Comparative analysis of microorganism adhesion on coated and uncoated orthodontic archwires: A prospective clinical study

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ABSTRACT

Objectives-Microorganism adhesion on coated (SS and NiTi) and uncoated (SS and NiTi) orthodontic archwires after clinical use. correlation between surface roughness (SR) and bacterial colonization was evaluated.

Methods-A total of 40 archwire segments (0.016 * 0.022-in) were equally divided into 4 groups: nickel-titanium coated, nickel-titanium uncoated, uncoated stainless steel, and coated SS. The archwires were randomly inserted. After 4 weeks of clinical use, the total number of microorganisms adhering to the archwire was quantified and transformed into colony-forming units. SR was evaluated using a SEM.

Result- All archwires presented microbial adhesion, but coated SS group demonstrating the lowest value ($P \leq 0.001$) . A statistically significant increase in SR was observed after clinical use for all groups ($P \leq 0.05$). Microbial adhesion occurred on all of the archwires tested, especially on the uncoated NiTi shows maximum adhesion.

Conclusion - Adequate implementation of each type of coated archwire can improve patients' comfort as well as reducing the treatment time. The personal orthodontist should always know and understand the needs and opportunities during each stage of treatment. It needs to use the desired features of a specific coated archwire type which has been chosen to satisfy the requirements of the current clinical situation. This would allow for the most optimal and efficient treatment results.

Keywords- Archwire, surface roughness, microbial adhesion, SEM

INTRODUCTION

Displacement of the teeth might have major impact on good oral and dental health, as well as on a person's appearance, contributing to an attractive smile, which can improve psychological self-esteemed self-confidence [1]. Orthodontic treatment is one of the most efficient path to revise the position of the teeth[2].

Arch wires are the primary means of generating forces for orthodontic treatment. They bring about various tooth movements through the medium of brackets and buccal tubes. The major role is that the wire act as springs in clinical orthodontics. Mechanical properties of a materialare determined by several factors such as variation of intrinsic properties alters the nature of the alloy itself, extrinsic properties are macroscopic features (diameter, length) and can be determined by the clinician. Three major properties of archwires are critical in defining their clinical usefulness: strength, stiffness and range.[3]

In turn,the aesthetic color of the archwires can be achieved by coating them with teflon, rhodium, epoxy resin, as well as the combination of silver and biopolymers and 24K gold [4]. The reasons why patients decide for aesthetic archwires are varied. Most patients choose them to make archwires less visible and obtain color similar to the tooth enamel.

During treatment with fixed braces, it is difficult to maintain oral hygiene. The bracket with the inserted archwire provides a medium for adhesion of dental plaque. Increased bacterial titer may be the cause of complications in orthodontic treatment, such as enamel

demineralization or carious lesions [5]. These complications may have a negative impact on aesthetics and cause significant periodontal complications. Bacteria can also affect the characteristics of orthodontic materials.

Attempts to reduce the risk of caries by covering the archwires with various coatings have been carried out for years[6].

Currently, studies are being conducted on different coating materials for archwires and brackets designed to reduce the growth of bacteria. The study by Zakaria et al.[7] reveals that the coating of archwires with titanium dioxide (TiO2) has strong antimicrobial and anticorrosive effects.

Some authors have suggested that the increase in surface roughness of archwires, their corrosion, and the debris occurring during their clinical use, could facilitate the adhesion of microorganisms.[8] However, there remains a scarcity of clinical evidence to confirm this correlation.[9,10]

The adhesion of microorganisms to orthodontic devices may have an influence on enamel demineralization[11] and the development of periodontal diseases,[12] in addition to the possibility of interfering in orthodontic mechanics.[13,14,15] Therefore, studies that evaluate the microbiologic aspects of orthodontic materials should be encouraged.

Thus, this study is aimed to compare the total number of microorganisms adhering to coated and uncoated orthodontic archwires after 4 weeks of clinical use. In addition, the correlation between SR and bacterial colonization was evaluated.

METHOD

1. EXPERIMENT DESIGN

The present study to consisted of 20 randomly selected patients undergoing orthodontic treatment from which microbiological screening was first undertaken. It was done to assess biofilm formation on 4 types of archwire material and microbial colonization on their surface. Four different archwire materials were used, with 10 wires in each group. Samples were collected at Dependent Of Orthodontics and DentofacialOrthopaedics , Jaipur Dental College And Hospital, Jaipur , Rajasthan, India.

EXPERIMENTAL POPULATION

This prospective study was approved by the University Ethics Committee , Jaipur Dental College And Hospital, Jaipur, Rajasthan , on 18/8/2021. The study was carried out between June 2022 to Dec 2022. The study was conducted with twenty volunteers , and Subjects aged between 17 to 25 years who visited the outpatient department of Orthodontics – Jaipur Dental College and Hospital, Jaipur, Rajasthan , India with complaints of malocclusion.

The inclusion criteria included patients with good oral hygiene and with no signs of redness edema or bleeding during brushing , with full complement of teeth erupted in the oral cavity, and zero decay / missing / filled teeth index (DMFT) were also included. Participants should not have been under antibiotics or antiseptic mouthwash for four weeks prior to the sample collection.

Exclusion criteria consisted of the presence of extensive dental caries, other oral disease, having any systemic diseases or undergoing antibiotic therapy and those with any preexisting periodontal conditions.

After explanationsof planned procedures and the study risk / benefits, the queries of participants were answered, and a written informed consent for participation in the study was obtained. After prophylaxis with a rubber cup and pumice stone, the participants had stainless steel brackets McLaughlin,Bennett, Trevisi prescription 3M Unitek Gemini, 0.022 slot brackets were bonded in both maxillary and mandibular arches in all cases.

Four types of rectangular orthodontic archwires that were available on the market at the time of the present study were selected (total number of archwire 10 in each group)
1. Coated nickel-titanium
2. Non coatedNiTi
3. Non coated stainless steel
4. Coated stainless steel

All archwireswere sterilized in an autoclave before use. Oral prophylaxis was performed just prior to archwire insertion. After the archwireswere inserted, the participants individually received basic instructions on oral hygiene and care regarding the orthodontic appliance. All the participants received the same commercial brand of orthodontic toothbrush and fluoridated toothpaste The participants were also instructed not to performmouth washing with any oral antiseptic during the period of the experiment to avoid changes in the oral microbiota. Before removal of the archwires , the participants were asked not to perform oral brushing for a minimum of 12 hours before the appointment.

SAMPLE COLLECTION METHOD

After 4 weeks of clinical use, the orthodontic archwire segments were carefully removed using sterile Mathieu tweezers carefully to avoid iatrogenic biofilm dislodgement. For samples of all types, a 30 mm piece was cut with a distal-end cutter from the end of 'as retrieved' archwires from right side of the posterior segment of both the arches.

Each piece of archwire was dipped in 10 mL of BHI (brain heart infusion) broth meant to be used as a microbial environment and transportation medium. It was placed in a sterilized culture tube. On the outer surface of each tube, the name of type of archwire were recorded. The adherent bacteria were then detached using vortex mixture.

For further analysis the same was immediately transferred to the Central Research Laboratory, Jaipur Dental College for incubation at 37°C for 24 hours.

MEASUREMENT OF MICROBIAL GROWTH

The measurement of oral microbes was attempted in the present study by the Colonies counting method.

In this method we Measure Bacterial Growth that can be achieved by counting bacterial colonies. Bacterial colonies are grown and developed on Solid media. Hence, colonies count method can be achieved by plating techniques. For such sample, we first perform dilution methods. To dilute the bacterial number, the sample is serially diluted 10 times and it is plated in appropriate medium. There are two different plating methods –

- Spread Plate
- Pour plate

Before plating, we need to dilute the sample by serial dilution

SERIAL DILUTIONS

A step-wise dilution of a substance in solution is called as serial dilution. In serial dilution the sample is to dilute the sample to spatially separate the cells in the liquid suspension. Usually the dilution factor at each step is constant, which results in a geometric progression of the concentration in a logarithmic fashion. In highly-diluted solutions, serial dilutions are used to accurately quantify microorganisms in viz. 1:10, 1:100, 1:1000, 1:10000, 1:100000, etc. sufficient enough to count on a Petri plate. To carry out serial dilution, we need 6 test tubes and labelled as 10-1 to 10-6, each containing 9ml of sterile distilled water. 1 ml of overnight grown sample is added in the first tube i.e.10-1 and mixes the content uniformly. From first test tube now take 1 ml of diluted sample add to second test tube. The same procedure is followed till the last test tube 10-6.

SPREAD PLATE TECHNIQUE

For the growth of the microbe of interest, about 0.1 ml of the serially diluted samples was transferred to a Petri dish with blood agar media. The solution was spread uniformly through the use of sterile bent - glass rods. A sterilized glass rod was used to spread the microbe-containing liquid on the plate uniformly. 0.1ml of diluted sample from 10-6 test tube is inoculated in the appropriate nutrient medium and is spread on plates using L shaped spreader. A sterilised glass rod was used to spread the microbe-containing liquid on the plate uniformly.0.1ml of diluted sample from 10-6 test tube is inoculated in the appropriate nutrient medium and is spread on plates using L shaped spreader.

The viable or live bacteria multiply and form a colony are called as Colony forming unit (CFU) and hence the number of cells are determined by number of CFU/ml the original sample. Such viable cells with ability to form colony Formula–

CFU/ml = (no. of colonies x dilution factor) / volume of culture plate

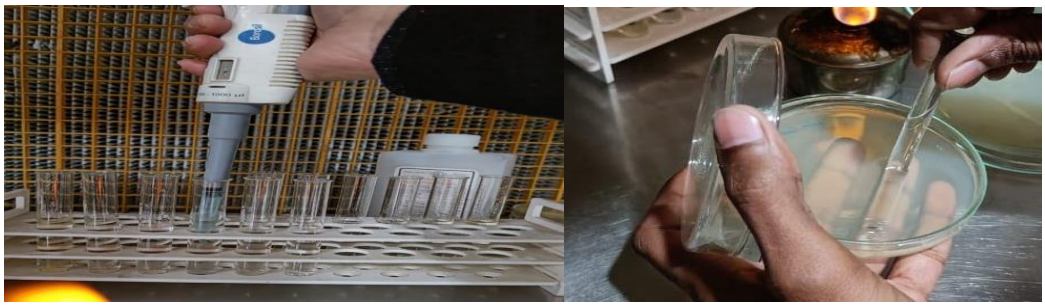

Figure 1: Serial dilution of bacterial suspension after incubation.
Figure 2: Spread plate technique

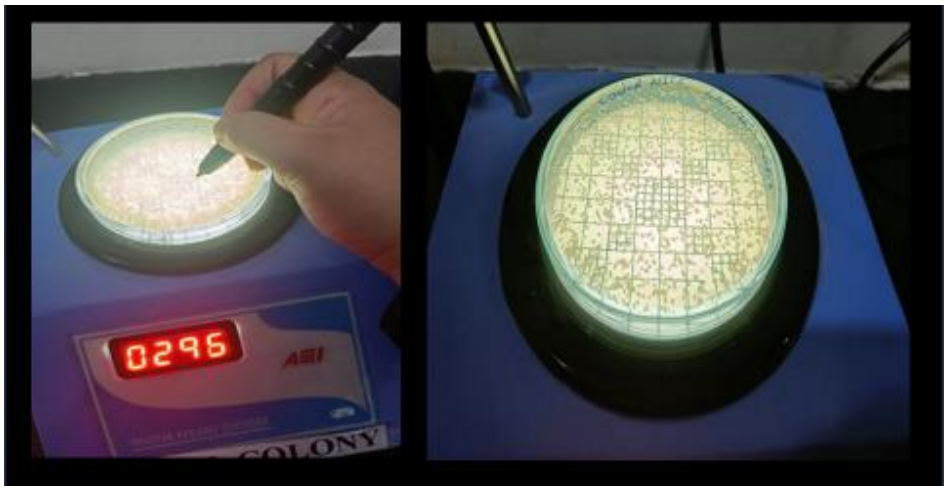

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Fig 3: After incubation of spread plate to evaluate microbial count through digital colony counter

The surface roughness of the orthodontic archwires was measured by SEM (scanning electron microscope) before they were inserted in the oral cavity and after their removal using a profilometer (from Material research centre MNIT JAIPUR).

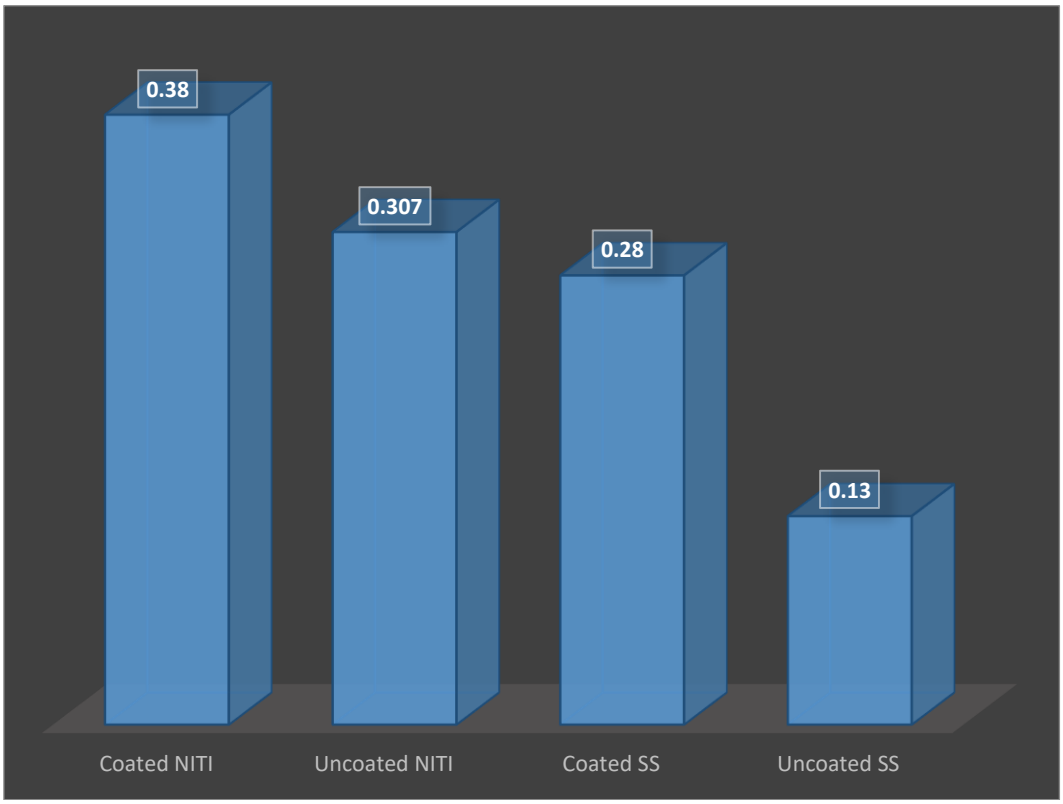
STATISTICAL ANALYSIS AND RESULTS

To evaluate the reproducibility of the roughness measurements, 40 archwire segments were remeasured after 4 weeks.

The data was coded and entered into Microsoft Excel spreadsheet. Analysis was done using SPSS version 20 (IBM SPSS Statistics Inc., Chicago, Illinois, USA) Windows software program. Descriptive statistics included computation of percentages, means and standard

deviations. The data were checked for normality before statistical analysis using Kolmogorov Simonov test. The Mann Whitney u test (for quantitative data to compare two independent observations) and ANOVA test (for quantitative data to compare two and more than two observations) with post hoc Dunnett's test were applied.Level of significance was set at P≤0.05.

SURFACE ROUGHNESS



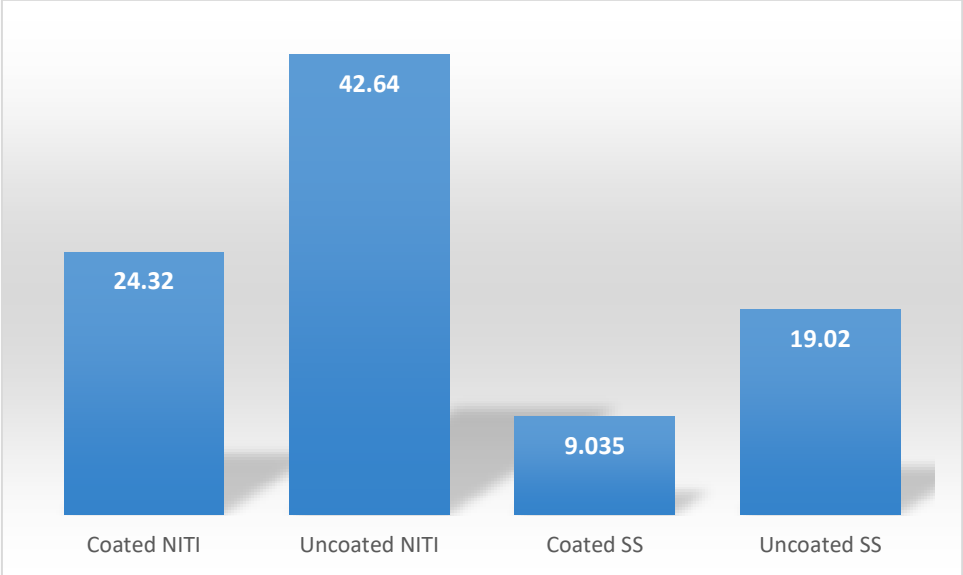
Intra group comparison of Surface Roughness

		Mean Difference	P value	95% Confidence Interval	
				Lower Bound	Upper Bound
Coated NITI	Uncoated NITI	.075	0.001 (S)	.0640	.0860
	Coated SS	.094	0.001 (S)	.0830	.1050
	Uncoated SS	.247	0.001 (S)	.2360	.2580
Uncoated NITI	Coated NITI	-.075	0.001 (S)	-.0860	-.0640
	Coated SS	.019	0.001 (S)	.0080	.0300
	Uncoated SS	.172	0.001 (S)	.1610	.1830
Coated SS	Coated NITI	-.094	0.001 (S)	-.1050	-.0830
	Uncoated NITI	-.019	0.001 (S)	-.0300	-.0080
	Uncoated SS	.153	0.001 (S)	.1420	.1640
Uncoated SS	Coated NITI	-.247	0.001 (S)	-.2580	-.2360
	Uncoated NITI	-.172	0.001 (S)	-.1830	-.1610
	Coated SS	-.153	0.001 (S)	-.1640	-.1420

Dunnett's test

Intra group comparison of Surface Roughness was showed statistically significant results.

MICROBIAL ADHESION



Intra group comparison of Microbial Growth

		Mean Difference	P value	95% Confidence Interval	
				Lower Bound	Upper Bound
Coated NITI	Uncoated NITI	-18.32	0.001 (S)	-18.5277	-18.1123
	Coated SS	15.285	0.001 (S)	15.0773	15.4927
	Uncoated SS	5.294	0.001 (S)	5.0863	5.5017
Uncoated NITI	Coated NITI	18.32	0.001 (S)	18.1123	18.5277
	Coated SS	33.605	0.001 (S)	33.3973	33.8127
	Uncoated SS	23.614	0.001 (S)	23.4063	23.8217
Coated SS	Coated NITI	-15.285	0.001 (S)	-15.4927	-15.0773
	Uncoated NITI	-33.605	0.001 (S)	-33.8127	-33.3973
	Uncoated SS	-9.991	0.001 (S)	-10.1987	-9.7833
Uncoated SS	Coated NITI	-5.294	0.001 (S)	-5.5017	-5.0863
	Uncoated NITI	-23.614	0.001 (S)	-23.8217	-23.4063
	Coated SS	9.991	0.001 (S)	9.7833	10.1987

Dunnett's test

Intra group comparison of Microbial adhesion showed statistically significant results.

DISCUSSION

1. Bacterial accumulation on orthodontic devices plays an important role in the pathogenesis of enamel demineralization during orthodontic treatment.(16)

Orthodontic archwires are manufactured from different metal alloys, present different mechanical properties and SR values, and may also be coated with specific materials to improve their esthetic appearance during treatment. The scarcity of controlled clinical studies that compare the number of microorganisms adhering to different types of orthodontic archwires exposed to the oral cavity justifies the relevance of the present study.

Archwires of the same cross-section (0.016 * 0.022 inch) were used to standardize the size and shape, allowing greater precision in the microbiologic evaluation. Moreover, this study was designed to enable all the archwires to be simultaneously exposed to the same factors, thereby eliminating biases such as differences in pH, food, cleaning, temperature, and bacterial flora, with each patient serving as his or her own control as well (17)

The detection of total microorganisms was positive on all archwires tested, corroborating the findings of previous studies.(18)

Our results showed a higher value in uncoated NiTi and lower value in coated SS of microorganism count in group 2. This finding may suggest the use of coated SS specially in patients with greater cariogenic potential or periodontal problem .

A statistically significant increase in SR was observed on all archwires tested , which is consistent with the results of previous studies evaluating the intra-oral aging of nickel-titanium,(19) stainless steel, and esthetic archwires.(20) The increase in SR was probably related to abrasion during tooth brushing, eating, and interactions between the archwires , brackets, and ligatures; this may be more evident in the groups with coated wires.

In our finding in table no 1 results shows that surface roughness is higher in non coatedNiTi and least surface roughness on non coated SS.

2. The color stability of coated archwires during orthodontic treatment is also clinically important. In the current study, the color stability of these archwirescould be reliably evaluated.

Ideally, the color of esthetic archwires should match that of natural teeth and esthetic brackets. However, the colors of natural teeth vary according to the color measurement protocols used and also by race, gender, and age.(21,22)

Generally, values in the range of one unit are considered exact color matches because they cannot be identified by independent observers.(23) Since instrumental measurements eliminate the subjective interpretation of visual color comparison, spectrophotometers are used instead of visual evaluation.

Many authors(24,25,26,27) have used DE* values to evaluate the ‘‘perceptibility’’ of color differences. However, it is noteworthy that the criteria for perceptibility adopted by each author were different. To counter such differences and disagreements in the criteria used, the NBS rating system is frequently used to determine the degree of color difference, since it offers absolute criteria by which DE* values can be converted to definitions with clinical significance.(28)

Some studies(29) concluded that coffee was the most chromogenic agent in comparison with other staining substances, such as tea and cola drinks. For this reason, a coffee solution was used in this study to evaluate the effect of staining.

3. Dayanne L D Silvaa et al suggested in his study that the color changes of Optis (composite) and Tecnident (nickel-titanium) archwires intensified with a longer immersion period, while the Orthometric (nickel-titanium), Aesthetic Shiny Bright (stainless steel), and Trianeiro (nickel-titanium) wires showed a significant color change between 7 and 14 days and were stable thereafter . When NBS values were evaluated after the 3-week immersion period, ‘‘extremely marked change’’ was observed in Optisarchwires and ‘‘perceivable change’’ was seen in the Ortho Organizers (chromium-nickel) and Trianeiroarchwires.

Arthur et al.(30) suggested that changes in the optical properties within a polymer could be responsible for the color changes seen clinically. They stated that chemical discoloration was caused by the oxidation of unreacted double bonds in the matrix of the polymer and the subsequent formation of degradation products from water diffusion or the oxidation of the polymer. This could explain the staining behavior of coated archwires over time.

Various types of coating are currently being investigated to improve the properties of materials used in medicine and dentistry.

SUMMARY AND CONCLUSION

This study aimed to compare the microorganism adhesion on coated (SS and NiTi) and uncoated (SS and NiTi) orthodontic archwires after clinical use. The correlation between

surface roughness (SR) and bacterial colonization was also evaluated. A total of 40 archwire segments (0.016 * 0.022-in) were equally divided into 4 groups: nickel-titanium coated, nickel-titanium uncoated, uncoated stainless steel, and coated SS. The archwireswere randomly inserted. After 4 weeks of clinical use, the total number of microorganisms adhering to the archwire was quantified and transformed into colony-forming units . SR was evaluated using a SEM. The data were checked for normality before statistical analysis using Kolmogorov Simonov test. The Mann Whitney u test and ANOVA test with post hoc Dunnett's test were applied. Level of significance was set at $P \leq 0.05$.

- All the archwires presented microorganism adhesion, with the coated SS group demonstrating the lowest value ($P \leq 0.001$) . A statistically significant increase in SR was observed after clinical use for all groups ($P \leq 0.05$. Microorganism adhesion occurred on all of the archwires tested, especially on the uncoated NiTi shows maximum adhesion.

So from the present study it can be concluded that:

1. the affinity for biofilm adhesion differs with different types of wires. Biofilm adhesion could be the significant determining factor in the selection of an orthodontic archwire for patients primarily at risk of dental caries or periodontal diseases, or both.
2. Adequate implementation of each type of coated archwire can improve patients' comfort as well as reducing the treatment time. The personal orthodontist should always know and understand the needs and opportunities during each stage of treatment. It needs to use the desired features of a specific coated archwire type which has been chosen to satisfy the requirements of the current clinical situation. This would allow for the most optimal and efficient treatment results.

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