi-square test, Fisher’s exact test, and likelihood ratio test were applied to verify the relationship between the studied environmental factors and the occurrence of CL and/or P or CP.

For the seven independent variables whose association value (P) with CL and/or P was <0.20 (race/color of skin, paternal age, maternal infection, maternal smoking, paternal smoking, contact with chemicals during pregnancy, and parity order), the prevalence ratio was obtained [Table 5].

The prevalence ratio was adjusted with the robust Poisson regression model with confidence interval of 95%. The results showed that maternal smoking (P < 0.001), paternal smoking (P = 0.042), and parity (second or over) (P = 0.042) were associated with the occurrence of CL and/or P. The race/color of the skin variable, although not statistically significant at the 0.05 level, remained in the model as an adjustment variable [Table 6].

**DISCUSSION**

Mato Grosso is the third largest state of Brazil located in the central-west region with the population of 3,035,122 inhabitants. Since the late 20th century, it is the national champion of grain, cattle, and wood production. The Craniofacial Rehabilitation Center in HGU/UNIC was implanted in 2004 in Cuiabá, capital of Mato Grosso, as the first specialized center for the care of oral cleft patients in the state.\(^{21,22}\)

The population of this study comprised 64.66% of male patients. Although the study does not reflect the totality of Mato Grosso’s oral cleft patients, this result is similar to previous studies that indicate greater incidence of clefts in men.\(^{16,9,23-26}\)

Among the study population, 46.02% were White, 34.51% Brown, 19.47% Black, and 2.60% Native American. Previous studies conducted in different localities also found oral cleft predilection for Caucasians and lower frequency in Afro-descendent population.\(^{3,23,24,27}\)

For the inferential analysis, CL and CLP were considered as one group (CL and/or P) as they are etiologically,
Among the variables related to the parents of children with clefts, we evaluated the age of the parents at the beginning of pregnancy. Maternal and paternal ages have been investigated as possible risk factors for the occurrence of CLP. Herkrath et al. conducted a meta-analysis analyzing parental age as a risk factor for CLP. They demonstrated that parents aged 40 or older had an increased risk of generating a child with CP and mothers aged 35–39 years were more likely to produce a child with CP. Mothers aged 40 or older had an increased risk of generating a child with CL and/or P. They also reported that no evidence was found associating younger parents with increased risk to the occurrence of clefts. In our study, 60.34% of mothers were aged between 20 and 34 years and 82.76% of the parents were aged between 20 and 39 years at the start of pregnancy, contrasting the results that suggest an association between advanced age of the parents and the occurrence of oral clefts. This study found no association between maternal and paternal age and the occurrence of CL and/or P or CP.

It was observed that 39.66% of the cleft patients were the result of the first pregnancy and 60.35% of the second pregnancy onward. It was suggested that the greater the number of pregnancies, greater the risk of a woman having a child with cleft perhaps because an excessive number of abortions could lead to damage or dysfunction in the female reproductive tract. Messer et al. demonstrated that a higher proportion of cleft cases was generated by women who had three or more previous pregnancies in Texas between 1999 and 2003. A case–control study in China with 713 cases of nonsyndromic oral clefts showed that the highest proportion of cases originated from the second pregnancy onward, similar to the result of this study, found significant association in the occurrence of CL and/or P in children born in the second or posterior pregnancies in the adjusted Poisson regression model (P = 0.042).

Regarding parental education, it was observed that mothers had more years of study than fathers since 56.03% of them had middle school education against 43.10% of men. It is noteworthy that only 11.21% of mothers and 12.07% of fathers had higher education. It has been shown in previous studies that parents of oral cleft children tend to have lower educational level.

The parental occupation variable was analyzed in this study because there could be a parental occupational exposure to different agents during pregnancy, favoring the development of congenital anomalies such as oral clefts. However, such association was not established in this study.

The association of oral clefts and the use of certain drugs during pregnancy have been suggested over the past decades. Medications such as anticonvulsants, benzodiazepines, and corticosteroids have been investigated. However, the association of these drugs with the occurrence of oral clefts is still inconclusive since many of these studies limited by a lack of statistical power showed weak evidence of association or even absence of association. In the present study, mothers reported the use of analgesics, antibiotics, benzodiazepines, anticonvulsants, and anti-inflammatorer. The use of home remedies was also questioned and although Omo-Aghoja et al. have found an increased relative risk to CP and unilateral CLP with the use of herbal medicines, this study found only 5.17% reported using home remedies during pregnancy.

On the other hand, the use of folic acid and vitamin supplements during pregnancy has been inversely related to the occurrence of oral clefts, but this relation still has consistency limitations and the mechanism of how this protective effect occurs is still unknown. In this study, it was observed that 55.17% of mothers administered vitamin supplements during pregnancy and 48.28% administered folic acid. This is an important finding because it contradicts an important public health strategy since the preventive administration of folic acid is also associated with a lower incidence of neural tube defects and congenital cardiac anomalies. It was recently shown that the quality of maternal diet could also be associated with a reduction in the risk of neural tube defects and oral clefts even more consistently than maternal administration of isolated nutrients.

A positive association between alcohol consumption during pregnancy and occurrence of oral clefts has been shown in several studies, but not in all. This inconsistency in the results may be due to the need of excessive consumption of alcohol for the expression of the anomaly, which is uncommon in pregnancy, thus having a small number of exposed women. In this study, 17.24% of the mothers reported any alcohol consumption in the first trimester of pregnancy, and there was no association with the occurrence of oral clefts.

We also assessed maternal smoking during the first trimester of pregnancy and paternal smoking. The association between maternal smoking and oral clefts has been investigated in many studies that have suggested a modest association. In this study, we found a significant association between maternal smoking and occurrence of CL and/or P in the adjusted Poisson regression model (P < 0.001) as in several studies. Likewise, paternal smoking in this study has also been associated with the occurrence of CLP (P = 0.042), confirming the results found by Jia et al. and Zhang et al.
Considering maternal contact with chemical agents in the first trimester of pregnancy, we found that 27.59% of the mothers came into contact with these products being 11.30% with cleaning agents, 7.83% with pesticides, and 6.09% with cosmetics. Garlantézec et al.\textsuperscript{18} showed that maternal exposure to solvents is associated with the occurrence of oral clefts, these solvents being present in cleaning agents and cosmetics and other products. As for pesticides, for decades, their relationship with birth defects including CLP has been investigated. Despite scientific evidence suggesting this association, it is not conclusive.\textsuperscript{14,19,34,49} Recent studies in the State of Mato Grosso showed the relationship between the use of pesticides and the occurrence of congenital malformations,\textsuperscript{50,51} the association, however, was not observed in this study.

CONCLUSION

The analysis of the environmental factors present during the pregnancy of children with oral clefts revealed a significant association between parity (second onward), maternal smoking, and paternal smoking and the occurrence of CL and/or P in this population.

The present data may subside oral clefts prevention and counseling programs.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Zhang B, Jiao X, Mao L, Xue J. Maternal cigarette smoking and the associated risk of having a child with orofacial clefts in China: A case-control study. J Craniofac Surg 2011;39:313-8.
2. Murray JC. Gene/environment causes of cleft lip and/or palate. Clin Genet 2002;61:248-56.
3. Herkath AP, Herkath FJ, Rebelo MA, Vettore MV. Parental age as a risk factor for non-syndromic oral clefts: A meta-analysis. J Dent 2012;40:3-14.
4. Stanier P, Moore GE. Genetics of cleft lip and palate: Syndromic genes contribute to the incidence of non-syndromic clefts. Hum Mol Genet 2004;13:R73-81.
5. Dixon MJ, Marariza ML, Beaty TH, Murray JC. Cleft lip and palate: Understanding genetic and environmental influences. Nat Rev Genet 2011;12:167-78.
6. Honein MA, Rasmussen SA, Reefhuis J, Romitti PA, Lammer EJ, Sun L, \textit{et al}. Maternal smoking and environmental tobacco smoke exposure and the risk of orofacial clefts. Epidemiology 2007;18:226-33.
7. Shi M, Christensen K, Weinberg CR, Romitti P, Bathum L, Lozada A, \textit{et al}. Orofacial cleft risk is increased with maternal smoking and specific detoxification-gene variants. Am J Hum Genet 2007;80:76-90.
8. Leite IC, Koifman S. Oral clefts, consanguinity, parental tobacco and alcohol use: A case-control study in Rio de Janeiro, Brazil. Braz Oral Res 2009;23:31-7.
9. Lebby KD, Tan F, Brown CF. Maternal factors and disparities associated with oral clefts. Ethn Dis 2010;20 1 Suppl 1:S146-9.
10. DeRoo LA, Wilcox AJ, Drevon CA, Lie RT. First-trimester maternal alcohol consumption and the risk of infant oral clefts in Norway: A population-based case-control study. Am J Epidemiol 2008;168:638-46.
11. Boyles AL, DeRoo LA, Lie RT, Taylor JA, Jugessur A, Murray JC, \textit{et al}. Maternal alcohol consumption, alcohol metabolism genes, and the risk of oral clefts: A population-based case-control study in Norway, 1996-2001. Am J Epidemiol 2010;172:924-31.
12. Krapels IP, van Rooij IA, Ocké MC, West CE, van der Horst CM, Steegers-Theunissen RP. Maternal nutritional status and the risk for orofacial cleft offspring in humans. J Nutr 2004;134:3106-13.
13. van Baynum IM, Kapusta L, Bakker MK, den Heijer M, Blom HJ, de Walle HE. Protective effect of periconceptional folic acid supplements on the risk of congenital heart defects: A registry-based case-control study in the northern Netherlands. Eur Heart J 2010;31:464-71.
14. Leite IC, Paumgarten FJ, Koifman S. Chemical exposure during pregnancy and oral clefts in newborns. Cad Saúde Pública 2002;18:17-31.
15. Hviid A, Melgaard-Nielsen D. Corticosteroid use during pregnancy and risk of orofacial clefts. CMAJ 2011;183:796-804.
16. Wallace GH, Arellano JM, Gruner TM. Non-syndromic cleft lip and palate: Could stress be a causal factor? Women Birth 2011;24:40-8.
17. Koifman S, Koifman RJ, Meyer A. Human reproductive system disturbances and pesticide exposure in Brazil. Cad Saúde Pública 2002;18:435-48.
18. Garlantézec R, Monfort C, Rouget F, Cordier S, Maternal occupational exposure to solvents and congenital malformations: A prospective study in the general population. Occup Environ Med 2009;66:456-63.
19. Shirangi A, Nieuwenhuysen M, Vienneau D, Holman CD. Living near agricultural pesticide applications and the risk of adverse reproductive outcomes: A review of the literature. Paediatr Perinat Epidemiol 2011;25:172-91.
20. Marshall EG, Harris G, Wartenberg D. Oral cleft defects and maternal exposure to ambient air pollutants in New Jersey. Birth Defects Res A Clin Mol Teratol 2008;80:205-15.
21. Khalil W, da Silva HL, Serafim KT, Volpato LE, Casela LF, Aranha AM. Recovering the personal identity of an elderly patient with cleft lip: A case report. Spec Care Dentist 2012;32:218-22.
22. Cuozzo FD, Espinosa MM, da Silva KT, de Barros YB, Bandeira MC, Aranha AM, \textit{et al}. Cleft lip and palate in a Brazilian subpopulation. J Int Oral Health 2013;5:15-20.
23. Martelli-Junior H, Porto LV, Martelli DR, Bonan PR, Freitas AB, Della Coletta R. Prevalence of nonsyndromic oral clefts in a reference hospital in the state of Minas Gerais, Brazil, between 2000-2005. Braz Oral Res 2007;21:314-7.
24. Martelli DR, Bonan PR, Soares MC, Paranaiba LR, Martelli-Júnior H. Analysis of familial incidence of non-syndromic cleft lip and palate in a Brazilian population. Med Oral Patol Oral Cir Bucal 2010;15:e898-901.
25. Jia ZL, Shi B, Chen CH, Shi JY, Wu J, Xu X. Maternal malnutrition, environmental exposure during pregnancy and the risk of non-syndromic orofacial clefts. Oral Dis 2011;17:584-9.
26. Yañez-Vico RM, Iglesias-Linares A, Gómez-Mendoza I, Torres-Lagares D, González-Moles MÁ, Gutierrez-Pérez JL, \textit{et al}. A descriptive epidemiologic study of cleft lip and palate in Spain. Oral Surg Oral Med Oral Pathol Oral Radiol 2012;114 5 Suppl:S1-4.
27. Poletta FA, Castella EE, Orioli IM, Lopez-Camelio JS. Regional analysis on the occurrence of oral clefts in South America. Am J Med Genet A 2007;143A:3218-27.
28. Sivertsen A, Wilcox AJ, Skjaerven R, Vindersen HA, Abyholm F, Harville E, et al. Familial risk of oral clefts by morphological type and severity: Population based cohort study of first degree relatives. BMJ 2008;336:432-4.
29. Bille C, Skytte A, Vach W, Knudsen LB, Andersen AM, Murray JC, et al. Parent’s age and the risk of oral clefts. Epidemiology 2005;16:311-6.
30. Messer LC, Luben TJ, Mendola P, Carozza SE, Horel SA, Langlois PH. Urban-rural residence and the occurrence of cleft lip and cleft palate in Texas, 1999-2003. Ann Epidemiol 2010;20:32-9.
31. Omo-Aghoja VW, Omo-Aghoja LO, Ugboko VI, Obuekwe ON, Saheeb BD, Feyi-Waboso P, et al. Antenatal determinants of oro-facial clefts in Southern Nigeria. Afr Health Sci 2010;10:31-9.
32. Lorente C, Cordier S, Bergeret A, De Walle HE, Goujard J, Aymé S, et al. Maternal occupational risk factors for oral clefts. Occupational Exposure and Congenital Malformation Working Group. Scand J Work Environ Health 2000;26:137-45.
33. Chevrier C, Dananché B, Bahuau M, Nelva A, Herman C, Francannet C, et al. Occupational exposure to organic solvent mixtures during pregnancy and the risk of non-syndromic oral clefts. Occup Environ Med 2006;63:617-23.
34. Thulstrup AM, Bonde JP. Maternal occupational exposure and risk of specific birth defects. Occup Med (Lond) 2006;56:532-43.
35. Saxén I. Cleft lip and palate in Finland: Parental histories, course of pregnancy and selected environmental factors. Int J Epidemiology 1974;3:263-70.
36. Safra MJ, Oakley GP Jr. Association between cleft lip with or without cleft palate and prenatal exposure to diazepam. Lancet 1975;2:478-80.
37. Hill L, Murphy M, McDowall M, Paul AH. Maternal drug histories and congenital malformations: Limb reduction defects and oral clefts. J Epidemiol Community Health 1988;42:1-7.
38. Carmichael SL, Shaw GM. Maternal corticosteroid use and risk of oral clefts. Am J Med Genet 1999;88:242-4.
39. Källén B. Maternal drug use and infant cleft lip/palate with special reference to corticoids. Cleft Palate Craniofac J 2003;40:624-8.
40. Pradat P, Robert-Gnansia E, Di Tanna GL, Rosano A, Lisi A, Mastroiacovo P; Contributors to the MADRE Database. First trimester exposure to corticosteroids and oral clefts. Birth Defects Res A Clin Mol Teratol 2003;67:968-70.
41. Werler MM, Ahrens KA, Bosco JL, Mitchell AA, Anderka MT, Gilboa SM, et al. Use of antiepileptic medications in pregnancy in relation to risks of birth defects. Ann Epidemiol 2011;21:842-50.
42. Carmichael SL, Shaw GM, Ma C, Werler MM, Rasmussen SA, Lammer EJ; National Birth Defects Prevention Study. Maternal corticosteroid use and orofacial clefts. Am J Obstet Gynecol 2007;197:585.e1-7.
43. Dolovich LR, Addis A, Vaillancourt JM, Power JD, Koren G, Einanson TR. Benzodiazepine use in pregnancy and major malformations or oral cleft: Meta-analysis of cohort and case-control studies. BMJ 1998;317:839-43.
44. Rosenettel, Mitchell AA, Piersellis JL, Pashayan H, Louik C, Shapiro S. Lack of relation of oral clefts to diazepam use during pregnancy. N Engl J Med 1983;309:1282-5.
45. Badovinac RL, Werler MM, Williams PL, Kelsey KT, Hayes C. Folic acid-containing supplement consumption during pregnancy and risk for oral clefts: A meta-analysis. Birth Defects Res A Clin Mol Teratol 2007;79:8-15.
46. Shaw GM, Carmichael SL, Laurent C, Rasmussen SA. Maternal nutrient intakes and risk of orofacial clefts. Epidemiology 2006;17:285-91.
47. Carmichael SL, Yang W, Feldkamp ML, Munger RG, Siega-Riz AM, Botto LD, et al. Reduced risks of neural tube defects and orofacial clefts with higher diet quality. Arch Pediatr Adolesc Med 2012;166:121-6.
48. Little J, Cardy A, Munger RG. Tobacco smoking and oral clefts: A meta-analysis. Bull World Health Organ 2004;82:213-8.
49. Gordon JE, Shy CM. Agricultural chemical use and congenital cleft lip and/or palate. Arch Environ Health 1981;36:213-21.
50. Pignati WA, Machado JM, Cabral JF. Major rural accident: The pesticide “rain” case in Lucas do Rio Verde city-MT. Cien Saude Colet 2007;12:105-14.
51. Oliveira NP, Moi GP, Atanaka-Santos M, Silva AMC, Pignati WA. Congenital defects in the cities with high use of pesticides in the state of Mato Grosso, Brazil. Ciênc. saúde coletiva 2014;19:4123-30.