




Article

Gingival Pigmentation Affected by Smoking among Different Age Groups: A Quantitative Analysis of Gingival Pigmentation Using Clinical Oral Photographs

Tomotaka Kato ^{1,*}, Shinsuke Mizutani ¹, Hiroya Takiuchi ¹, Seiichi Sugiyama ², Takashi Hanioka ³  and Toru Naito ^{1,*}

¹ Section of Geriatric Dentistry, Department of General Dentistry, Fukuoka Dental College, Fukuoka 814-0193, Japan; mizutani@college.fdcnet.ac.jp (S.M.); takiuchi@college.fdcnet.ac.jp (H.T.)

² Sugiyama Dental Clinic, Yachiyo 276-0027, Japan; sdcss@pastel.ocn.ne.jp

³ Section of Oral Public Health, Department of Preventive and Public Health Dentistry, Fukuoka Dental College, Fukuoka 814-0193, Japan; haniokat@college.fdcnet.ac.jp

* Correspondence: tkatou@college.fdcnet.ac.jp (T.K.); naito@college.fdcnet.ac.jp (T.N.); Tel.: +81-92-801-0411 (T.K. & T.N.)

Academic Editor: Zubair Kabir

Received: 14 June 2017; Accepted: 1 August 2017; Published: 4 August 2017

Abstract: The presence of any age-related differences in gingival pigmentation associated with smoking, particularly in a young population, remains to be fully investigated. The purpose of this study was to determine the age-related differences in smoking gingival pigmentation. Gingival pigmentation was analyzed using the gingival melanosis record (GMR) and Hedin's classification with frontal oral photographs taken at 16 dental offices in Japan. Participants were categorized into 10-year age groups, and their baseline photographs were compared. In addition, to evaluate the effect of smoking cessation on gingival pigmentation, subjects were divided into a former smoker group (stopped smoking) and current smoker group. A total of 259 patients 19 to 79 years of age were analyzed. People in their 30s showed the most widespread gingival pigmentation. In addition, subjects in their 20s showed a weak effect of smoking cessation on gingival pigmentation. These findings suggested that the gingival pigmentation induced by smoking was more remarkable in young people than in middle-aged people. This information may be useful for anti-smoking education, especially among young populations with a high affinity for smoking.

Keywords: gingival melanosis; age difference; smoking; clinical oral photographs

1. Introduction

Smoking is known to cause health problems [1]. Recently, there have been many reports on the effects of secondhand smoking in children, underscoring the severity of smoking in children [2–5]. In the field of oral health, the effects of secondhand smoking in children have also begun to be described [6,7]. Hanioka et al. reported the association of melanin pigmentation in the gingiva of children with parents who smoke [6]. Therefore, proactive efforts toward smoking cessation in young populations must be enacted in not only the medical field but also the dental field.

In the general dental field, one side effect of smoking is gingival pigmentation. Gingival pigmentation can be induced by not only smoking, but also by tattoos, and they can lead to both toxicity and systemic diseases [8,9]. Gingival pigmentation develops following the accumulation of melanin, which is produced by melanocytes [10,11]. Hedin et al. first reported that smokers showed greater gingival pigmentation than non-smokers [12]. In addition, it was reported that quitting smoking

resulted in a decrease in gingival pigmentation in qualitative studies [12–14]. Recent studies have evaluated gingival color using a dental photoelectric colorimeter [15,16].

However, the relationship between the duration of smoking cessation and reduction of gingival pigmentation is unclear at present. It is difficult to evaluate gingival pigmentation quantitatively, and gingival pigmentation has traditionally been analyzed in a subjective manner. Ono reported the quantitative evaluation of gingival pigmentation in their study [17]. However, a special device was required to evaluate the gingival pigmentation in those reports, and its reproducibility was unclear.

Therefore, we developed a new assessment method—the gingival melanosis record (GMR)—that evaluates gingival pigmentation quantitatively using common devices and examined the relationship between smoking cessation and reduction of gingival pigmentation [18]. Given that the presence of age-related differences in smoking gingival pigmentation, particularly in a young population, has been unclear, we attempted to describe the influence of smoking and the effects of smoking cessation on gingival pigmentation in different age groups.

2. Materials and Methods

2.1. Patients and Ethical Considerations

The subjects were 283 smoker patients who visited 16 general dental offices that were members of the Japan Health Care Dental Association. The subjects were divided into current smokers and former smokers. “Current smokers” were those who had smoked greater than 100 cigarettes in their lifetime and had smoked in the previous 30 days, and “former smokers” were those who had smoked greater than 100 cigarettes in their lifetime but had not smoked in the last 30 days, according to the Japanese Administration of Health, Labor, and Welfare Ministry. They visited these dental offices for a regular checkup and had a series of oral photographs taken. In total, more than 900 oral photographs were analyzed.

This study was approved by the Ethics Committee for Clinical Research at Fukuoka Dental College (approval number 194). To maintain anonymity, patients were given an identification number. Therefore, analysts did not have to use patients’ individual information. All patients provided their informed consent in writing.

2.2. Measurement of the Gingival Color

Gingival pigmentation was analyzed using frontal oral photographs taken with non-contact-type dental cameras. These cameras were commonly available, and frontal oral photographs were obtained using standardized techniques. All oral photographs were reviewed on a monitor with a liquid crystal display (Multi Sync LCD 2490WUXi2, NEC Corp., Tokyo, Japan), and the same estimator evaluated all of the photographs. These oral photographs were randomized, and the analyzed photographs were completely masked.

The frontal oral photographs were analyzed using the GMR for quantitative measurement as well as using Hedin’s classification for qualitative measurement [18]. The GMR determined the presence or absence of gingival pigmentation at the target site, which was defined as below (Figure 1). The GMR was used to evaluate the presence or absence of gingival pigment at the assessment sites, and it was represented as a percentage by dividing the pigmented assessment sites by all assessment sites [18].

Hedin’s classification uses the classical evaluation method of gingival pigmentation, and gingival pigmentation was qualitatively analyzed on a scale of 0 to 4 (Figure 2) [12]. The subjects of this study were applied to the samples in our previous study [18], and assessment procedures included confirmed standardized conditions and a validity check. To assess the validity of the GMR, we compared the GMR with Hedin’s classification, and intraclass correlation coefficients (ICC) were calculated to assess the GMR reproducibility. ICC (1,1) was used to evaluate repeatability by the same examiner, and ICC (2,1) was used to evaluate stability between examiners [18].

Defined line

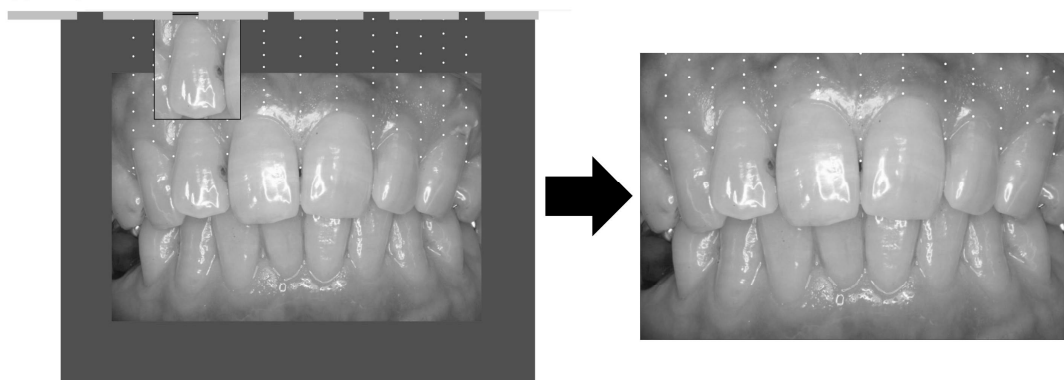


Figure 1. Measurement of gingival pigmentation (gingival melanosis record: GMR, Left was outline and right was evaluating coverage).

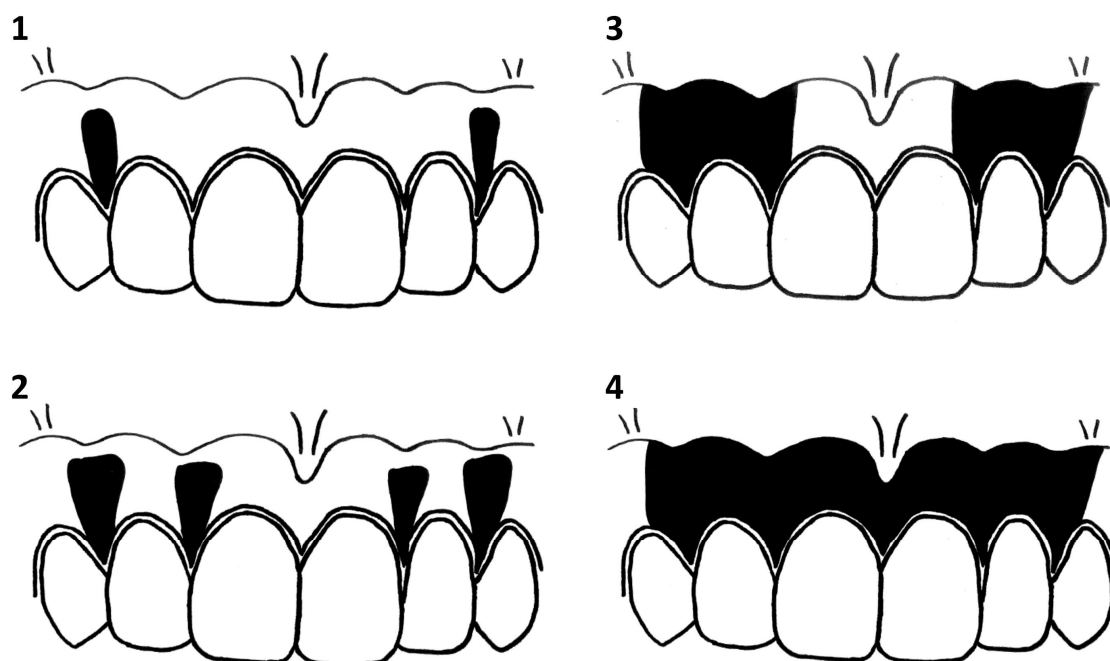


Figure 2. Degrees of gingival pigmentation (Hedin's classification). (1) A baseline was drawn horizontally on the maxillary gingiva at the crown length of the maxillary right lateral incisor. (2) Between the right and left maxillary canines, a vertical line was drawn from the baseline to the cervical line of each tooth. (3) Nine points were plotted to separate eight equal parts on the vertical line, and it was adopted that measuring sites were the points in the attached gingiva, and the vertical lines were placed at mesial and central and distal points of the frontal teeth. (4) The presence of pigmentation at the measuring sites was evaluated and the percentile with pigmentation was calculated for the gingival melanosis record (GMR).

2.3. Age-Related Differences in the Effect of Smoking Cessation on Gingival Pigmentation

The relationship between age and smoking history was analyzed using Spearman's rank correlation coefficient. To evaluate age-related differences in gingival pigmentation due to smoking, all patients were divided into subgroups by their age (10-year intervals), and baseline photographs were evaluated using GMR and Hedin's classification. To determine the age-related effect of smoking cessation on smoking-induced gingival pigmentation, subjects were separated into a former smoker group (stopped smoking) and current smoker group (presently smoking). In addition, the two groups

were divided into subgroups by age, and the baseline and follow-up photographs were compared using GMR and Hedin's classification.

2.4. Statistical Analyses

We conducted all analyses using the SPSS software program, version 22.0 (IBM, Tokyo, Japan). The significance level was set at 5%. An analysis of variance was used to evaluate the age differences at the baseline. The age-related differences in pigmentation between the former smoker group and current smoker group were analyzed by a paired *t*-test. Comparisons between the baseline and follow-up photographs of the former smoker group were also analyzed by a paired *t*-test.

3. Results

3.1. Subjects

The characteristics of the study subjects are shown in Table 1. A total of 259 patients (mean age 45.9 years; range 19–79 years, 126 current smokers, and 133 former smokers) attended the baseline examination. Twenty-four patients were excluded because of unreadable photographs or the absence of routine checkups. No statistically significant differences between the current smoker group and former smoker group were observed in the baseline characteristics and GMR. A significant correlation was found between the duration of smoking and subject age using Spearman's rank coefficient ($p < 0.01$, correlation coefficient = 0.85). There were no statistically significant differences among subgroups for the follow-up period or duration of smoking cessation (Table 2).

Table 1. Demographic characteristics of the patients at baseline.

Patient Characteristics	Former Smokers	Current Smokers	Total	<i>p</i> Value
Number of subjects	133	126	259	
Male/Female	88/45	79/47	167/92	0.30
Age (years)	46.8 ± 13.4	44.6 ± 13.0	45.9 ± 13.2	0.60
Duration of smoking (years)	27.3 ± 12.6	27.7 ± 12.4	27.5 ± 12.5	0.65
GMR (%)	26.0 ± 18.4	27.5 ± 21.1	26.7 ± 19.8	0.89
Follow-up period (years)	4.50 ± 2.15	3.79 ± 1.17	4.15 ± 1.77	0.01
Duration of smoking cessation (years)	3.14 ± 1.95	-		

Table 2. Demographic characteristics of the subgroups.

Patient Characteristics	Age of Former Smokers (Years)					Age of Current Smokers (Years)				
	19–29	30–39	40–49	50–59	≥60	19–29	30–39	40–49	50–59	≥60
Number of subjects	13	32	29	32	27	17	26	32	36	15
Male/Female	8/5	23/9	21/8	16/16	20/7	11/6	18/8	19/13	23/13	8/7
Duration of smoking (years)	9.5	18.3	26.1	32.5	41.4	9.0	17.9	27.7	37.2	43.3
Follow-up period (years)	5.01	4.40	4.73	4.19	4.47	3.60	4.00	3.52	3.78	4.22
Duration of smoking cessation (years)	2.62	2.75	3.31	3.38	3.67					

3.2. Smoking-Induced Gingival Pigmentation in Different Age Groups

The 259 patients were analyzed for their gingival pigmentation using GMR and Hedin's classification at baseline. Figure 3 shows the baseline GMR separated by age group. It was significantly associated on an analysis of variance ($p < 0.05$) in subgroups. The GMR was the highest among patients in their 30s with a decreasing trend after the 40s. Hedin's classification showed the same tendency, with patients in their 30s showing the highest proportion of scores of 3 and 4 among all age groups.

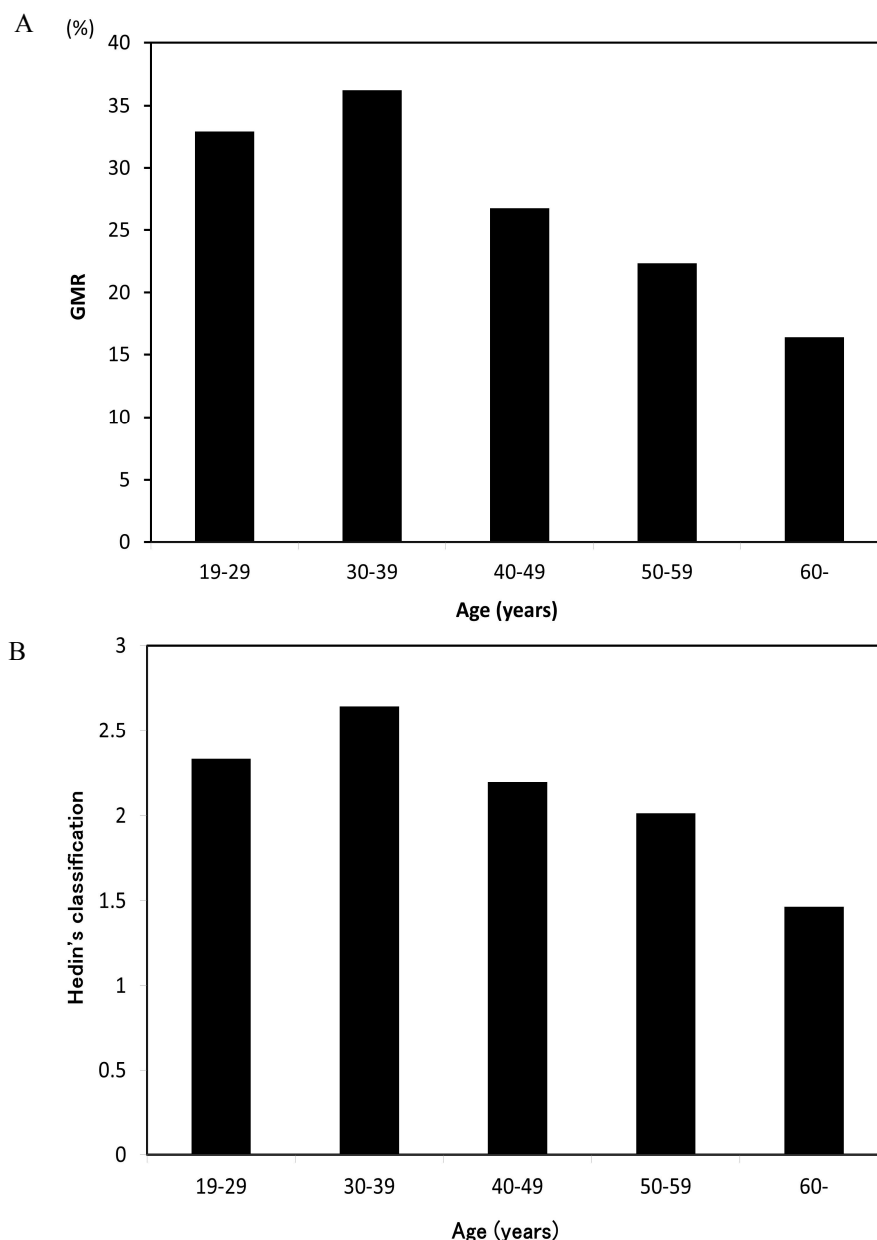


Figure 3. Mean GMR score (A) and Mean Hedin's classification (B) grouped according to age at baseline.

3.3. Age-Related Differences in Decreased Gingival Pigmentation Due to Smoking Cessation

The average follow-up period in the former smoking group was 4.50 ± 2.15 years, and the mean duration of smoking cessation was 3.14 years. In the current smoking group, the average follow-up period was 3.79 years. Figure 4 shows a comparison of the GMR between baseline and follow-up in both groups. The GMR decreased significantly with age in both groups, and Hedin's classification showed the same tendency. Each age group among former smokers showed less gingival pigmentation than the current smokers. Figure 5 shows the differences in the GMR at baseline and follow-up for each age group among current smokers. The GMR decreased with age, but the amount of change among those in their 20s was clearly less marked than in other age groups, and only those in their 20s showed no significant difference in the score between baseline and follow-up. The same tendency was noted in Hedin's classification, and the improvement of gingival pigmentation with smoking cessation was less obvious among those in their 20s than in other age groups.

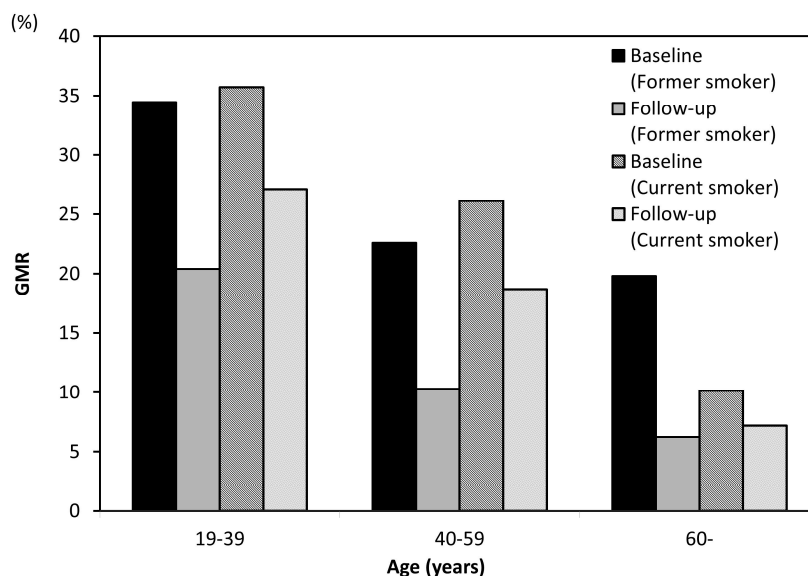


Figure 4. Mean GMR scores of former smokers vs. current smokers at baseline and follow-up.

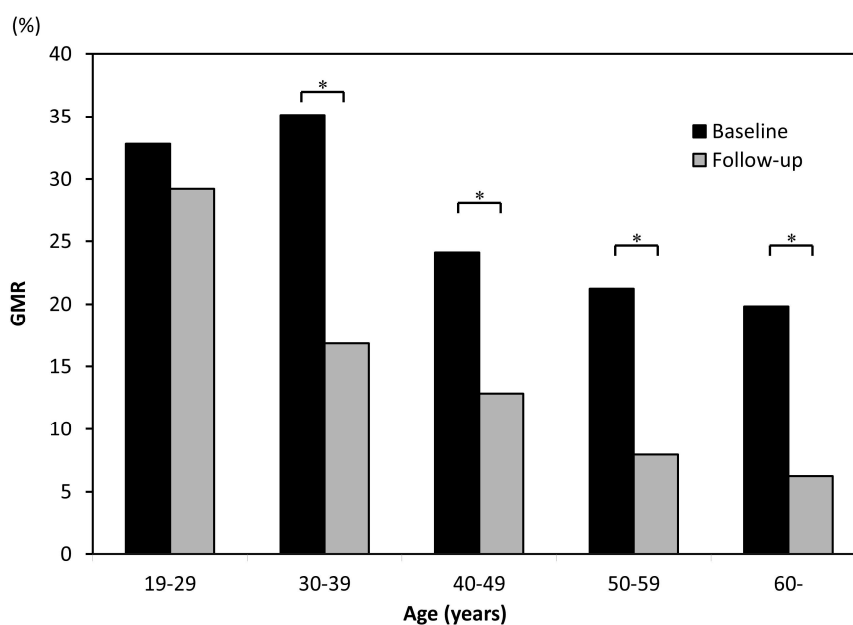


Figure 5. A comparison of the mean GMR scores of former smokers at baseline and follow-up, grouped according to age * $p < 0.05$ (paired t -test).

4. Discussion

In this study, we analyzed the gingival pigmentation of 259 patients from 19 to 79 years of age. The results showed that patients in their 30s had the most widespread gingival pigmentation. Furthermore, the effect of smoking cessation on gingival pigmentation was weak among those in their 20s. These findings suggest that smoking significantly affected gingival pigmentation in young people.

Regarding why these effects of smoking on gingival pigmentation were so much more marked in young people than in others, gingival pigmentation has a number of causes and develops due to the accumulation of melanin by melanocytes [10]. The activation of melanocytes can trigger the accumulation of melanin. Histologically [19], many reports have noted that young people have high melanocyte activity. As such, the widespread gingival pigmentation in young people may have been

due to their high activity of melanocytes. Reports from the dermatological field also suggest that young people have many melanocytes and high melanocyte activity [20,21], and we feel that these reports support our findings.

However, there have been a few reports of pigmentation increasing with age. For example, Fry reported that buccal pigmentation increased with age [22]. However, the participants in that study, which was conducted in Britain, were of Caucasian and African descent, while our study included only Japanese people. Differences in races or species seemed important concerning gingival pigmentation [23–25]. We therefore feel that our present findings are not necessarily negated by these previous ones.

Several limitations associated with the present study warrant mention. First, there was an evaluation limit for gingival pigmentation. Hedin's classification, which is the gold standard for analyzing gingival pigmentation, evaluates pigmentation on a scale of 0 to 4 in a subjective manner, and it is not a quantitative evaluation [12]. In contrast, GMR is a quantitative method of analyzing the gingival pigmentation that uses common logistics [18]. However, GMR requires a longer analysis time than Hedin's classification on a per capita basis. Therefore, a more refined method is yet needed. Second, information on the smoking amount was limited, and we were unable to determine the smoking index because subjects were unclear about the number of cigarettes smoked per day. However, there have been reports that gingival pigmentation has no relationship with the number of cigarettes smoked per day [26]. We therefore feel that this limitation does not deny our outcome.

This study found that younger patients showed more significant smoking gingival pigmentation than older ones. This result is very important for clinical practice. While the smoking rate is decreasing in developed countries year over year [27], the rate of young smokers has been on the rise for the past several years in some countries [28]. In addition, smoking in young people is associated with not only concerns for their own health but also that of potential children [2–5]. Therefore, antismoking education is a very important and pressing issue. In the dental field, describing the effects of smoking on gingival pigmentation is effective in educating patients on smoking cessation. Our findings may therefore expand anti-smoking education to include pointing out the effects of smoking on gingival pigmentation.

In addition, all age groups among former smokers showed less gingival pigmentation than the current smokers (Figure 4). However, current smokers also had decreased gingival pigmentation. The cause of decrease in the current smoker group was thought to be the quantity of smoking. Araki et al. reported that the prevalence of gingival pigmentation was significantly higher in smokers who smoked more than 10 cigarettes per day [29]. Additionally, the absence of oral pigmentation was recognized in a patient who initially smoked four cheroots per day but reduced the amount to one cheroot per day for nine years [14]. Taken together, these findings suggest that the quantity of smoking reduction might lead to a reduction in gingival pigmentation.

In order to further clarify the effects of smoking on gingival pigmentation, it is important to conduct long-term follow-up examinations of the gingival pigmentation. In the present study, the mean follow-up period was 4.15 years, which was relatively short; 5 or 10 years' follow-up may be better. Furthermore, increasing the number of subjects might better depict the effects of smoking cessation in the short term. Regardless, it is important to conduct further research in order to ensure that smoking cessation education has a sound scientific basis.

5. Conclusions

The present findings suggested an age-related difference in the smoking-induced gingival pigmentation. Gingival pigmentation due to smoking was more significant in younger patients than in older ones, and the reduction in the gingival pigmentation by smoking cessation was less marked in younger patients than in older ones. Therefore, anti-smoking programs should strongly target younger generations.

Acknowledgments: We thank the members and staff of the Japan Health Care Dental Association for providing and formatting the oral photographs and patient records. We also express our appreciation to Hidetoshi Akimoto

for coordinating the survey. We thank our colleagues for their support during the present study. A part of this work was supported by a Grant-in-Aid for Young Scientists (B: No. 26861826) from the Japan Society for the Promotion of Science.

Author Contributions: Tomotaka Kato and Toru Naito performed the experiments. Tomotaka Kato, Shinsuke Mizutani, and Hiroya Takiuchi analyzed the data. Seiichi Sugiyama managed the collected data. Tomotaka Kato, Shinsuke Mizutani, Takashi Hanioka, and Toru Naito wrote the paper. Tomotaka Kato and Toru Naito conceived and designed the experiments.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Katanoda, K.; Marugame, T.; Saika, K.; Satoh, H.; Tajima, K.; Suzuki, T.; Tamakoshi, A.; Tsugane, S.; Sobue, T. Population attributable fraction of mortality associated with tobacco smoking in Japan: A pooled analysis of three large-scale cohort studies. *J. Epidemiol.* **2008**, *18*, 251–264. [[CrossRef](#)] [[PubMed](#)]
2. Tabuchi, T.; Fujiwara, T.; Nakayama, T.; Miyashiro, I.; Tsukuma, H.; Ozaki, K.; Kondo, N. Maternal and paternal indoor or outdoor smoking and the risk of asthma in their children: A nationwide prospective birth cohort study. *Drug Alcohol Depend.* **2015**, *147*, 103–108. [[CrossRef](#)] [[PubMed](#)]
3. Tanaka, K.; Miyake, Y. Association between prenatal and postnatal tobacco smoke exposure and allergies in young children. *J. Asthma* **2011**, *48*, 458–463. [[CrossRef](#)] [[PubMed](#)]
4. Yamasaki, A.; Hanaki, K.; Tomita, K.; Watanabe, M.; Hasagawa, Y.; Okazaki, R.; Igishi, T.; Horimukai, K.; Fukutani, K.; Sugimoto, Y. Environmental tobacco smoke and its effect on the symptoms and medication in children with asthma. *Int. J. Environ. Health Res.* **2009**, *19*, 97–108. [[CrossRef](#)] [[PubMed](#)]
5. Yolton, K.; Dietrich, K.; Auinger, P.; Lanphear, B.P.; Hornung, R. Exposure to environmental tobacco smoke and cognitive abilities among U.S. children and adolescents. *Environ. Health Perspect.* **2005**, *113*, 98–103. [[CrossRef](#)] [[PubMed](#)]
6. Hanioka, T.; Tanaka, K.; Ojima, M.; Yuuki, K. Association of melanin pigmentation in the gingiva of children with parents who smoke. *Pediatrics* **2005**, *116*, 186–190. [[CrossRef](#)] [[PubMed](#)]
7. Tanaka, K.; Miyake, Y.; Arakawa, M.; Sasaki, S.; Ohya, Y. Household smoking and dental caries in schoolchildren: The Ryukyus child health study. *BMC Public Health* **2010**, *10*, 335. [[CrossRef](#)] [[PubMed](#)]
8. Çiçek, Y.; Ertas, U. The normal and pathological pigmentation of oral mucous membrane: A review. *J. Contemp. Dent. Pract.* **2003**, *4*, 76–86. [[PubMed](#)]
9. Moravej-Salehi, E.; Moravej-Salehi, E.; Hajifattahi, F. Relationship of gingival pigmentation with passive smoking in women. *Tanaffos* **2015**, *14*, 107–114. [[PubMed](#)]
10. Hedin, C.A.; Larsson, A. The ultrastructure of the gingival epithelium in smokers' melanos. *J. Periodontal Res.* **1984**, *19*, 177–190. [[PubMed](#)]
11. Schroeder, H.E. Melanin containing organelles in cells of the human gingiva. *J. Periodontal Res.* **1969**, *4*, 1–18. [[CrossRef](#)] [[PubMed](#)]
12. Hedin, C.A. Smokers' melanos: Occurrence and localization in the attached gingiva. *Arch. Dermatol.* **1977**, *113*, 1533–1538. [[CrossRef](#)] [[PubMed](#)]
13. Hedin, C.A.; Axéll, T. Oral melanin pigmentation in 467 Thai and Malaysian people with special emphasis on smoker's melanos. *J. Oral Pathol. Med.* **1991**, *20*, 8–12. [[CrossRef](#)] [[PubMed](#)]
14. Hedin, C.A.; Pindborg, J.J.; Axéll, T. Disappearance of smoker's melanos after reducing smoking. *J. Oral Pathol. Med.* **1993**, *22*, 228–230. [[CrossRef](#)] [[PubMed](#)]
15. Ho, D.K.; Ghinea, R.; Herrera, L.J.; Angelov, N.; Paravina, R.D. Color range and color distribution of healthy human gingiva: A prospective clinical study. *Sci. Rep.* **2015**, *5*. [[CrossRef](#)] [[PubMed](#)]
16. Wang, J.; Lin, J.; Seliger, A.; Gil, M.; da Silva, J.D.; Ishikawa-Nagai, S. Color effects of gingiva on cervical regions of all-ceramic crowns. *J. Esthet. Restor. Dent.* **2013**, *25*, 254–262. [[CrossRef](#)] [[PubMed](#)]
17. Ono, T.; Naito, T.; Makino, M.; Sato, M. A color analysis of smoker's melanos using a non-contact type dental spectrophotometer. *J. Oral Hyg. Health* **2015**, *2*, 160–164.
18. Kato, T.; Takiuchi, H.; Sugiyama, S.; Makino, M.; Noguchi, S.; Katayama-Ono, T.; Hanioka, T.; Naito, T. Measurement of reduced gingival melanos after smoking cessation: A novel analysis of gingival pigmentation using clinical oral photographs. *Int. J. Environ. Res. Public Health* **2016**, *13*, 598. [[CrossRef](#)] [[PubMed](#)]

19. Gilhar, A.; Pillar, T.; David, M.; Eidelman, S. Melanocytes and Langerhans cells in aged versus young skin before and after transplantation onto nude mice. *J. Investig. Dermatol.* **1991**, *96*, 210–214. [[CrossRef](#)] [[PubMed](#)]
20. Montagna, W.; Carlisle, K. Structural changes in aging human skin. *J. Investig. Dermatol.* **1979**, *73*, 47–53. [[CrossRef](#)] [[PubMed](#)]
21. Haddad, M.M.; Xu, W.; Medrano, E.E. Aging in epidermal melanocytes: Cell cycle genes and melanins. *J. Investig. Dermatol. Symp. Proc.* **1998**, *3*, 36–40. [[PubMed](#)]
22. Fry, L.; Almeyda, J.R. The incidence of buccal pigmentation in caucasoids and negroids in Britain. *Br. J. Dermatol.* **1968**, *80*, 244–247. [[CrossRef](#)] [[PubMed](#)]
23. Monash, S. Normal pigmentation of the oral mucosa. *Arch. Dermatol.* **1932**, *26*, 139–147.
24. Ando, Y.; Suetaka, T.; Sakuma, I. A statistical investigation of gingival pigmentation. *Dent. Abstr.* **1956**, *1*, 749–750.
25. Axell, T.; Hedin, A. Epidemiologic study of excessive oral melanin pigmentation with special reference to the influence of tobacco habits. *Scand. J. Dent. Res.* **1982**, *90*, 434–442. [[PubMed](#)]
26. Marakoğlu, K.; Gürsoy, U.K.; Toker, H.C.; Demirer, S.; Sezer, R.E.; Marakoğlu, I. Smoking status and smoke-related gingival melanin pigmentation in army recruitments. *Mil. Med.* **2007**, *172*, 110–113. [[CrossRef](#)] [[PubMed](#)]
27. WHO Report on the Global Tobacco Epidemic 2015. Available online: http://www.who.int/tobacco/global_report/2015/en/ (accessed on 6 March 2017).
28. Chen, Z.; Peto, R.; Zhou, M.; Iona, A.; Smith, M.; Yang, L.; Guo, Y.; Chen, Y.; Bian, Z.; Lancaster, G.; et al. Contrasting male and female trends in tobacco-attributed mortality in China: Evidence from successive nationwide prospective cohort studies. *Lancet* **2015**, *386*, 1447–1456. [[CrossRef](#)]
29. Araki, S.; Murata, K.; Ushio, K.; Sakai, R. Dose-response relationship between tobacco consumption and melanin pigmentation in the attached gingiva. *Arch. Environ. Health* **1983**, *38*, 375–378. [[CrossRef](#)] [[PubMed](#)]



© 2017 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<http://creativecommons.org/licenses/by/4.0/>).