The Clinical Study of Detecting the Endodontic Vitality Clinical Reference Range of Anterior Teeth in Children Used By Laser Doppler Flowmetry

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Research Article

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Abstract

Aim: To measure the pulpal blood flow (PBF) of the upper incisors using the laser Doppler flowmetry (LDF) and to calculate the endodontic vitality clinical reference range using PBF as an indicator in the healthy children. To clarify the factors that might interfere the detection and interpretation of the results.

Methods: Seven to twelve-year-old school-age children were recruited randomly. The basic characteristics of the subjects are described in terms of age and gender. The mean, standard deviation, and reference range of the subjects' PBF values are described based on their age, sex, and tooth position. Multi-factor analysis of variance was implemented, mainly including age, gender, and tooth position.

Results: 1) The clinical reference range of the anterior teeth PBF in children of different ages was preliminary defined that was approximately among the range of 7-14. 2). A statistically significant correlation between PBF and age in school-aged children ($P < 0.05$) was detected and no statistical significance between genders ($P > 0.05$) was detected. The PBF detection value of lateral incisors was higher than that of central incisors, and the difference was statistically significant ($P < 0.05$).

Conclusion: The determination of the range of clinical reference of anterior teeth PBF in children by LDF could provide a good theoretical basis and reference basis for further promotion of its application.

Introduction

Dental trauma and dental pulp diseases are common and difficult problems in dental diagnosis and treatment. The reasonable formulation of the diagnosis and treatment plan depends largely on the accurate judgment of the state of the dental pulp [1]. The accurate dental pulp vitality detection is more conducive to reserve the dental pulp to the hilt so as to promote the continuous development of tooth roots since the young permanent teeth have not yet been fully developed [2]. In addition, orthodontic treatment, whitening, repair or revascularization of teeth also requires accurate determination of pulp vitality. The common methods for the detection of pulp vitality in clinical practice currently fall into two major categories, namely, temperature sensing and electrical vitality testing [3]. Although these detection methods have a long history of application, they still cannot fully meet the actual clinical needs. In particular, the young permanent teeth [4] cannot form a high-resistance loop due to the open apex and the early stage of dental trauma [5] is mostly in the shock phase which might result in the spurious results or no reaction absolutely to the sensory test methods frequently. In addition, the children's coordination is poor due to their high sensitivity to painful stimuli especially after the tooth trauma, so it is difficult to apply sensory tests to obtain accurate subjective feedback. The inaccurate judgment of the dental pulp condition poses a great challenge to the development of the clinical diagnosis and treatment plan. It will increase the fragility of the teeth to remove the dental pulp blindly during treatment and increase the risk of future dental fractures [6]. However, blindly-preserved dental pulp may cause some complications due to necrosis of the dental pulp leading to irreversible internal and external absorption of roots and then lead to the tooth loss and other serious complications [7]. Therefore, whether or not dental pulp could be accurately determined will directly affect the diagnosis, treatment and prognosis of the suffering teeth.

Extensive research on dental pulp vitality testing methods have been conducted which proposed a completely new concept that the decisive factor that influenced the pulp vitality was the blood circulation system rather than the nervous system [8–10] which opened up a new direction for dental pulp vitality detection. Laser Doppler Flowmetry (LDF) is one of the outcomes under the guidance of this new concept which have been researched relatively mature. The principle of LDF is using the helium-neon or semiconductor diode as the laser light source and the reflected light of frequency drift caused by the movement of blood cells according to the doppler phenomenon would be absorbed, and then the changes in the flow and the flow rate of the blood cells according to the intensity and the frequency drift are detected [11]. The test object of LDF is the local microcirculation system of the teeth [12] which could reflect the state of pulp vitality through the change of PBF. The characteristics of LDF are non-invasive, painless, objective and non-radioactive [13] which can effectively avoid the pseudo-response caused by nerve shock or hypersensitivity. The test results are not affected by the subjective will of the tester or the subject [14] and the output result is displayed as an intuitive data and waveform which could help to judge the state of the dental pulp accurately by reading the value immediately. The operation is simple and easy to be accepted by the subject especially children with dental diseases.

A high sensitivity and specificity [15–17] in the detection of pulp vitality by LDF have been recognized by majority of scholars after continuous research and development. Studies have shown [18] that LDF had a wide range of clinical applications such as the dental pulp vitality detection for the teeth after trauma even though the traumatic teeth were still in the endothelium shock period and could be used in the young permanent teeth and temporary teeth and many more. There is an irreplaceable advantage of LDF in the detection of pulp vitality in primary teeth and young permanent teeth compared to the traditional methods and LDF can provide a powerful reference for the diagnosis and treatment of young permanent teeth and traumatic teeth. In a word, there is a wide range of application prospects in the field of children's dental medical treatment.

However, although LDF has a history of nearly 50 years, it still had not been widely used in clinical practice which is mainly because of the lack of standard clinical reference range in its application process [19]. Many factors and difficulties may affect the establishment of the reference range of the pulp vitality measured by LDF such as the consistency of the detection accuracy of different types of instruments, the anti-interference performance of the instrument itself, the lack of standardized detection methods, the large sample size required for research, the randomness of the sample size determination and the difficulty of the exclusion of bias and so on. This present study obtained a stable clinical reference value through developing a standardized testing process to collect the anterior teeth PBF of adolescents in the Han nationality. The required sample size was calculated basing on the pre-experimental results and using a uniform testing instrument by the same tester. The reference value would provide a good foundation for LDF's extensive clinical application.

Materials And Method

1.1 Objects

This study was approved by the Ethical Committee of Stomatological Hospital of the Air Force Military Medical University (grant number IRB-REV-2016044), and the healthy 7-12-year-old school-age children who were voluntary willing to participate in the project were recruited open to the public. The inclusion and exclusion criteria of this study were formulated according to the application precautions of LDF, and the target population that met the experimental requirements was screened. The experimental method and purpose was informed to the subjects and their guardians and the written informed consent was obtained from the guardians, and meanwhile, the subject's medical personal file was established respectively.

1.1.1 Inclusion criteria

1) Physical and mental health, no systematic, infectious or genetic diseases;

2) No history of anterior tooth trauma, surgery, repair or orthodontic treatment;

3) No obvious pigmentation, caries, crowding or malocclusion;

4) The eruption height of the test teeth meet the testing needs that the occlusal surface is at least 5mm above the gingival margin, the periodontal and oral health is good;

5) Subjects can effectively cooperate to complete the test.

1.1.2 Exclusion criteria

1) Don’t meet the inclusion criteria;

2) Suffering hematological or other systemic illnesses;

3) Use the cardiovascular drugs recently;

4) Sensitive to the testing materials;

5) Incomplete collection of the basic information or refuse to participate the test.

1.2 PBF data acquisition

In order to unify the standard, all the data collected in this study were taken under the room temperature (20°C) and the subjects were kept in a seated and calm state. A silicone rubber impression (3M□ExpressTMSTD□3M ESPE Dental Products□Minnesota, USA) was made for the subject after the maxillary anterior testing area was cleaned thoroughly. The thickness of the lip side of the impression was better to maintain about 3-5mm so that an effective retention for the probe could be formed to enhance the stability of the detection after punching. The position of drilling corresponding to the test teeth in the impression was situated in the middle of the tooth surface and distanced from the gingival margin about 3-5mm [20], and the diameter of the drilling hole was consistent with the probe. The subjects were asked to keep in the seated, calm and relaxed state after the LDF was preheated and calibrated, and then the silicone rubber impression was reset to the detected area in the test area (Fig 1[21]). Test subjects were asked to maintain normal and steady breath and keep head static as far as possible in order to ensure the stability of the detected data. Generally, the time for detection was 1-3min after which the output waveform was relatively stable, and the test results were taken from a relatively stable waveband which the required time was not less than 30s and SD < 4 in the output result was regarded as a valid testing result. The PBF of the 12-22 teeth would be detected following this step in turn and the test results were recorded as mentioned above. The subjects were asked to have a quiet rest for 10 minutes after the test was completed and then the above test would be repeated and the corresponding record was recorded as the second measurement. The mean value the two tests was considered as the final results. Finally, photos of the maxillary anterior teeth and root tip films were taken. The instruments used in this study were LDF (Perimed, PF 5001, Perimed AB, Stockholm, Sweden) and LDF probe (Perimed, DP 416, Perimed AB, Stockholm, Sweden).

1.3 Statistical Analysis

The test results were statistically analyzed using SPSS22.0 software. The basic characteristics of the subjects are described in terms of age and gender. The mean, standard deviation, and reference range of the subjects' PBF values are described based on their age, sex, and tooth position. Multi-factor analysis of variance was implemented, mainly including age, gender, and tooth position; the test level α was set to 0.05.

Results

2.1 Sample information

Table 1
General characteristics of the sample population

gender			
	male	239	52.53%
	female	216	47.47%
age			
	7	95	20.88%
	8	104	22.86%
	9	127	27.91%
	10	67	14.73%
	11	38	8.35%
	12	24	5.27%

According to the inclusion and exclusion criteria, 455 (male 239 and female 216) were included in the study finally. The age distribution of the sample population is shown in the Table 1. There were 909 central incisors and 785 lateral incisors (some of the subjects had no lateral incisors or the eruption height could not meet the requirement for detecting) which met the experimental sample size requirements.

2.2 Clinical Reference Range

The PBF detection values of 12–22 teeth in different sex samples were statistically described and the clinical reference range of the anterior teeth PBF in children aged 7–12 years was calculated in Table 2, 3 and 4:

Table 2
PBF values of maxillary anterior teeth in females of different ages

age	tooth position											
	11			12			21			22		
	Mean	SD	Reference range	Mean	SD	Reference range	Mean	SD	Reference range	Mean	SD	Reference range
7	9.94	2.13	(5.76,14.11)	8.56	1.49	(5.64,11.48)	8.59	1.61	(5.44,11.73)	9.74	1.62	(6.57,12.91)
8	10.38	1.78	(6.89,13.86)	8.77	1.42	(5.98,11.55)	8.68	1.68	(5.38,11.97)	10.25	1.85	(6.62,13.88)
9	10.66	2.00	(6.73,14.58)	9.14	1.50	(6.20,12.08)	9.06	1.40	(6.31,11.81)	10.82	1.79	(7.30,14.33)
10	10.78	1.87	(7.12,14.45)	9.27	1.66	(6.02,12.53)	9.31	1.58	(6.22,12.41)	10.41	1.93	(6.63,14.20)
11	11.50	1.99	(7.61,15.40)	9.26	1.40	(6.51,12.01)	9.48	1.59	(6.36,12.59)	11.26	1.93	(7.48,15.05)
12.00	9.79	2.94	(5.12,14.47)	8.25	1.12	(6.47,10.03)	7.83	1.04	(6.18,9.48)	9.26	2.08	(5.95,12.57)

Table 3
PBF values of maxillary anterior teeth in males of different ages

age	tooth position											
	11			12			21			22		
	Mean	SD	Reference range	Mean	SD	Reference range	Mean	SD	Reference range	Mean	SD	Reference range
7	8.67	1.16	(6.39,10.95)	8.52	1.41	(5.75,11.29)	8.54	1.27	(6.04,11.03)	8.85	1.11	(6.67,11.03)
8	9.93	1.62	(6.76,13.10)	8.99	1.62	(5.82,12.15)	8.99	1.56	(5.94,12.04)	9.97	2.03	(5.99,13.94)
9	10.47	1.90	(6.76,14.19)	8.95	1.48	(6.05,11.85)	9.05	1.27	(6.57,11.54)	10.62	1.82	(7.05,14.18)
10	10.87	1.94	(7.08,14.66)	9.38	1.52	(6.40,12.37)	9.49	1.62	(6.31,12.67)	11.04	2.01	(7.09,14.99)
11	10.54	1.47	(7.65,13.42)	8.91	1.17	(6.61,11.20)	8.93	1.27	(6.44,11.42)	10.56	1.78	(7.06,14.05)
12.00	11.56	1.52	（10.28,12.83）	9.14	1.52	（7.87,10.41）	9.12	0.89	（8.37,9.86）	11.26	1.91	（9.66,12.85）

Table 4
PBF values of maxillary anterior teeth in children diffrent ages

age	tooth position											
	11			12			21			22		
	Mean	SD	Reference range	Mean	SD	Reference range	Mean	SD	Reference range	Mean	SD	Reference range
7	9.34	1.83	(5.74,12.93)	8.54	1.41	(5.77,11.31)	8.57	1.27	(6.07,11.06)	9.32	1.11	(7.14,11.50)
8	10.07	1.62	(6.90,13.24)	8.90	1.62	(5.74,12.07)	8.86	1.56	(5.81,11.90)	10.06	2.03	(6.08,14.04)
9	10.53	1.90	(6.82,14.25)	9.03	1.48	(6.12,11.93)	9.04	1.27	(6.56,11.53)	10.69	1.82	(7.12,14.25)
10	10.79	1.94	(6.99,14.58)	9.32	1.52	(6.34,12.31)	9.44	1.62	(6.25,12.62)	10.77	2.01	(6.82,14.72)
11	11.12	1.47	(8.24,14.00)	9.14	1.17	(6.85,11.44)	9.31	1.27	(6.82,11.79)	10.97	1.78	(7.48,14.47)
12.00	10.97	2.14	(9.61,12.33)	8.84	1.41	(7.95,9.74)	8.69	1.09	(7.99,9.38)	10.59	2.11	(9.25,11.93)

2.3 Factors Affecting PBF

A multi-factor analysis of variance was performed on the PBF data of all survey subjects(Table 5). The results showed that the difference in PBF values of children's deciduous teeth between different genders was not statistically significant (p > 0.05); there were differences between different ages (p < 0.001), central incisor The mean PBF increased with age, and gradually decreased after 10 years of age and stabilized. The PBF of the lateral incisor also showed a trend similar to that of the central incisor, and gradually decreased after the age of 11 and stabilized (Fig. 1); PBF values of different tooth positions are also different. Specifically, the PBF differences between teeth 11 and 21, and teeth 12 and 22 are not statistically significant (P 0.05), while the PBF value of central incisors is lower than that of lateral incisors The difference in PBF detection value was statistically significant (P < 0.001). In addition, the interaction effect between gender and age factors is significant (p < 0.01), that is, among sample populations of different genders, age factors have different effects on PBF values. The mean PBF of the central incisor continues to increase with age, and gradually declines and stabilizes after the age of 10 years. The PBF of the lateral incisor also shows a trend of outcome similar to that of the central incisor, and gradually decreases after the age of 11 and tends to Stable (Fig. 1). The conclusion that the PBF detection value of young permanent teeth is significantly correlated with age is further verified, and it is speculated that PBF is closely related to the degree of root development. The author also performed a statistical analysis of the root development of the upper central incisor and lateral incisor of the children in this experiment. The trend chart of the change with age is shown in Fig. 2.

Table 5
Multivariate analysis of variance values of the LDF

Factor	Type III sum of squares	df	Mean square	F	P
Correction Model	1196.650	47	25.461	9.172	.000
Intercept	108963.355	1	108963.355	39252.815	0.000
gender	2.014	1	2.014	.725	.395
age	210.227	5	42.045	15.146	.000
position	539.876	3	179.959	64.828	.000
gender * age	48.140	5	9.628	3.468	.004
gender * position	5.743	3	1.914	.690	.558
age * position	39.010	15	2.601	.937	.522
gender * age * position	22.799	15	1.520	.548	.914
Error	4569.193	1646	2.776		
Total	163071.192	1694			
Total number of post-correction	5765.842	1693			

2.4 PBF and Root Development

A statistical analysis of tooth root development among the children aged 7–12 years in this study was made considering that the degree of tooth root development in this age group was quite different and the degree of root development was closely related to the volume of the medullary cavity which would be closely related to the PBF detection value. The standard of grouping was according to the method of Nolla subsection that as: the roots form 2/3, horn-shaped apex was group 1, the roots close to fully developed but the aperture opening was group 2, the roots fully developed was group 3 and the subject of statistics was the anterior root tip slices of the group of study samples. The distribution of tooth root development was shown in Fig. 2. The results showed that the degree of individual development of tooth roots in children was relatively large that with more than 50% of the central incisors was developed completely at the age of 10 years and this phenomenon of the lateral incisors was 11 years old. The development of lateral incisors was lagged behind central incisors and the root canal diameters of lateral incisors in the same age were larger than those in the central incisors. This individual difference might be the factor that led to a corresponding trend of PBF with age and a difference between PBFs in the central incisors and lateral incisors.

Figure 1 shows the trend of the PBF of 12/11/21/22 teeth with ages that the PBF gradually increased with age and then tended to be stable or decreased slightly.

Figure 2 shows the changes about root development of incisors with ages that the individual differences in each age were obvious regardless of the central incisors(A) or lateral incisors(B).

Discussion

In this study, the range of the PBF clinical reference for the anterior teeth of healthy children in different age groups was sought to be explored. The possible factors that might affect the PBF in these age groups included gender, tooth position, root development, age and the interference of the gingival blood flow signals, etc. The anterior teeth erupt at 7–8 years old and the roots develop completely at about 11–12 years old. 7–12 year-old school-age children were selected as the research objects in this study aiming to explore the relationship between the anterior teeth PBF and age, gender, tooth position and root development in different age groups. The children was grouped by age as each year a group because the analysis of the degree of root development showed that the degree of development of the root at certain ages could not be generalized in this age group, and the difference in PBF grouped according to the degree of root development was still required further exploration and research. The clinical reference ranges of anterior teeth PBF in children with young permanent teeth and the permanent teeth with stable root were determined.

As can be seen from the results of the study, there was no statistical significance between different genders in the 7-12-year-old group. The reasons for this phenomenon were considered might mainly relate to the different blood circulation system indicators of genders with age for the subjects because the detected object of LDF was the changes of blood flow velocity in the pulp cavity capillary which would change with the body's own blood pressure, heart rate and so on [22]. It had been documented that the detection of pulp vitality by LDF was not suitable for patients with hypertension, obvious anemia and other hematological system diseases [23]. There are differences between different genders on the blood pressure and blood cell contents. Some domestic scholars have shown that there was no difference in blood pressure between male and female [24] when the age of children were stratified by 7 to 11 years old. However, this conclusion was not the same among different ethnic groups. A study [25] showed that the mean blood pressure of girls was slightly higher than the mean among children aged 7–12 years. In addition, considering the age-related changes of root development in children, the comparative analysis of the root development of children showed that there was no statistical significance in the developmental level of root in this age group between different genders. Therefore, it could be considered that the range of clinical reference for the anterior teeth PBF was the same for both male and female of the children group, but whether or not the results would be applicable to the different races still required further research.

There was a statistical significance of PBF detection value between the central incisors and lateral incisors of the same gender and the results showed that the PBF of lateral incisor was slightly higher than that of the central incisors in the children group. This result was considered that might mainly relate to the changes of the shape of the anterior pulp cavity with age. It could be seen from the root canal development with the trend of age in the 7–12 years old that the root development of lateral incisors was lagging to the central incisors and the pulp cavity was in a relatively more open state. The larger the volume of the pulp cavity is, the more the pulp blood flow will be in the unit time. Therefore, the volume of the pulp cavity of the lateral incisors was relatively greater than that of the central incisors in the age group of 7–12, while the volume of the central incisors would be greater than lateral incisors with the gradual development of the root apical foramen closed in the youth group and the changes of PBF also showed the same trend. However, the change of PBF could not be completely explained by the degree of root development simply because the trend of PBF of 12–22 teeth was not performed as the higher the proportion of trumpet-shaped was, the higher PBF detection value was but showed a trend of increasing gradually and then being steady or slightly declining with age. Comprehensive analysis to consider this phenomenon might be related to the height of the eruption of teeth and the location of the gums. Specifically, the eruption height increased gradually and the gingival position gradually receded with age that would change the position where the LDF probe should be placed on the teeth indirectly which was equivalent to decrease the amount of laser light transmitted into the marrow cavity because of the detection position was high deviated to the edge of the teeth early. With the continuous eruption of the teeth and the gradual retraction of the gingival position, the position of the probe gradually reached to the standard testing position and the amount of laser light transmitted into the pulp cavity increased resulting in an increase of the doppler shift which led to the phenomenon that the PBF detection value showed a gradually increasing trend. At the same time, the blood flow into the pulp cavity would gradually decrease after the root gradually developed and the doppler shift generated by the same laser transmission would also gradually decrease. There was a "turning point" which was when most of the roots developed completely in the curve of the PBF with age. Therefore, we can consider that the factors affected the PBF in children were not only the local microcirculation system factors of the tooth itself, but the degree of root development, location of gingiva and other comprehensive factors.

This study also found a statistically significant correlation between PBF and age at ages 7–12. This phenomenon was considered might be related to the degree of root development and the location of the gingiva and the principle of its influence might be similar to the related impact mentioned above. The vast majority of anterior teeth have developed at the age about 20 and the PBF changes have been relatively stable. Regardless of the female or male sample, the PBF detection value of central incisors of children was slightly lower than that of the youth group and the difference was statistically significant. A comprehensive analysis of age-related changes in blood pressure and root canal diameter was conducted in response to this phenomenon. Some studies found [26] that the age-related changes in root canal of female were faster than that of male but the average root canal diameter was larger than that of male which might lead the result that the PBF among female group was slightly higher than male. However, the level of blood pressure in female increasing gradually with age was slightly slower than that in male and the variation in female lateral incisors was more common. Therefore, many possible and complicated factors might lead to the diffrence between male and female group, the specific micro-mechanism was not fully clear and further exploration and research were still required.

In conclusion, the determination of the clinical reference range of PBF for young adolescents of different ages in this study would effectively fill the gap that traditional pulp vitality detection methods could not accurately judge the status of pulp of the young permanent teeth. At the same time, we also applied LDF to the detection of dental pulp vitality in children with traumatic injury so as to further validate the reliability of the reference range and provide a reference for its further research and the expansion of its clinical application. However, some studies have shown that the usage of LDF had some limitations and clinically uncontrollable factors [27] such as the individual differences of PBF and vascular nerve responses, different optical characteristics of tooth structure, ethnic differences, tooth morphology, and periodontal tissue, etc. and this conclusion also needed more clinical studies to verify. Some research showed that LDF had a certain limitation in the elderly population. The PBF changes with the age of the order of magnitude was very small according to the pulp cavity volume of the elderly decreased in addition to the pulp cavity around the lower blood flow velocity and other factors which determined that LDF was not suitable for elderly people with age-related changes in the pulp cavity [28] while the exact age limit was still unclear. Considering the age-related changes in the root canal, the clinical reference of PBF in more relevant age groups should be obtained in subsequent studies to further improve the conclusion. In addition, the application of LDF in the detection of dental pulp vitality can also be explored accordingly to give full play to its objective and accurate superiority in the field of children's oral diagnosis and treatment.

Conclusions

There was a unique advantage and a wide development prospect for the detection of pulp vitality by LDF especially in the field of children's oral medical. The determination of its clinical reference value was conducive to provide a powerful reference for clinical diagnosis and treatment. Of course, the results of this study were still needed to further verify whether the range was same in other ethnic groups and regions and the range of PBF reference value of the anterior teeth among other age groups was still required further explorations and studies.

Declarations

Ethical approval:

"All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards."

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Figures

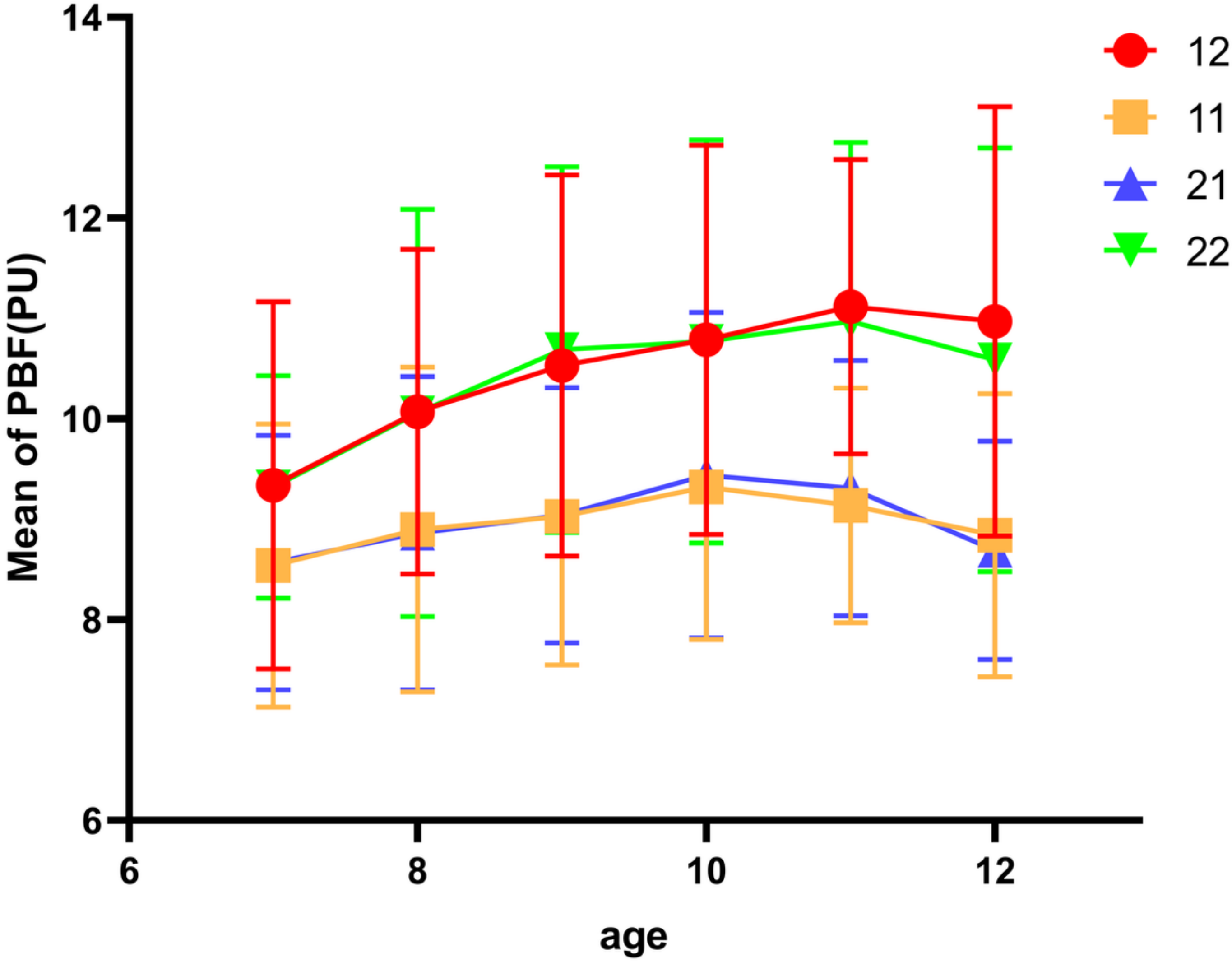

Figure 1

The trend of PBF value in children of different ages

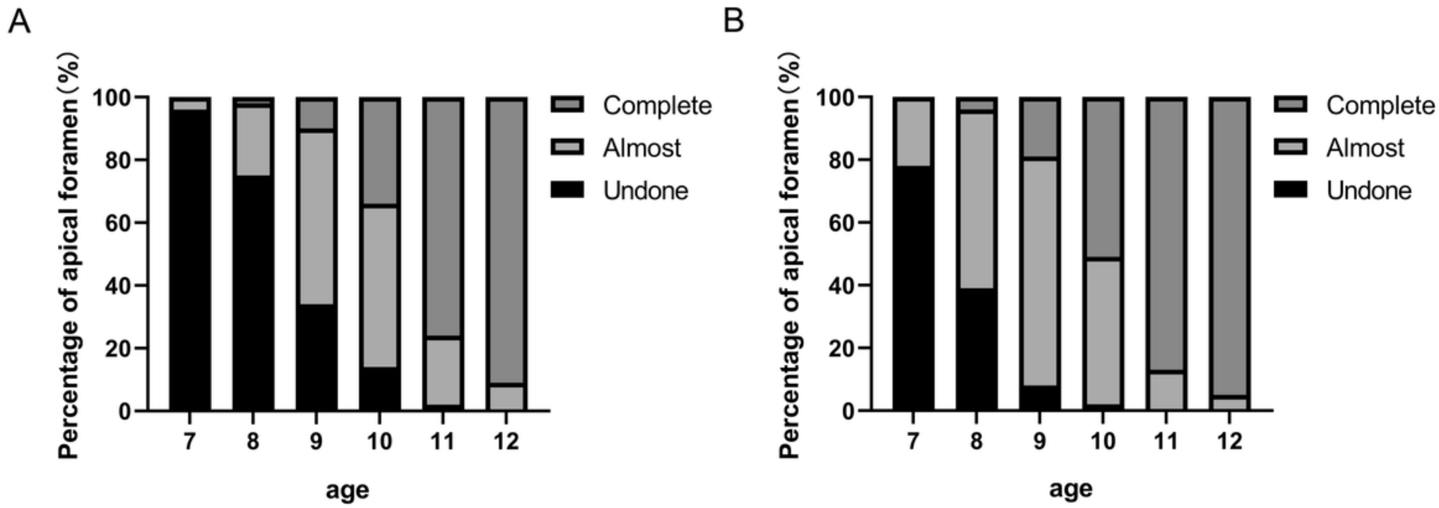

Figure 2

Apical foramen of central incisors and lateral incisors in children of diffrent ages