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**Influence of breastfeeding and timing of gluten introduction on the onset of
celiac disease in infants**

Утицај дојења и времена увођења глутена на појаву целијачне болести код
деце узраста до две године

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Influence of breastfeeding and timing of gluten introduction on the onset of celiac disease in infants

Утицај дојења и времена увођења глутена на појаву целијачне болести код деце узраста до две године

SUMMARY

Introduction/Objective The classic type of celiac disease (CD) is most common in children under two years of age.

The aim of this study was to investigate whether breastfeeding, particularly breastfeeding during gluten introduction, and timing of gluten introduction, influence the onset of CD at this age.

Methods We retrospectively analyzed medical records of 93 children, 40 in the first and 53 in the second year, with a classic CD diagnosed at the University Children's Hospital, Belgrade between 2000 and 2010. Diagnose of CD was based on the criteria of the European Society for Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) from 1989.

Results Duration of breastfeeding reduced the onset of the CD in the first year $p = 0.039$ (OR = 1.43 95% CI 1.019–1.899). Also, breastfeeding at the time of gluten introduction significantly delayed the age at diagnosis ($F = 1.671$, $t = 2.39$, $p = 0.029$). The timing of gluten introduction did not affect the age of occurrence of CD in these group of children.

Conclusion Longer breastfeeding, and breastfeeding at the time of gluten introduction, postponed the onset of classic CD in patients up to 2 years. The association between the occurrence of CD and the time of introduction of gluten in this age group of patients has not been established.

Keywords: classic celiac disease; children up to 2 years; breastfeeding; age of gluten introduction

САЖЕТАК

Увод/Циљ Класични тип целијачне болести (ЦБ) се најчешће јавља код деце узраста до две године.

Циљ ове студије је био да се испита да ли дојење, посебно дојење током увођења глутена, и време увођења глутена, утичу на почетак ЦБ у овом добу.

Метод Ретроспективно смо анализирали медицинску документацију 93 деце, 40 у првој и 53 у другој години, са класичном формом ЦБ дијагностикованој у Универзитетској дечјој клиници у Београду између 2000. и 2010. године. Дијагноза ЦБ заснована је на критеријумима Европског удружења за дечју гастроентерологију, хепатологију и нутрицију (ESPGHAN) из 1989. године.

Резултати Трајање природне исхране значајно редукује појаву ЦБ у првој години живота ($p = 0,039$, $OR = 1,43$, $95\% CI = 1,019-1,899$). Такође, природна исхрана у време увођења глутена сигнификантно одлаже појаву болести ($F = 1,671$, $t = 2,39$, $p = 0,029$). Време увођења глутена није утицало на узраст појаве ЦБ у овој групи деце.

Закључак Природна исхрана, а посебно природна исхрана у време увођења глутена, одлаже појаву класичне ЦБ код деце узраста до две године. Повезаност појаве ЦД и времена увођења глутена у овој узрастој групи пацијената није доказана.

Кључне речи: класична целијачна болест; деца до две године; дојење; узраст увођења глутена

INTRODUCTION

Celiac disease (CD) is an immune-mediated systematic disease caused by the ingestion of gluten that appears in genetically predisposed individuals. The most important genetic factor is the human leukocyte antigen (HLA) locus DQ2 and HLA-DQ8 haplotypes, while the gluten, most important environmental factor, is required to trigger the disease [1, 2]. Other factors may contribute to the pathogenesis and expression of CD, namely additional genetic loci, gender, breastfeeding, timing of gluten introduction, gut microbiota, mode of delivery,

metabolic profile of patients, etc. [3]. The disease may be symptomatic, with the occurrence of gastrointestinal and non-gastrointestinal symptoms. In addition, the course of the disease may be asymptomatic. Symptomatic form includes classic and atypical presentation. The classic form of the disease, which primarily occurs in children aged 9–24 months, is characterized by chronic diarrhea, vomiting, abdominal distention, and malnutrition [4].

Some recent randomized controlled trials concluded that there is no influence of timing of gluten introduction on the risk of developing CD [5, 6]. New observational studies showed that breastfeeding, never during gluten introduction, influenced the risk of developing CD [5, 6]. Obviously, there is a need to further clarify the role of environmental factors in pathogenesis of CD.

The aim of this study was to investigate whether breastfeeding, particularly breastfeeding during gluten introduction, and timing of gluten introduction, influenced the onset of CD in infants.

METHODS

We retrospectively analyzed medical records of 93 infants (children up to 2 years of age; 61 girls and 32 boys), diagnosed with classic form of CD at the University Children's Hospital in Belgrade, between the year 2000 and 2010. The study protocol was approved by the hospital Ethics Committee. Of the 93 infants, 40 were diagnosed in the first year and 53 in the second, and all of them had severe gastrointestinal symptoms, characterized by a chronic diarrhea, poor appetite, and failure to thrive. Diagnose of CD was based on the criteria of the European Society for Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) [7]. In all participants, the enteropathy was destructive, in 90% total or subtotal and in 10% partial.

In medical records, we analyzed duration of breastfeeding before the diagnosis, timing of gluten introduction, the duration of symptoms, and the age at diagnosis. Having the retrospective study, we did not insist on exclusively breastfeeding, we assumed that the infant had been breastfed if mother had produced enough milk for a minimum of three complete nursing per day.

Participant descriptive statistics are shown in Table 1.

The participants were divided into two groups based on breastfeeding status at the time of gluten introduction. Twenty-four participants were breastfed at the time of gluten introduction, and 69 were not. In addition, according to new ESPGHAN position paper [8], we divided participants regarding timing of gluten introduction: before the fourth month ($n = 12$) and after ($n = 81$).

Binary logistic regression was conducted to estimate whether duration of breastfeeding and timing of gluten introduction influenced the risk of disease occurrence in the first year of life. Differences regarding age at diagnosis between the groups of infants formed based on timing of gluten introduction and breastfeeding during gluten introduction were determined using χ^2 test. For all statistical analyses OpenStat (Iowa, USA) software for Windows, version 11.9.08 (<http://openstat.en.softonic.com/>) was used.

RESULTS

Binary logistic regression showed that duration of breastfeeding and timing of gluten introduction influenced the risk of CD occurrence in the first year of life. The model with statistical significance influenced the risk ($\chi^2 = 16.14$; $df = 4$; $p = 0.001$), and explained 24.3–32.4% variance. The only variable with significant prediction was duration of breastfeeding, which reduced the onset of the disease in the first year $p = 0.039$ (OR = 1.43 95%, CI = 1.019–1.899) (Table 2).

Since the regression model suggested that timing of gluten introduction was not associated with the postponing the diagnosis, we compared the two groups of infants based on the timing of gluten introduction. Although in the first group (gluten introduced prior to the fourth month) the onset of the disease was earlier (12.75 ± 4.15 months) compared with the second group (14.69 ± 4.90 months), there was no significant difference ($F = 1.036$; $p = 0.197$) between them.

Longer breastfeeding was associated with delayed diagnosis of CD. Infants with the diagnosis made in second year were breastfed for 5.27 ± 4.68 months, while those diagnosed in the first year had shorter duration of breastfeeding (1.63 ± 3.99 months) and the difference between groups was significant ($F = 5.657$, $t = -4.15$; $p < 0.01$) (Figure 1).

In the group of infants that were not breastfed at the time of gluten introduction, the mean age at diagnosis was 13.20 ± 5.01 , and in the other group it was 16.31 ± 3.99 months, and the age at diagnosis was significantly delayed into second group ($F = 1.671$, $t = 2.39$, $DF = 91$, $p = 0.029$) (Figure 2).

Also, we found that there was twice as much girls than boys diagnosed with classic form of CD, but gender did not significantly affect age at diagnosis ($t = 0.87$, $DF = 91$, $p = 0.39$) nor the duration of symptoms ($t = -1.33$, $DF = 91$, $p = 0.18$).

DISCUSSION

CD is one of the most common diseases worldwide with the prevalence of 1% among Caucasians [8, 9]. Some epidemiological studies showed that the prevalence has increased over past decade, with no significant change in human genome [10]. It points out to the environmental factors and renews their role in the onset of the disease.

In our study, we found that breastfeeding delayed onset of CD in infants. Particularly, breastfeeding during gluten introduction postponed the onset of the disease. According to our study, timing of gluten introduction did not influence the occurrence of CD.

A protective effect of breastfeeding on CD has long been assumed and it occurs through various mechanisms, including presence of numerous nonnutritive factors, like lysozyme lactoferrin, s IgA and others [11, 12]. In addition, breastfeeding is excellent protection from gastrointestinal infections and repeated gastrointestinal infections have been reported to increase the risk of CD [13]. That is why breastfeeding may confer indirect protection from CD [12, 13]. Some studies point to the importance of continuing breastfeeding at the time of gluten introduction [11, 14]. Previous retrospective studies suggested a 'window of opportunity' for primary prevention by introducing gluten between four and six months of age during which breastfeeding provided a protective effect [14, 15, 16]. Small amount of gluten in breast milk helps induce oral tolerance, as is the case with other food allergens [17, 18]. In one recent study, no protective effect of breastfeeding on the development of CD was observed, while in another there was no significant difference in the percentage of children that developed CD among children that were introduced gluten during breastfeeding and in those that were not breastfed at the time of gluten introduction [19, 20, 21].

In our study, the duration of breastfeeding was generally short, and we can't be absolutely positive about its exclusiveness, but it obviously delayed the onset of the disease. Today we know that the type of milk, as well as the mode of the delivery, antibiotics and stress of any kind, strongly influence gastrointestinal microbiota [22, 23]. We did not investigate the gut microbiota in our participants, but we speculate that breastfeeding can promote and sustain healthy microbiome [24, 25]. This healthy pattern, along with other protective factors in human milk, can promote gluten tolerance, and delay occurrence of the

disease, even in patients with strong genetic predisposition [22, 23]. Although we are aware that there are studies that claim breastfeeding at the time of gluten introduction is not protective, in our study we showed that breastfeeding during gluten introduction postponed the disease, which is important, because in our group of patients duration of symptoms for only a few months at the early age was critical for growth, especially for weight gain.

On contrary to old ESPGHAN recommendation that gluten should not be introduced before 17 weeks and not later than at 26 weeks, preferably concurrent with the period of breastfeeding, new ESPGHAN position claims that gluten can be introduced to the infant's diet between the ages of 4 and 12 months [15, 16, 25]. The age of gluten introduction in infants of this age does not seem to influence the absolute risk of developing CD during childhood. Those recommendations were based on some new prospective, randomized trials [19, 21]. We also did not find the difference in the onset of CD regarding timing of gluten introduction. In our study, only small number of participants consumed gluten prior to the fourth months, and possible in small quantities. Another possible factor, which we did not take into account, was the amount of gluten consumed by the infants.

Additionally, in our study, we found that girls were twice as often affected than boys were which is in accordance to the fact that CD is autoimmune disease, and shares some important features with other autoimmune diseases that are being more prevalent in females than in males [26]. In addition, in our study, gender did not significantly affect the age at diagnosis or the duration of symptoms.

It is well known that genetic predisposition, which all our participants clearly possess, and exposure to gluten are the two most important factors necessary for CD to develop [19, 20, 21]. Early nutrition practices have been studied long and hard, but new studies pointed to other environmental factors to contribute the CD risk [22, 27].

CONCLUSION

In this study, we found that the duration of breastfeeding, and breastfeeding at the time of gluten introduction, postponed the onset of CD in genetically predisposed children up to two years old. We did not confirm the timing of gluten introduction influenced the age of occurrence of CD in these group of children. Our research is a contribution to a very complex nature of CD that requires more investigations not only in the field of genetic predisposition, but also regarding influence of various nutritive and nonnutritive environmental factors on its expression.

Conflict of interest: None declared.

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Table 1. Descriptive statistics of 93 infants with classic CD

| | |
|-------------------------------------|--------------|
| Age (months) at celiac diagnosis | 14.28 ± 4.78 |
| Duration (months) of symptoms | 2.08 ± 1.84 |
| Duration (months) of breastfeeding | 3.33 ± 3.77 |
| Age (months) at gluten introduction | 4.64 ± 1.23 |

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Table 2. Binary logistic regression for the analysis of association between duration of breastfeeding and occurrence of Celiac disease after the first year in 93 infants diagnosed with classic form of Celiac disease

| Variable | B | SE | p | Exp (B) or OR | 95% CI for OR |
|---------------------------|--------|-------|-------|---------------|---------------|
| Duration of breastfeeding | 0.383 | 0.141 | 0.010 | 1.439 | 1.019–1.899 |
| Constant | -2.369 | 1.467 | 0.106 | 0.094 | |

OR – odds ratio; CI – confidence interval; SE – standard error

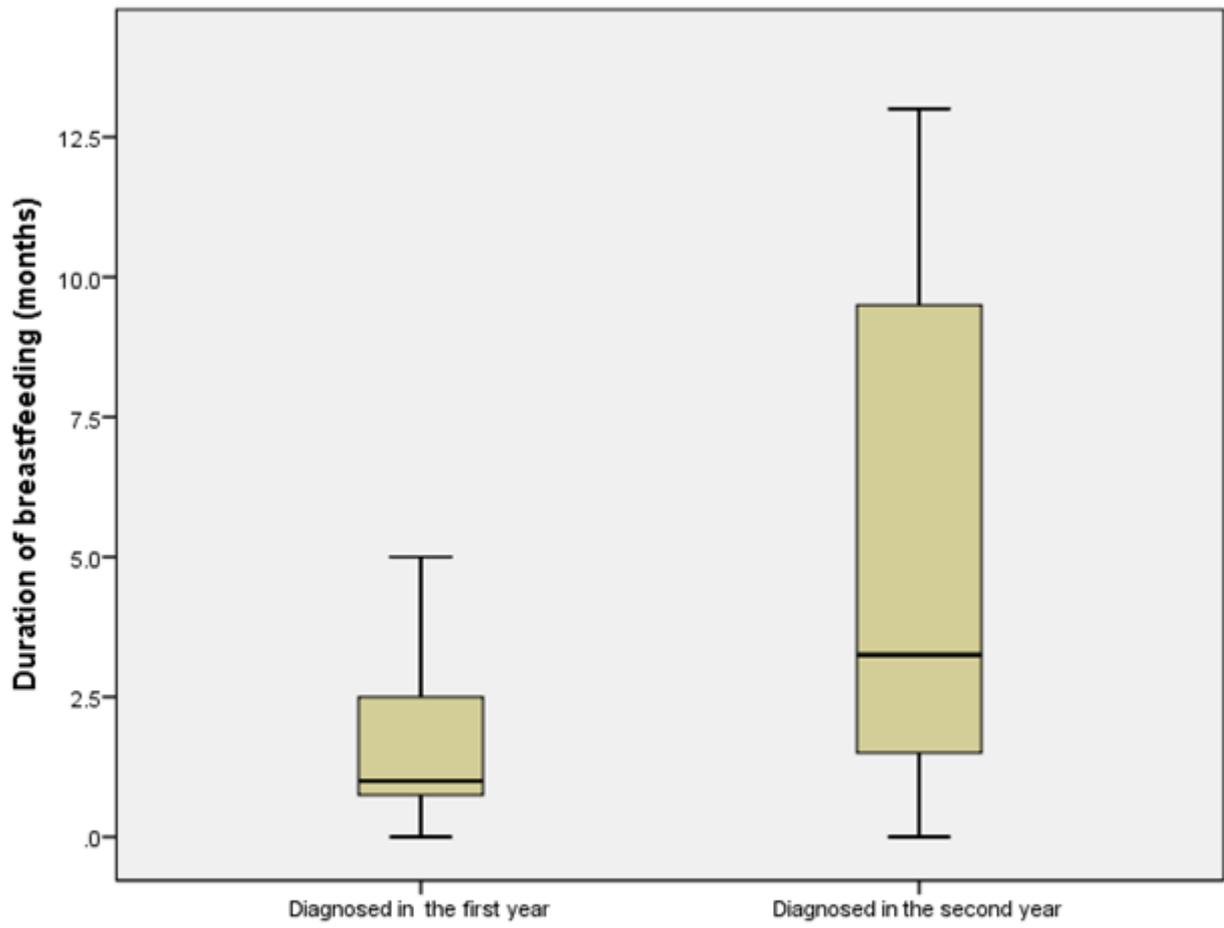


Figure 1. Duration of breastfeeding in infants with classic CD diagnosed in first and in the second year

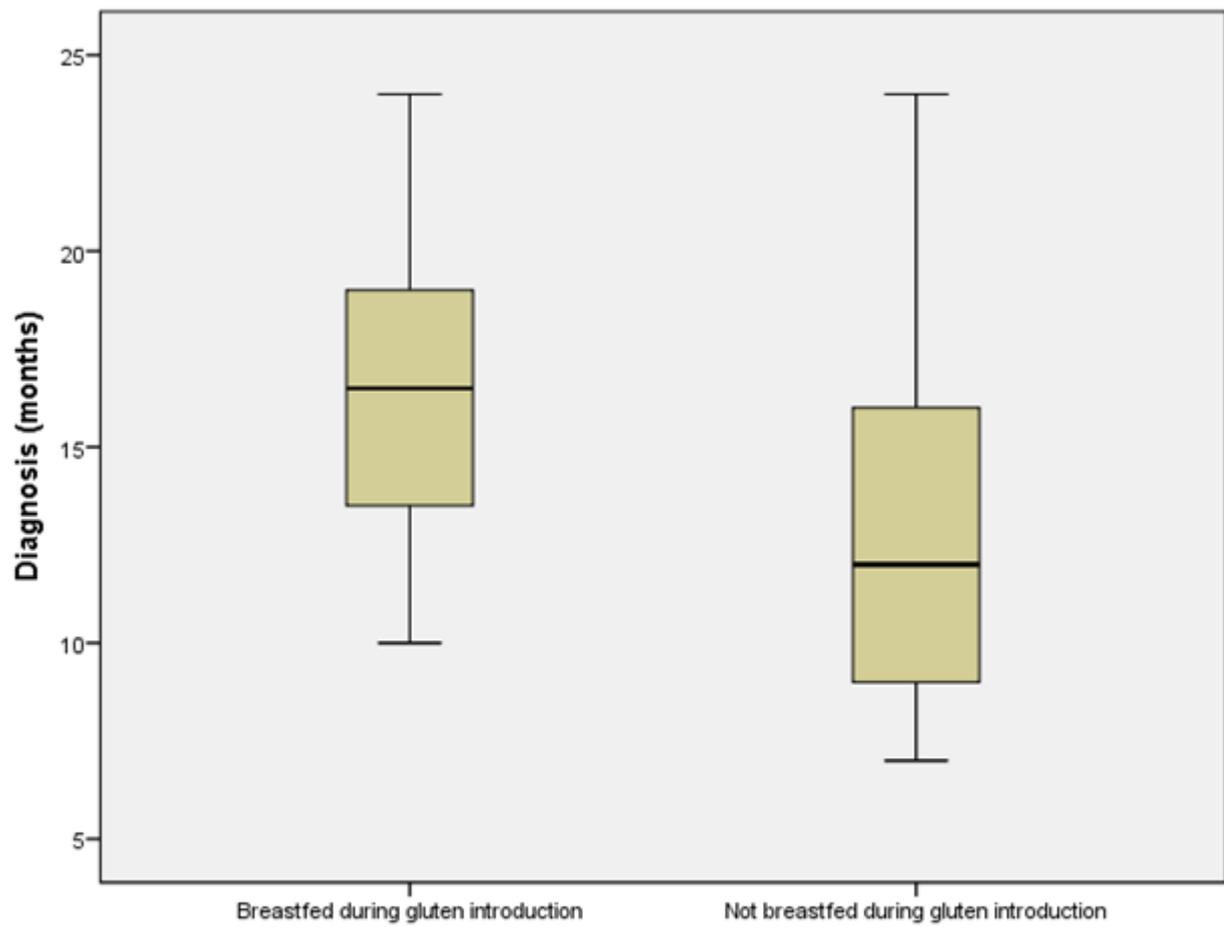


Figure 2. Occurrence of the CD in two groups of infants based on breastfeeding status during gluten introduction