

Association of Salivary ABO Antigen Expression and Dental Caries Experience

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Abstract

This study aimed to determine ABO blood group phenotypes and secretor status from saliva samples and to evaluate their association with dental caries experience among Iraqi primary schoolchildren. A cross-sectional study was conducted among 88 children aged (6-12) years attending governmental dental centers in Baghdad, Iraq. Unstimulated whole saliva samples were collected and analyzed using the absorption-inhibition (hemagglutination method) to determine ABO blood group phenotype and secretor status. Dental caries experience was assessed using DMFT/dmft index according to World Health Organization criteria. Statistical analyses were performed to compare caries scores according to secretor status and ABO blood group. Of the 88 participants, 81 (92%) were identified as secretors and 7 (8%) as non-secretors. Among secretors, Blood group B was the most prevalent (45%), followed by O (31%), A (17%), and AB (7%). No statistically significant differences were observed in DMFT scores between secretors and non-secretors in permanent dentition ($p=0.177$), nor in dmft scores in primary dentition ($p=0.461$). Similarly, no significant association was found between ABO blood group phenotype and caries experience in either permanent or primary teeth (DMFT: $p=0.542$; dmft: $p=0.359$). Most Iraqi children in this study were secretors, with blood group B being the most common phenotype. Neither secretor status nor ABO blood group phenotype showed no association with dental caries experience.

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Introduction

A highly prevalent oral disease worldwide, commonly known as tooth decay or Dental caries, affecting all age groups of people and socioeconomic backgrounds [1]. At early life of human this disease can start and if not treated it may progress and end up with tooth loss [2]. Dental caries happens due to a mix of different factors. It involves the host factors like the vulnerability of teeth and the quality of saliva, along with dietary sugars and cavity-causing bacteria, all coming together over a period [3]. The heterogeneous etiology of these diseases

explains the difficulty to achieve its control and has a direct impact not only on the oral health of the individual but also about their general health [4].

Austrian Karl Landsteiner discovered the ABH blood group system in 1901 [5]. The A and B antigens in the ABO blood group system are derived from a precursor known as H substance. This transformation begins around 5 to 6 weeks into fetal development. It's important to note that the conversion of H substance into either A or B antigens is only partial. For individuals with blood group O, there is no conversion of H substance at all [6].

Blood group is a genetically determined factor, and it is a unique character of every individual [7].

These group-specific substances, known as ABH, are not just limited to red blood cells; you can find them in various body fluids as soluble forms, including sweat, digestive secretions, and even breast milk, and tears except cerebrospinal fluid. One of the richest and most available sources is saliva [5]. That group of people who secrete ABH soluble substances in secretion are known as secretor [6].

The FUT 2 (Se) gene is responsible for the body's ability to release A, B, and H substances in a water-soluble form into the body fluids [7]. While there are not any diseases directly caused by not expressing ABO blood group antigens, some studies suggest that a person's ABO phenotype might influence their susceptibility to various diseases. These connections are still up for debate, but there have been observations linking certain bacteria to specific blood groups—like *Pseudomonas* with group A, *Helicobacter pylori* with group O, and *Salmonella typhi* with group B. Additionally, individuals with the Histo-blood group A have been found to have a higher incidence of several types of cancer, including rectal, cervical, gastric, breast, and ovarian cancers [8]. Every person can either be a secretor (Se) or a non-secretor (se), and this trait is separate from their blood type—whether it's A, B, AB, or O. This means you could be an A secretor or an A non-secretor, a B secretor or a B non-secretor [9]. When it comes to bodily fluids, those with blood group “O” only secrete the H element, while blood group “A” secretes both A and H substances, and blood group “B” secretes both B and H substances [10].

Generally, in a population, there are around 80% secretors and 20% non-secretors [7], but with some racial and ethnic differences [11]. There is evidence to suggest a link between secretor status and the appearance of caries. It has been proposed that secretion of these antigens into the saliva might be caries-preventive [12]. The results of a previous study showed that the dental caries status was more in the secretors than non-secretors [5].

Llena-Puy (2006) has been suggested that the way blood group antigens are expressed in saliva could alter how microorganisms interact with their salivary glycoprotein receptors. This change might play a role in both the development and prevention of oral infectious diseases [13]. As a result, some researchers have proposed that the presence of AB antigen in saliva could act as a protective agent against cavities [14], and patients who are secretors had less dental caries than non-secretors [15]. However, other researchers showed that the secretor status unrelated to the caries scores [16].

An Icelandic study indicated that blood group antigens might play a role in hindering the ability of *Streptococcus mutans* to stick to teeth, which could help prevent cavities [11,17].

Saliva has emerged as an important biological fluid for diagnostics,

Its noninvasive collection, economical, and easy to collect, especially in pediatric populations, make it a compelling alternative to traditional blood-based diagnostics [18].

Detection of ABO blood group antigens from saliva can be performed using the absorption–inhibition hemagglutination method [19].

The aims of this study were to determine ABO secretor status and ABO blood group phenotype through salivary analysis, evaluate dental caries experience using the dmft/DMFT index, and assess the association between ABO secretor status, and ABO blood group type with the dmft/DMFT index.

Materials and Methodos

A cross-sectional study carried out from, 29 December 2025, to 25 February 2026, at 3 Baghdad government dental centers.

Institutional Review Board (IRB) College of Medicine-Al-Nahrain University approved the study protocol with number 198 on November 16, 2025.

Salivary samples from Ninety-three (6-12) years old children who attended government dental centers.

Whole unstimulated saliva was collected over a time span of 5 min between 9 to 11am. The child was instructed to stop consuming food, chewing gum, and drinking beverages one hour before to saliva collection, then we asked them to sit on a chair and lean their head forward over the test tube, The participant was instructed to keep his mouth open to facilitate the drainage of saliva into the tube.

The saliva samples were kept at 4°C and were transferred to the laboratory up to 20 min and stored at -20°C for Absorption–Inhibition Method (hemagglutination) [20].

We evaluated dental caries experience by using the Decayed, Missing, and Filled Teeth (DMFT/dmft) index, following the criteria set by the World Health Organization. Examination conducted under adequate illumination (artificial light) with mouth mirror and probe. Each tooth is counted once only. Usually 28 teeth included (excluding third molars) in DMFT and 20 teeth in dmft index.

We excluded teeth can be missing for a variety of reasons, including trauma, orthodontic extractions, or periodontal disease, and restorations were done either for cosmetic purposes or to apply preventive resin treatments.

For this test, a one drop of heat inactivated saliva samples (heated in boiling water for 10 minutes) was added to 1 drop of appropriately diluted anti-A, anti-B or anti-H (Lorine Laboratories Ltd, Berks, UK) in micro titration plate with 96 round bottom wells and left at room temperature for 10 minutes, the one drop of the appropriate indicator 5% RBC suspension was added to each well as the following: A cells with anti-A, B cells with anti-B and O cells with anti-H (Figure 1). The plate was incubated at room temperature for 30 minutes, and then cell agglutination at well bottom was

inspected. The results were independently evaluated by two qualified specialists (Hematologist and Biologist, Table 1). The principle of hemagglutination inhibition test based on neutralizing action of soluble ABH substances present in saliva to their corresponding antibodies.

We gathered, summarized, analyzed, and presented the data using the Statistical Package for the Social Sciences (SPSS) version 26 and Microsoft Office Excel 2024. For qualitative (categorical) variables, we reported the results as numbers and percentages. On the other hand, quantitative (numeric) variables were assessed for normal distribution using the Shapiro-Wilk test and were presented as means and standard deviations (SD), median and range. Mann Whitney U test was used for comparison of parameters between two groups while Kruskal Wallis H test was used when comparison among more than 2 groups. Spearman correlation as bivariate correlation between parameters (as data were not normally distributed) and expressed as (r) and its p value. For all comparisons and correlations, the level of significance was considered at P-value of less than 0.05.

Results

A total of 88 children participated in the study out of which 40 (45.5%) were males and 48 (54.5%) were females, the age ranged from 6-12 years with a median of 8 years.

In respect to the salivary secretor status in figure (1A), the results of hemagglutination test revealed indicate that (92%) of the participants are secretors while (8%) are non-secretors.

Total number of participants was 88 children; Eighty-one of them are secretor, their ABO blood groups were illustrated in Figure (1B), According to ABO blood groups; the majority of participants 45 % having B phenotype (n=36) , while 31% (n=25) have O phenotype, and 17 % (n=14) out of them have A phenotype. AB phenotypes constitute the minority of patients 7% (n=6).

Comparison of DMFT/dmft between Secretors and Nonsecretors is presented in Table 2. There was no statistically significant difference in DMFT status between secretors and nonsecretors in permanent dentition (p=0.177). also, statistically no significant difference was found in dmft between secretor and nonsecretor statuses in mixed dentition (p =0.461).

However, Table 3 demonstrates the Comparison of caries score experience (DMFT, dmft) according to ABO blood groups for the secretor child , For permanent teeth, median DMFT scores were 2 (0-4), 2 (0-7), 2 (0-10) and 3 (2-6) for blood groups A, B, O, and AB, respectively, with no statistically significant difference observed between the groups (p = 0.542).

Similarly, median dmft scores for primary teeth were 7 (0-17), 6 (0-14), 5 (0-13), and 3 (2-10) for blood groups A, B, O, and AB, respectively, and again no significant difference was detected ($p = 0.359$).

Discussion

The literature consistently reports that approximately 80% of the general population are secretors, while 20% are non-secretors [7]. In the present study, the hemagglutination inhibition test demonstrated that 92% of the participants were secretors ($n=81$) and only 8% were non-secretors ($n=7$), which exceed the globally established figure and may reflect population-specific genetic variation within the Iraqi pediatric population.

Regarding ABO blood group distribution among secretor children, blood group B was the most prevalent (45%, $n=36$), followed by blood group O (31%, $n=25$), blood group A (17%, $n=14$) and blood group AB (7%, $n=6$). This distribution is consistent with the recognized ABO phenotype frequencies in the population and is comparable to previous findings [21-25].

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Regarding secretor status and dental caries (Table 2), the present study found no statically significant difference in caries experience between secretor and non-secretor in either the primary or permanent dentition. Although secretors showed numerically higher median DMFT scores in permanent teeth 2 (0-10) compared to non-secretor 1 (0-4), these differences did not reach statistical significance ($p=0.177$ and $p=0.461$, respectively) as determined by Mann-Whitney U test. These findings are agreement with several previously published studies. Chung et al. (1965), in a study of 137 children aged 14-17 years, found no apparent relationship between caries experience of the permanent teeth and secretor status of ABO substances [26].

Also, the results of current study align with those of a cross-sectional study conducted in Makkah, Saudi Arabia by Al-Sihli, et al. (2018), which reported that no correlations were found between ABO blood grouping, salivary secretor status and dental caries [25]. However, the findings contrast with those of other studies that did report a significant association. Arneberg et al. (1976) found that secretors had significantly lower caries prevalence than non-secretors across all blood groups [15]. Holbrook and Blackwell reported that non-secretors had significantly higher DMFT scores compared to secretors [17], and more recently, Kárpáti et al. (2014) studied 130 Hungarian children and adolescents aged 6–18 years and found that children who were secretors had significantly fewer caries in their primary teeth compared to non-secretors [12]. It is also

worth noting that the relatively small number of non-secretors in the present sample ($N=7$) compared to secretors ($N=81$) may have limited the statistical power to detect any potential differences, and this should be considered when interpreting the results.

As shown in Table 3, no statically significant differences in caries experience were found among the four blood groups in either the primary or permanent dentition ($p = 0.542$ and $p = 0.359$, respectively). Although blood group AB recorded the highest median DMFT score in permanent teeth, this agrees with Govindaraju et al. (2018) [27], and blood group A recorded the highest median dmft score in primary teeth, these differences did not reach statistical significance. These results are in accordance with Mazumdar et al. (2014), who found no significant difference in caries scores among different blood groups, concluding that there is no correlation between blood group and dental caries [22], and noting that further studies on larger populations are needed to derive any conclusive results. In contrast to the present study, the results are inconsistent with those reported by Almalki et al., (2024), who observed a significantly higher DMFT prevalence among individuals with blood group AB [28], and by Yadav et al., (2018), who suggested a positive association between ABO blood groups and DMFT scores [24]. The conflicting results observed across studies are likely due to differences in study population, sample sizes and study designs. Also, the absence of a significant association may be explained by the multifactorial nature of dental caries, where biological factors such as secretor status and blood group phenotype may be overshadowed by behavioral and environmental determinants. Many factors work together to either cause or prevent caries, such as oral hygiene habits, the use of fluoride toothpaste, diet, and access to dental care, this explanation was also suggested by Holbrook (2015) [17], these factors may be strong enough to hide or reduce any possible effect that secretor status has on caries experience. In other word, even if blood group substances in saliva play some protective role, this effect might not be visible when other powerful factors are involved at the same time so that the interaction of multiple cariogenic and protective factors makes it difficult to detect a clear link between secretor status and caries scores.

Conclusion

In this study, 92% of the children (participants) were found to be secretors and only 8% were non-secretors.

Regarding the ABO blood group distribution, blood group B was the most common, followed by O, A, and AB.

No statistically significant difference in caries experience was found between secretors and non-secretors in either the primary or permanent dentition. Similarly, no statistically significant differences in caries experience were found among the four ABO blood groups in either dentition.

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Table 1. Hemagglutination results interpretation.

Anti-A + (A RBCs)	Anti-B (B RBCs)	Anti-H (O RBCs)	Salivary Antigens Detected	Secretor Status	ABO Inference	Interpretation
(–)	(–)	(–)	A, B, H	Secretor	AB	Complete inhibition of anti-A, anti-B, and anti-H indicates presence of all antigens in saliva
(–)	(+)	(–)	A, H	Secretor	A	Inhibition of anti-A and anti-H; anti-B remains active
(+)	(–)	(–)	B, H	Secretor	B	Inhibition of anti-B and anti-H; anti-A remains active
(+)	(+)	(–)	H only	Secretor	O	Only anti-H inhibited; confirms presence of H substance
(+)	(+)	(+)	None	Non-secretor	Not determined	No inhibition; absence of salivary ABO antigens
(–)	(–)	(+)	A, B (low H)	Secretor	AB	Anti-A and anti-B inhibited; anti-H active due to low H level

Table 2. Comparison of caries score in primary and permanent teeth among participants according to secretor status.

Variable		Secretor N=81	Non-secretor N=7	p-value
DMFT (permanent)	Mean $\pm$ SD	2.44 $\pm$ 2.025	1.43 $\pm$ 1.397	0.177
	Median (Range)	2 (0-10)	1 (0-4)	
dmft (primary)	Mean $\pm$ SD	5.2 $\pm$ 4.032	3.86 $\pm$ 3.671	0.461
	Median (Range)	6 (0-17)	2 (0-8)	

p-value by Mann-Whitney U test

Table 3. Comparison of caries score in primary and permanent teeth among participants according to ABO blood groups.

p-value by Kruskal-Wallis H test

Variable		A N=14	B N=36	O N=25	AB N=6	p-value
DMFT (permanent)	Mean $\pm$ SD	2.14 $\pm$ 1.51	2.19 $\pm$ 1.82	2.76 $\pm$ 2.59	3.33 $\pm$ 1.51	0.542
	Median (Range)	2 (0-4)	2 (0-7)	2 (0-10)	3 (2-6)	
dmft (primary)	Mean $\pm$ SD	6.79 $\pm$ 5.09	5.36 $\pm$ 3.94	4.20 $\pm$ 3.59	4.67 $\pm$ 3.14	0.359
	Median (Range)	7 (0-17)	6 (0-14)	5 (0-13)	3 (2-10)	