Frontiers in Health Informatics
ISSN-Online: 2676-7104
www.healthinformaticsjournal.com
2024; Vol 13: Issue 3
Open Access

# Effectiveness Of Probiotic Mouthwash And Anti-Ca-gtf-IgY Lozenges In High Caries Risk Children Using Cariogram - A Randomized Clinical Trial

***Running title: Effectiveness Of Probiotic Mouthwash And Anti-Ca-gtf-igy Lozenges In High Caries Risk Children***

**Shankar P, Jeeva Rathan, Kabita Sahoo, , Trophimus J, Nancy S, Kalapana H**

[1]https://orcid.org/0000-0003-4524-6875 Reader, Department of Pediatric and Preventive Dentistry, SRM Dental College, Ramapuram, Chennai- 600087 Email: pedoshankar@gmail.com
[2]Professor, Department of Pediatric and Preventive Dentistry Sree Balaji dental college and hospital, Chennai
[3]Dental Surgeon, Health and Family Welfare Department, Govt. of Odisha, India
[4]https://orcid.org/0000-0002-8285-7424 Reader, Department of Pediatric and Preventive Dentistry, SRM Dental College, Ramapuram, Chennai
[5]https://orcid.org/0000-0001-6337-9300 Senior Lecturer, Department of Pediatric and Preventive Dentistry, SRM Dental College, Ramapuram, Chennai
[6]Professor, Department of Anatomy, Sree Balaji Medical College and Hospital, Chennai

Conflict of interests: None declared.

Ethics approval: Study was approved by the Institutional Ethical Review board of M&HS/SRMDC/2020/PG/024. Also registered in clinical trial registry of india REF/2020/02/031636

Funding: Self-funded.

Author contributions: All authors participated in the study design, implementation and writing of the manuscript.

Acknowledgements: None

Cite this paper as: Shankar P, Jeeva Rathan, Kabita Sahoo, , Trophimus J, Nancy S, Kalapana H (2024) Effectiveness Of Probiotic Mouthwash And Anti-Ca-gtf-IgY Lozenges In High Caries Risk Children Using Cariogram - A Randomized Clinical Trial . *Frontiers in Health Informatics, 13* (3), 10416-10422

***Abstract***

***Introduction –*** *Dental Caries is the most prevalent disease worldwide. Its multifactorial origin necessitates the risk factors to be monitored. Cariogram is one such risk assessment tool that graphically represents the several risk factors that support the development of dental caries.*

***Aim-*** *To estimate the efficacy of probiotic mouthwash and anti-CA-gtf IgY lozenges in high caries risk children using cariogram.*

***Methods-*** *Sixty children (6 to 9 years) with high caries risk were randomly distributed into 3 groups. Group A control and group B Probiotic mouthwash (Darolac) and group C with fluoridated toothpaste and anti-CA-gtf IgY lozenges (No Decay). After an interval of one month, cariogram was again constructed. Data was collected and put to statistical analysis.*

***Results-*** *The intergroup comparison showed that Group C had the greatest reduction in the red sector than Group A and B (**p=0.03** and p=1.0) respectively. The intergroup comparison showed the greatest increase in green sector in Group B followed by Group C and A respectively, however which showed no statistically*

*significance.*

***Conclusion-*** *Probiotic mouth wash and Anti-CA gtf Ig Y lozenges was effective in reducing bacterial load thereby increasing the chance to avoid new caries.*

***Key words-*** *Cariogram, dental caries, glucosly-transferace, lozenges, probiotic mouthwash*

## Introduction

Dental caries is the most prevailing oral disease worldwide.[1-3] Its occurrence is mainly due to the contribution of different bacterial colonization, thus forming a biofilm over the tooth surface.[2] The initiation and development of caries may be imparted to multiple threat factors like oral microflora, fluoride experience, salivary components and diet.[4,5] Caries risk assessment approaches helps in identifying these risk factors and also planning preventive programs that reduces the development of caries.[6] "Cariogram", is one such computer based graphical representation tool that helps identify the risk to develop new caries by the interaction of several factors that contribute to dental caries, It also facilitates in evaluating the efficacy of any interventions provided in preventing caries in at-risk patients.[7]

The gold standard for controlling plaque and caries has been described in literature as mechanical brushing and flossing techniques.[8] However, with the advancing years, focus has been shifted to the use of chemical methods as a supplement to mechanical methods thus, augmenting in plaque and caries control by the modulations in the oral micro flora.

Procter and Gamble in 1955, had first introduced a clinically tested fluoridated dentifrice since then, it has been widely used throughout the world and proven as an effective anticaries and anti-plaque agent.[9]

Among the various existing chemical aids like mouthwashes, prophylactic pastes and lozenges, probiotics is considered as one of the breakthrough aid which is effective in reduction of pathogenic habitants and plaque in the oral cavity.[10]

It is believed that Streptococcus mutans is the primary culprit behind dental caries development and are known to synthesis the most virulent factor, the cell associated glucosyltransferase (CA-gtf).[2] Therefore, they serve as an essential molecular target for preventing caries. Recently lozenges consisting of egg yolk based antibody against CA-gtf had been successfully used to reduce Streptococcus mutans colonization but studies comparing the effectiveness of probiotics and Anti-CA-gtf IgY lozenges in caries reduction among children are scarce, Therefore, the current study's objective was to assess the effectiveness of the probiotic mouthwash and the Anti-CA-gtf lozenges in preventing caries among the kids who have a higher risk of cavities using cariogram.

## Materials and techniques

A three arm paralleled single masked randomized controlled clinical trial was the design of the current investigation. The research and control groups were chosen and vetted from a Chennai, Tamil Nadu, India, home for single children. The Institutional Review Board (M&HS/SRMDC/2020/PG/024) granted an ethical clearance certificate, and the trial was also listed in the Indian Clinical Trial Registry (CTRI/ 2019/09/021033). The relevant children's home authorities gave their informed consent. The study's entire process was depicted in the CONSORT flow chart (Figure -1).

G* Power Software 3.1.9.2 version was used to calculate the sample size based on the values of the key article (Di Pierro Fet al. Results of a randomized, controlled trial on the cariogram outcome following a 90-day oral treatment with Streptococcus salivarius M18 in children at high risk for dental caries). Sixty kids, aged six to nine, who had a high risk of dental caries, were part in the study. The children who met the exclusion criteria included those who had systemic diseases, had a history of sensitivity to drugs, mouthwash, or eggs, had received corticosteroid therapy, antibiotics, or mouthwash in the previous 30 days, had undergone professional treatment for the removal of plaque and calculus in the previous 15 days, or had an orthodontic appliance or

removable prosthesis.

Using a computer-generated database of random numbers, the chosen cohort was randomly assigned in a 1:1:1 ratio to each of the three groups. The study's interventions were only blinded to the investigator. However, because the interventions were in recognized forms, it was not viable to blind the individuals.

**Study protocol:**

A single trained investigator KS had screened and assessed all the children. One hundred children from the children's home were screened for the study. The baseline cariogram was constructed for each child, Children with values at day zero <20 in the green sector (chance to keep away new caries or the high caries risk), were incorporated into the study. 60 children were recruited and allocated into 3 groups. Another dentist SP was in charge of distribution as well as weekly monitoring of the interventions provided to all the children incorporated into the investigation, under the supervision of the children's home authority.

Group A: were told to use a soft-bristle toothbrush and fluoridated toothpaste twice a day.

Group B: Were trained to brush twice daily with fluoridated toothpaste and use a commercially available probiotic sachet (Darolac) dispensed in a 10ml of distilled water as a mouthwash, swish and swallow once daily at night.

Group C: were told to use fluoridated toothpaste twice a day and to consume (No Decay) anti-CA-gtf IgY Lozenges twice daily after food as instructed by the manufacturer.

After an interval of one month, the cariogram was again constructed for all the three groups by KS to assess the difference in green sector or chance to avoid new caries.

**Construction of cariogram:**

The cariogram software (English Version 3.0, Department of Cariology, Malmo University, Sweden) was exercised to assess the the shift in the risk profile for caries. Out of ten, eight variables- experience with caries, associated diseases, diet composition, frequency of diet, quantity of plaque, streptococcus mutans, fluoride program and clinical judgement were considered for the cariogram construction in the study. Intra-oral clinical examination and Silness-Loe Plaque Index were performed on each subject to score the caries experience and the accumulation of plaque respectively. History was taken to record the related disease, diet frequency and fluoride program. Diet content was scored using the lactobacillus count in saliva. The microbial count, both the lactobacillus and streptococcus mutans were estimated from the unstimulated whole saliva accumulated by the draining method. Prior to the saliva collection, subjects were limited from tooth-brushing and any other oral hygiene measures for the last 24 hours. All the collection was made between a standard time interval (8-11am) when the subjects were in a fasting state. The subjects were asked to sit upright and tilt their head down and mouth open, to collect the saliva in the floor of the mouth for a minute and to drool it passively down the lower lip into a sterile plastic vials. The collected sample was immediately inoculated on selective agar plates ( Mitis Salivarius Agar Base and Rogosa LBS Agar ) for streptococcus mutans and lactobacillus respectively and cultivated at $37^{O}$ C for 48 hours.

**Data gathering and statistical analysis:**

A Microsoft Excel worksheet was used to record and enter the cariogram's results at days 0 and 30. SPSS (IBM SPSS Statistics for Windows, Version 26.0, Armonk, NY: IBM Corp., Released 2019) was used for the data analysis.The mean values reported for each variable in the study were compared using a one-way ANOVA. To determine the pairwise comparison between the groups, a post hoc test was conducted. The significance level was set at 5% ($\alpha = 0.05$) and a P-value of less than 0.05 was deemed statistically significant.

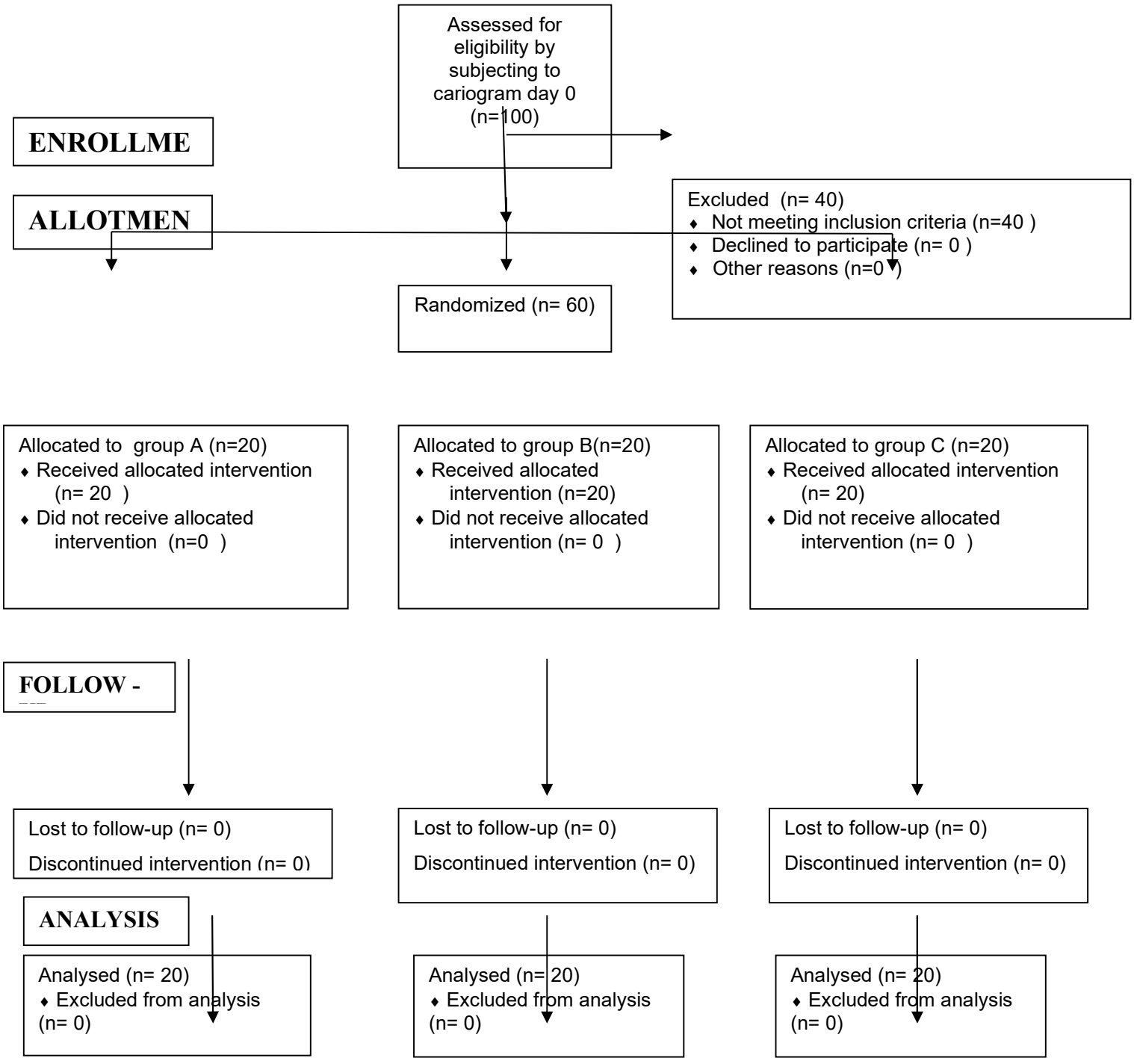
Assessed for eligibility by subjecting to cariogram day 0 (n=100)
ENROLLME
ALLOTMEN
Excluded (n= 40)
Not meeting inclusion criteria (n=40 )
Declined to participate (n= 0 )
Other reasons (n=0 )
Randomized (n= 60)
Allocated to group A (n=20)
Received allocated intervention (n= 20 )
Did not receive allocated intervention (n=0 )
Allocated to group B(n=20)
Received allocated intervention (n=20)
Did not receive allocated intervention (n= 0 )
Allocated to group C (n=20)
Received allocated intervention (n= 20)
Did not receive allocated intervention (n= 0 )
FOLLOW -
Lost to follow-up (n= 0)
Discontinued intervention (n= 0)
Lost to follow-up (n= 0)
Discontinued intervention (n= 0)
Lost to follow-up (n= 0)
Discontinued intervention (n= 0)
ANALYSIS
Analysed (n= 20)
Excluded from analysis (n= 0)
Analysed (n= 20)
Excluded from analysis (n= 0)
Analysed (n= 20)
Excluded from analysis (n= 0)

**Figure 1: CONSORT FLOWCHART**

**RESULT:**

The study comprised sixty kids from a children's home who had a significant risk of dental cavities. All of the kids were taken into consideration for statistical analysis because there were no dropouts. Groups A, B, and C had mean ages of 7.0±0.81, 6.7±0.8, and 7.1±0.87, in that order. Regarding age and gender, there was no statistically significant difference between any of the three groups ($p>0.05$). The findings of the Shapiro-Wilks, Kolmogorov-Smirnov, and normality tests showed that the study had a normal distribution. Therefore, parametric tests were used to analyze the data.

In the present study, changes in the dark blue (lactobacillus count/diet content and diet frequency), red (streptococcus mutans and plaque amount) and green (chance to avoid new caries) sector could be appreciated in all the three groups, (Table 1).

The inter-group comparisons were done using post hoc analysis. Group C showed the greatest reduction in red sector from day 0 to day 30 than Group A and B. (**p=0.03** and p=1.0). There was a greater reduction in the red sector in group B than Group A however was not statistically significant (p=0.2). The mean difference at day 0 and day 30 was greatest in Group C followed by Group B and Group A respectively (Table 2)

The intergroup comparison showed a statistical difference in the reduction of dark blue sector among all the three groups. Group B had shown greater reduction than Group A (p=0.058) and Group C (p= 0.00) which was statistically significant (Table 2) but no statistically substantial change was detected among the groups in green sector (Table 2).

**Table 1: changes in Mean values among the sectors during the study period**

| | **Group A** | | **Group B** | | **Group C** | |
|---|---|---|---|---|---|---|
| | **Day 0** | **Day 30** | **Day 0** | **Day 30** | **Day 0** | **Day 30** |
| **Dark blue sector** | 20.3 ± 2.8 | 16.0 ± 0.0 | 20.9 ± 2.7 | 8.0 ± 0.0 | 20.9 ± 2.7 | 13.0 ± 0.0 |
| **Red sector** | 31.8 ± 1.9 | 21.0 ± 0.0 | 31.4 ± 2.0 | 8.0 ± 0.0 | 31.4 ± 2.0 | 2.0 ± 0.0 |
| **Green sector** | 14.6 ± 2.2 | 32.0 ± 0.0 | 14.3 ± 2.0 | 62.0 ± 0.0 | 14.3 ± 2.0 | 58.0 ± 0.0 |

**Table 2: Inter group comparison between the groups using post hoc analysis**

| | Red sector | Green sector | Dark blue sector |
|---|---|---|---|
| VARIABLES | Sig. | Sig. | Sig. |
| | | | |

| | | | | |
|---|---|---|---|---|
| Group A | Group B | .224 | .065 | **.058** |
| | Group C | **.033** | .137 | **.000** |
| Group B | Group A | .224 | .065 | **.058** |
| | Group C | 1.000 | 1.000 | **.000** |
| Group C | Group A | **.033** | .137 | **.000** |
| | Group B | 1.000 | 1.000 | **.000** |

P value ≤ 0.05 is considered significant

**Discussion:**

An important first step in learning more about a patient's caries profile when they visit the dentist is to get a caries risk assessment and chairside care has found Cariogram software to be a useful tool to assess caries risk assessment.[11] Therefore in the current investigation cariogram was employed to assess the alteration in caries risk profile subsequent to the use of probiotic mouthwash and Anti-CA GTF lozenges.

Live microorganisms, such bacteria or yeast, that colonize a host and provide benefits are known as probiotics. Probiotics that are most well-known include Bifidobacterium and types of lactic acid bacteria. These bacteria have characteristics like enzyme action and acidity that enable them to withstand difficult circumstances inside the host organism. By performing biological tasks and managing the microbiome, they may invade the host and improve health. Additionally, these probiotics release bacteriocins, which have a bacteriocidal impact on Streptococcus mutans..[12]

Dental plaque is formed when S. mutant adheres to the tooth surface and synthesizes water-insoluble glucan from dietetic sucrose via the cell associated glucosyl-transferase (CA-gtf) enzyme, a crucial component of virulence that is present on the surface of bacteria. and Lozenges bearing selective egg yolk antibodies (IgY) against CA-gtf of S. mutans had a brutal impact on S. mutans colonization of the oral cavity.[13]

In the present study probiotic mouthwash and Anti-CA GTF lozenges displayed a significant drop in streptococcus mutans count in high risk children similar to the study done by Di Pierro F et al 2015 and Nguyen SV et al 2011.

**CONCLUSION**

Both probiotic mouthwash and Anti-CA GTF lozenges significantly reduced bacterial count in high caries risk children thereby increases the chance to avoid new caries.

**Refernces**

1. Pitts NB, Zero DT, Marsh PD, Ekstrand K, Weintraub JA, Ramos-Gomez F, Tagami J, Twetman S, Tsakos G, Ismail A. Dental caries. Nature reviews Disease primers. 2017 May 25;3(1):1-6.
2. Jayakaran TG, Rekha CV, Annamalai S, Baghkomeh PN. Salivary peptide human neutrophil defensin1–3 and its relationship with early childhood caries. Dental Research Journal. 2020 Nov 1;17(6):459.
3. Amudha S, Moses J, Vijayakumar M, Shankar P. Prevalence of Dental Caries among Different Socioeconomic Status and their Treatment Needs among 5–15-year-old School-going Children in Maduravoyal Area, Chennai. Int J Clin Pediatr Dent 2021;14(3):403–411. DOI: 10.5005/jp-journals-10005-1963
4. Fejerskov O, Larsen MJ, Richards A, Baelum V. Dental tissue effects of fluoride. Advances in dental research. 1994 Jun;8(1):15-31.
5. Bagramian RA, Garcia-Godoy F, Volpe AR. The global increase in dental caries. A pending public health crisis. Am J dent. 2009 Feb 1;22(1):3-8.

6. Celik EU, Gokay N, Ates M. Efficiency of caries risk assessment in young adults using Cariogram. European journal of dentistry. 2012 Jul;6(03):270-9.
7. Di Pierro F, Zanvit A, Nobili P, Risso P, Fornaini C. Cariogram outcome after 90 days of oral treatment with Streptococcus salivarius M18 in children at high risk for dental caries: results of a randomized, controlled study. Clinical, cosmetic and investigational dentistry. 2015 Oct 3:107-13.
8. Sgolastra F, Fidanza F, Carosi D, Petrucci A, Calò G, Gatto R. An interdisciplinary approach to a survey on dental caries in a group of 3-year-olds in Ascoli Piceno (Italy). European Journal of Paediatric Dentistry. 2010 Sep 1;11(3):137-40.
9. Baig A, He T. A novel dentifrice technology for advanced oral health protection: A review of technical and clinical data. Compendium of Continuing Education in Dentistry (Jamesburg, NJ: 1995). 2005 Sep 1;26(9 Suppl 1):4-11.
10. Purunaik S, Thippeswamy HM, Chavan SS. To evaluate the effect of probiotic mouthrinse on plaque and gingivitis among 15-16 year old school children of Mysore City, India-randomized controlled trial. Glob J Med Res. 2014 Aug;14:9-14.
11. Poorni S, Nivedhitha MS, Srinivasan M, Balasubramaniam A. Effect of probiotic Streptococcus salivarius K12 and M18 lozenges on the cariogram parameters of patients with high caries risk: a randomised control trial. Cureus. 2022 Mar;14(3
12. Silva DR, Sardi JD, de Souza Pitangui N, Roque SM, da Silva AC, Rosalen PL. Probiotics as an alternative antimicrobial therapy: Current reality and future directions. Journal of Functional Foods. 2020 Oct 1;73:104080
13. Nguyen SV, Icatlo Jr FC, Nakano T, Isogai E, Hirose K, Mizugai H, Kobayashi-Sakamoto M, Isogai H, Chiba I. Anti–cell-associated glucosyltransferase immunoglobulin Y suppression of salivary mutans streptococci in healthy young adults. The Journal of the American Dental Association. 2011 Aug 1;142(8):943-9.