THESIS APPROVED BY

4/13/17

Date

Stephen Gross, Ph. D., Chair

Mark Latta, D.M.D., M.S., Dean

Shawn Powell, D.D.S, M.S.

Barbara O'Kane, Ph. D.

Gail M. Jensen, Ph. D., Dean

FLEXURAL STRENGTH OF ORTHODONTIC CEMENT CONTAINING MICROCAPSULES

By

ERIC ADLER

A THESIS

Submitted to the faculty of the Graduate School of Creighton University in partial fulfillment of the requirements for the degree of Masters of Science in the Department of Oral Biology

---

Omaha, NE

April 13, 2017

ABSTRACT

Caries on the mineralized tooth structure and interface of dental materials continues to be a significant problem in oral health care. The objective of this study was to demonstrate the effect of microcapsules on the flexural strength of orthodontic resin cement formulations. Microcapsules demonstrate a slow, controlled release of fluoride, calcium, and phosphate ions. Orthodontic resin cements that contained microcapsules were incorporated into formulations at 5 w/w% [2 w/w% 0.8 M NaF, 2 w/w% 5.0 M $Ca(NO_3)_2$, and 1 w/w% 3.0 M K_2HPO_4] with one series exception of 6-10 w/w% microcapsules. In this study TEGDMA was the diluent monomer while Bis-GMA was the toughening monomer. The ratio of “toughening monomer:diluent monomer” was explored over a range of 1:1 to 4:1. Barium boroaluminosilicate glass was loaded over a range of 45-75 w/w%, fumed silica was loaded over a range of 0-5 w/w%. The potential number of formulations analyzing these variables is substantial. Therefore, a range of formulations was executed to further investigate the individual variables. These four variables were continuously altered in order to gain better understanding of the role these ingredients had in the presence of microcapsules on flexural strength. The flexural strength was examined as a function of each individual variable. Further analysis examined whether specific variable combinations lead to ideal flexural strength.

ACKNOWLEDGEMENTS

I would like to thank everyone who has spent time and energy into getting this thesis completed. Especially to my advisor and teacher Dr. Stephen Gross who graciously spent countless hours teaching the mechanics of microcapsules while also revising my writing. I would like to thank Dr. Mark Latta for his experience in research to drive this project forward. I would also like to thank fellow students Jacob Gubrud, Grant MacKinnon, and Keyro Bottros who all collaborated in completing the data and brought out the best in myself. Last but not least I'm grateful to Creighton University School of Dentistry's faculty and students, specifically to Dr. Neil Norton, for accepting my application into this esteemed program.

TABLE OF CONTENTS

FIGURES AND TABLES

Section 1

Introduction

1.1 Oral Environment

The environment of the oral cavity is unique and complex. A significant number of microbial species thrive in this moist, warm environment making the barrier of the external and internal environment crucial. The oral mucous membrane serves as that barrier to withstand changing pH and diet [1]. The oral cavity is home to over 700 bacterial species living in symbiosis with their human host. While some bacteria are harmful such as *Streptococcus mutans* and *Lactobacillus*, many can be advantageous in preventing disease. Local factors influence the oral environment including dental plaque, tartar, teeth alignment, bite occlusion, and incompatible artificial prosthesis. Also, systemic and lifestyle factors such as smoking, alcohol consumption, diet, hygiene, medications, obesity, stress, hormones, and genetics play roles [2]. The microbiome evolves from childhood into adulthood. As the child maturates the oral microbe loads increase while the microbe diversity decrease. Hence, the beginning microbe communities play an important role in development [3].

1.2 Tooth Enamel Remineralization/Demineralization

The enamel matrix contains the highest percentage of minerals of the body (96%) with hydroxyapatite being the predominant inorganic mineral. Hydroxyapatite crystalline structure consists of calcium, phosphate, and hydroxide ions [4,5]. The enamel surface attracts salivary glycoproteins and bacterial products creating a thin pellicle layer. This thin protein layer forms on the enamel surface within minutes after consuming carbohydrates protecting the tooth from acids [6]. Bacterial plaque biofilm is a sticky colorless deposit that grows on and around the tooth surfaces. As time progresses the plaque turns into a hardened calculus, brown or pale yellow in color [7]. While plaque is a normal process, it can turn into dental caries. Through the fermentation of carbohydrates, *S. mutans* releases acids that demineralize the tooth and form a carious lesion [8]. Demineralization is defined as the dissolution of the hydroxyapatite crystals resulting in calcium ions, phosphate ions, and hydroxide ions [9]. The process of attaining dental caries requires four key components: a susceptible host, cariogenic bacteria (e.g. *S. mutans*, *Lactobacillus*), high carbohydrate diet and time [10]. When given enough time, the lesion can break through the enamel into the dentin leaving the tooth without the ability to self-heal or remineralize [8]. To control the spread of caries, the goal is to prevent further demineralization whilst reinforcing the repair process of remineralization. Fluorapatite is mineral found in the enamel matrix that helps prevent tooth decay. It presents itself when exposed to fluoride ions through water fluoridation and fluoride-containing toothpaste [11]. The critical pH is the point below which saliva and plaque cease to be saturated with calcium and phosphate ions, leading hydroxyapatite to dissolve [12]. When the critical pH is below 4.5 for fluoroapatite and 5.5 for hydroxyapatite, dental erosion can occur. The lower critical pH and solubility product (K_{sp}) of fluoroapatite makes the tooth structure more resistant to caries attack. When exposed to the sugars in carbohydrates, the oral environment's pH drops under the critical point for roughly 30 to 45 minutes. Closely tied meals or frequent snacking along with high sugar diets, lowers the pH of the oral cavity leading to demineralization [9,13].

1.3 Orthodontic White Spot Lesions

The apparatus of dental braces does well to align and straighten the teeth for those people that so desire them for cosmetic and structural reasons. With over 4 million people wearing braces and the average American spending roughly $5,000, it continues to be an important area of study. The typical person can wear their braces anywhere from six months to 2.5 years, which leaves time for the area underneath the bracket to become a white decalcified spot when the bracket is removed. These white spot lesions (WSLs) develop due to demineralization and appear white, opaque, and chalky caused by surface and subsurface mineral loss. All teeth have the potential for developing these WSLs but the maxillary anterior teeth are the most common [14]. Having poor oral hygiene in association with brackets, bands, arch wires, and other orthodontic devices complicate conventional measures. This leads to prolonged plaque accumulation and possible WSL development [15]. Another complication is the possibility of defects in the orthodontic adhesives under the bracket that leads to microleakage. Microleakage penetration under the orthodontic brackets may cause problems, such as enamel discoloration, corrosion, and decreased bond strength of the adhesive [16]. With the use of a fixed orthodontic appliance, such as braces, this creates a larger surface area for carious attacks. The natural cleansing action of saliva is negated, leading to more intense and frequent biofilm acidification. This then leads to a more rapid caries progression [17].

1.4 Current Developments

Research has now taken dentistry into the area of prevention. One way in which this is done is with preventative bioactive dental materials. With the goal of preventing local biofilm accumulation and acid demineralization, bioactive glass-ceramics are incorporated into the resin. Bioactive glasses have many applications in the dental field for example: enamel remineralization, implant coatings, and bone graft materials for periodontal repair. These bioactive glasses differ from traditional glass because of their ability to release fluoride, phosphate, and calcium ions into the tooth restoration interface [18-21].

Resin-modified glass ionomer cements (RMGICs) are an example of an approach in which bioactive glasses are incorporated. The RMGIC properties are similar to that of conventional glass ionomer cement (CGIC) but are refined with 10-20% additional resin monomers. Advantages of RMGICs are fluoride release, no initial sensitivity, mechanical and chemical bonding, and a more controlled setting time. The disadvantages include poor mechanical properties and only being able to release fluoride for 1-2 weeks while the bracket could sit on the tooth for 2.5 years [22,23].

A new bioactive component is ion permeable microcapsules, which can be incorporated into the resin composite. These microcapsules have a permeable polyurethane membrane that allows fluoride, calcium, and phosphate ions to be released at the tooth interface. A study carried out by Falbo et al. was able to demonstrate the release of ions from microcapsules embedded in resin based composite as well as rosin based varnish formulations. The ion permeable membrane that encapsulates these salts has the ability to be controlled with a slow, sustained release. The ion release depends on the chemical structure of the membrane, initial concentration of salt in the microcapsule, and type of ion salt. In attempt to develop a bioactive orthodontic cement with improved mechanical properties, the effect of different formulation variables on the flexural strength was studied. This novel approach has the potential to combat the early stages of caries throughout the entirety of orthodontic treatment [24-28].

1.5 Mechanical Property Testing

The mechanical property of flexural strength has a crucial bearing on the long-term success and durability of the orthodontic cement. With the complexities of the oral environment as well as masticatory forces it is of importance to not undermine the material's integrity. Flexural strength is measured in terms of stress, and is defined as the stress in the material just before it yields in a flexure test. It represents the highest stress experienced within the material at its moment of failure in bending [29,30]. Flexural strength was measured in megapascals (MPa), which is the SI unit of pressure used to express internal pressure, stress, and Young's modulus [31]. Tensile strength is defined as a stress measured as force (in newtons) per unit area. Tensile strength is crucial when considering orthodontic resin cement because it measures the maximum stress it can withstand while being stretched before breaking. With the orthodontic resin cement being a brittle material it was subjected to the three-point bend test with a crosshead speed of 1mm/min [32].

Section 2

Materials and Methods

2.1 Prepolymer Synthesis

Microcapsules were prepared with a polyurethane shell via a prepolymer synthesis in an inert environment at 70 °C. A cyclohexanone solvent was used to react ethylene glycol (Fisher, New Jersey) with isophorone diisocyanate (Sigma-Aldrich, Steinheim) and left overnight. The prepolymer was then dried by vacuum [33].

2.2 Microcapsule Synthesis

After drying, the prepolymer was added to an emulsifying agent and methyl benzoate (Acros Organics, New Jersey). Aqueous salt solutions of 3.0 M potassium phosphate dibasic (Fisher Scientific, New Jersey), 5.0 M calcium nitrate tetrahydrate, (Alfa Aesar, Massachusetts) and 0.8 M sodium fluoride (MP Biomedicals, Ohio) were prepared. A reverse emulsion was used to create an environment for an interfaced polymerization of the urethane to encapsulate these salt solutions [33].

A reverse emulsion forms when the prepolymer oil solution is agitated and the aqueous salt solution is introduced. The oil solution was agitated in a custom made reactor at 70 °C while the aqueous salt solution was added slowly. The reaction was then quenched using ethylene glycol to finish preparing the microcapsules. These microcapsules were centrifuged in a Fisher Centrific 288 (Fisher Scientific, New Jersey) centrifuge using a diluent, rinsed, and prepared for formulation into an orthodontic resin cement [33].

2.3 Orthodontic Cement Formulations

The formulations conducted in this study were prepared with the same rheological properties as some of the industry leaders in orthodontic bracket and band cement, but with the addition of microcapsules. Orthodontic resin cements 3M Transbond XT, Ormco

Grēngloo, and Reliance Assure were all tested for flexural strength. The formulations consisted of w/w% loadings of monomer (BISGMA and TEGMA), glass, fumed silica, and microcapsules. The band cements were prepared with loadings of 5 w/w% microcapsules containing a mixture of 2 w/w% 0.8 M sodium fluoride, 2 w/w% 5.0 M calcium nitrate tetrahydrate, and 1 w/w% 3.0 M potassium phosphate dibasic aqueous salt solutions (2/2/1) with all ions needed for remineralization. All bracket cements were prepared with the same 5 w/w% microcapsule loading with the exception of five formulations as seen on Table 1. The formulations in Table 1 have increasing w/w% of sodium fluoride to examine how much microcapsule loading can be accomplished while maintaining an acceptable flexural strength.

% Loading of Microcapsules	Distribution of Microcapsule Type
5	2% 0.8M NaF; 2% 5.0M $Ca(NO_3)_2$; 1% 3.0M K_2HPO_4
6	3% 0.8M NaF; 2% 5.0M $Ca(NO_3)_2$; 1% 3.0M K_2HPO_4
7	4% 0.8M NaF; 2% 5.0M $Ca(NO_3)_2$; 1% 3.0M K_2HPO_4
8	5% 0.8M NaF; 2% 5.0M $Ca(NO_3)_2$; 1% 3.0M K_2HPO_4
9	6% 0.8M NaF; 2% 5.0M $Ca(NO_3)_2$; 1% 3.0M K_2HPO_4
10	7% 0.8M NaF; 2% 5.0M $Ca(NO_3)_2$; 1% 3.0M K_2HPO_4

Table 1: Formulations of bracket cement containing all three microcapsule types

2.4 Mechanical Testing

The orthodontic resin cement was made into twelve rectangular prism shaped specimens, using a Teflon mold with the dimensions specified by the ISO 4049/2000 specification (25 mm x 2 mm x 2 mm). The Teflon mold was positioned over a mylar strip and glass slide while filled with the formulation. The formulation was syringed in a single increment with excess removed using a razor blade from the mold before polymerization. A halogen curing light unit (Optilux 501) was used to polymerize and the light intensity was measured prior to use. The resin composite was cured for 30 seconds in three evenly spaced locations along the rectangular prism shaped specimen and then turned over to repeat on the other side. Once the specimen was removed from the mold, the excess flash material was polished with 600 grit sandpaper. The twelve specimens were stored in an oven for a minimum of 7 days. They were submitted to a three-point bend test with a universal testing machine (MTS Insight Electromechanical – 1 kN standard length) with a crosshead speed of 1 mm/min. The maximum force loads were obtained by using the following formula: $\sigma=3FL/2bd^2$ where the flexural strength (σ) was calculated in megapascals (MPa); F is the maximum load in newtons (N); L is the distance between supports in millimeters (mm); b is the width of the specimen in mm; d is the thickness of the specimen in mm.

Section 3

Results

3.1 Introduction

The incorporation of microcapsules that release remineralizing ions is a new advance in the development of bioactive dental materials. In this study most of the orthodontic resin cement formulations used 5 w/w% total microcapsules [2 w/w% 0.8 M NaF, 2 w/w% 5.0 M $Ca(NO_3)_2$, and 1 w/w% 3.0 M K_2HPO_4]. With the exception being one series that had 6-10 w/w% of varying microcapsules. A critical aspect for microcapsule incorporation into dental orthodontic cement is the understanding of how the microcapsules could potentially impact flexural strength. Orthodontic cements have a varying number of components in a formulation. These components include the ratio of toughening monomer to diluent monomer, the loading of glass, and the loading of fumed silica. In this study Bis-GMA was used as the toughening monomer while TEGDMA was the diluent monomer. The ratio of "toughening monomer:diluent monomer" was examined over a range of 4:1 through 1:1. The barium boroaluminosilicate glass was loaded over a range of 45-75 w/w% while the fumed silica was loaded over a range of 0-5 w/w%. A range of formulations was experimented that explored all of the potential number of variables.

The four components were consistently varied in order to thoroughly understand the role of each individual component in the presence of microcapsules on flexural strength. Although multiple variables change during each formulation, the flexural strength was examined as a function of monomer, glass, fumed silica, and microcapsule loading.

3.2 Bis-GMA:TEGDMA Ratio

The effect of monomer ratio with respect to both Bis-GMA and TEGMA content in each formulation is reported in Table 2. The monomer ratio is reported as four different ratios used in the formulations. They were in ratios of Bis-GMA to TEGDMA in the forms of 50/50; 60/40; 70/30; and 80/20. Multiple variations existed in terms of the percentage of each monomer in the continuous phase, glass, fumed silica, and microcapsule loading. However this data only considers average flexural strength and standard deviation for each monomer ratio as reported in Table 2.

Bis-GMA:TEGDMA Ratio	50/50	60/40	70/30	80/20
Flexural Strength	102.2	95.7	93.3	94.9
Standard Deviation	18.3	17.8	15.8	21.6

Table 2: The average flexural strength for orthodontic resin cements as a function of the Bis-GMA and TEGDMA ratio.

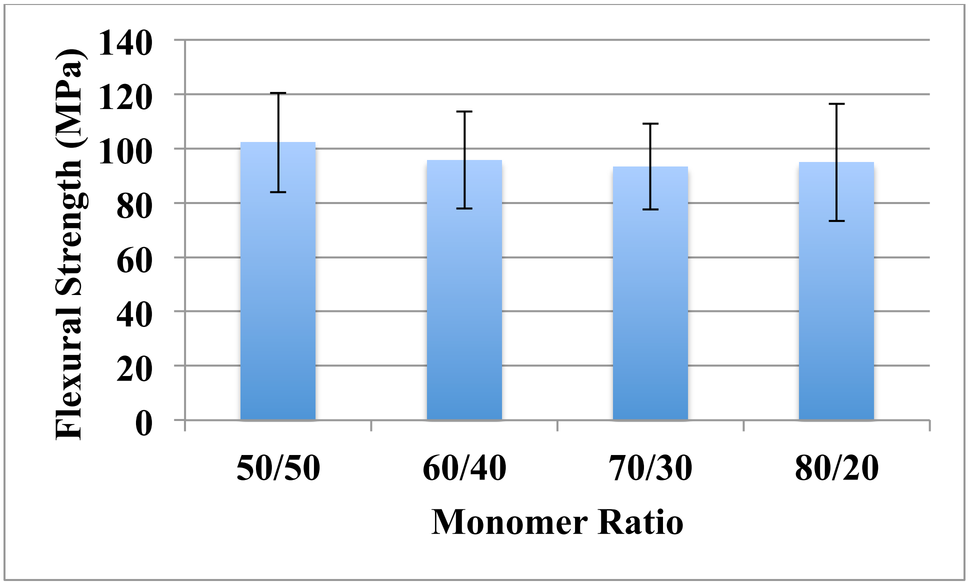

Figure 1. The role of the monomer ratio on the flexural strength was measured. Across all the formulations prepared, the flexural strength of formulations with different monomer ratio were averaged and divided into four categories. This was done to compare the impact that the ratio of Bis-GMA and TEGDMA had on the formulations that incorporated microcapsules.

3.3 Monomer Loading

The effect of monomer loading with respect to all the ratios of Bis-GMA and TEGDMA content in the formulations is reported in Table 3. Multiple formulations existed in terms of how much total monomer was used, the glass, fumed silica, and microcapsule loading. However this data only considers average flexural strength and standard deviation for the monomer loading as reported in Table 3.

% Monomer	Flex Strength	SD
17.4	67.7	15.6
18.4	84.2	15.7
19.4	92.3	25.9
20.4	113.4	23.6
21.4	103.1	15.3
21.4	125.2	25
22.2	104	17.4
22.3	104	18
22.4	105.9	18.9
22.5	92.1	17.6
22.6	104.7	18.9
22.7	108.4	12.5
22.8	123.5	17.7
22.9	110.1	16.6
23	107	15.7
23.2	116.6	14.8
23.4	92.5	12.8
23.6	99.6	15.8
24	99.2	18.6
24.2	107.8	18.1
24.25	94.2	19
24.4	98.9	19
24.5	115.4	20.4
24.8	104.2	19.9
25	99	13.9
25.5	92.8	12.2
26.4	100.7	26.2
30	95.5	23
35	98.1	16.4
40	88.2	30
45	102.2	16

Table 3: The average flexural strength for orthodontic resin cements as a function of the percent monomer loading.

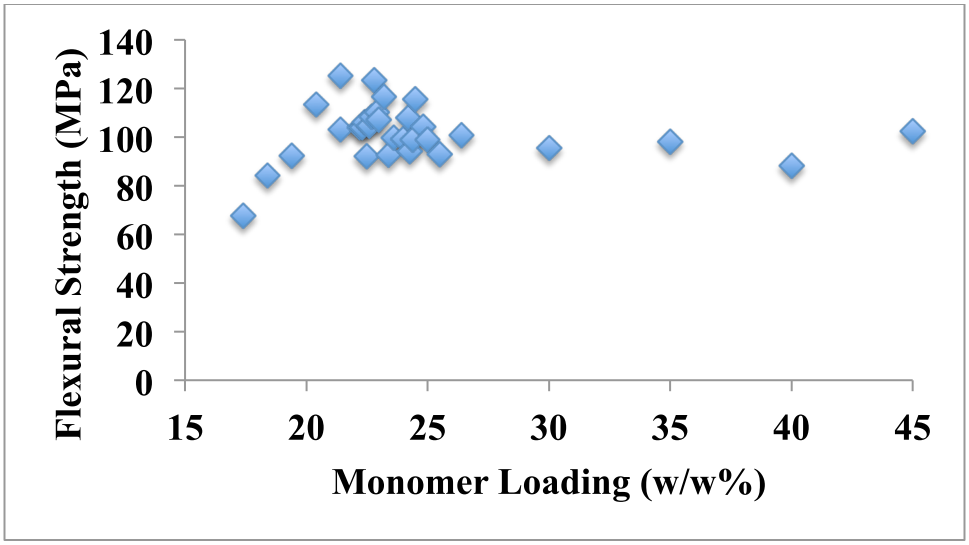

Figure 2. The flexural strength (MPa) was measured as a function of percent monomer loading. Every formulation was taken into account with multiple trials of monomer loading being averaged. This was done to determine if the w/w% of the continuous phase in the formulation effected flexural strength regardless of the Bis-GMA /TEGDMA ratio, glass, fumed silica, and microcapsule loading.

3.4 Glass Loading

The effect of glass loading with respect to all the ratios of Bis-GMA and TEGMA content in the formulations is reported in Table 4. Multiple variations of the continuous phase existed in terms of the monomer ratio, fumed silica, and microcapsule loading. This data only considers average flexural strength and standard deviation for the glass loading as reported in Table 4.

% Glass	Flex Strength	SD
45	102.2	16
50	88.2	30
55	98.1	16.4
60	95.5	23
65	93.5	10.1
66	100.7	26.2
67	92.8	12.2
68	94.2	19
70	101.9	17.9

Table 4: The average flexural strength for orthodontic resin cements as a function of the percent glass loading.

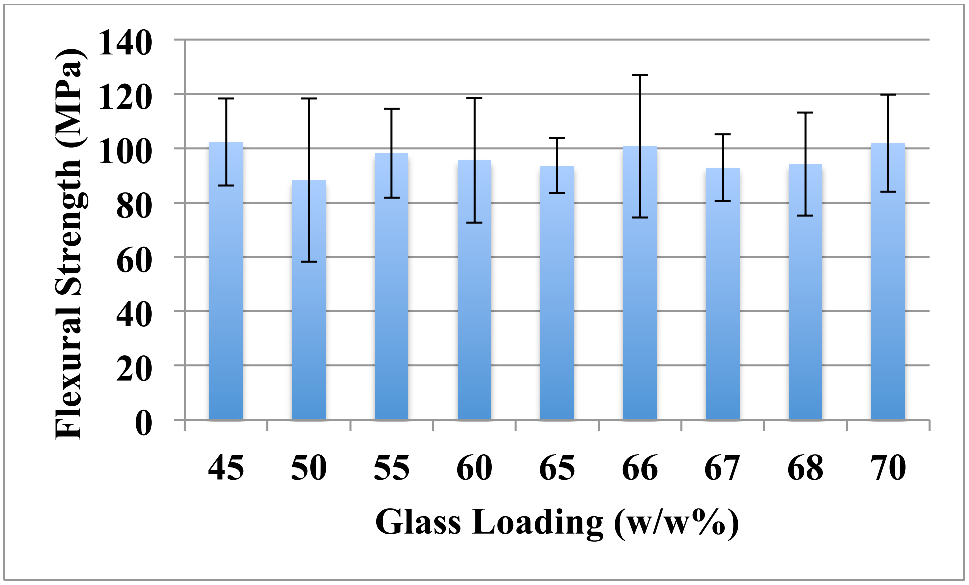

Figure 3. The flexural strength (MPa) was measured as a function of the glass loading. Every formulation was taken into account with multiple trials of identical glass loading.

3.5 Glass Loading with constant Bis-GMA/TEGDMA Ratio

The effect of decreasing glass loading with respect to increasing monomer loading in each formulation is reported in Table 5. While the monomer loading did increase, the monomer ratio of 80/20 Bis-GMA/TEGDMA was held constant in this set of experiments. Fumed silica and microcapsule loadings were also held constant at 5 w/w% for each formulation. This data only considers average flexural strength and standard deviation for each monomer and glass loading percentages as reported in Table 5.

% Monomer	% Glass	Flex Strength	SD
45	45	102.2	16
40	50	88.2	30
35	55	98.1	16.4
30	60	96.1	25

Table 5: The average flexural strength for orthodontic resin cements as a function of the percent monomer and glass loading.

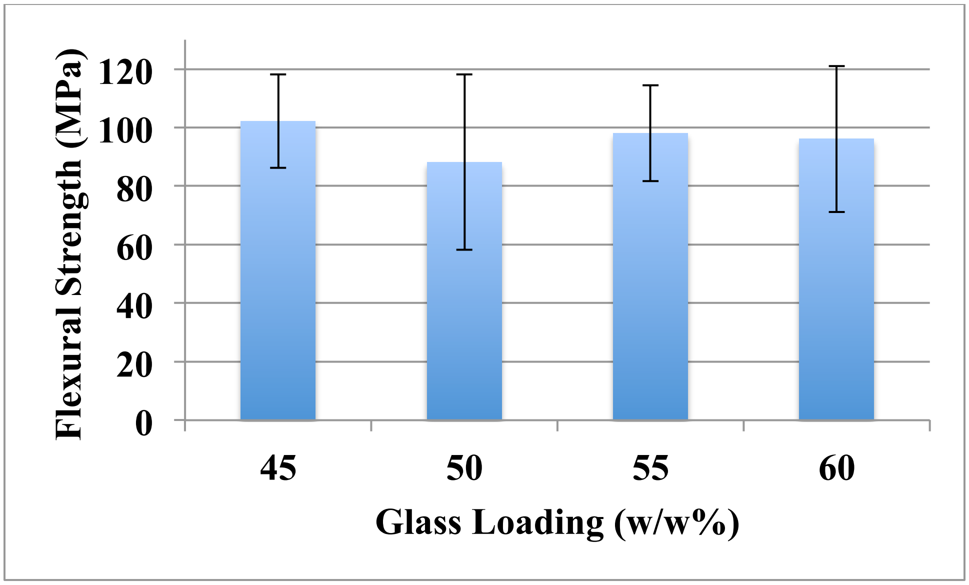

Figure 4. The flexural strength was measured as a function of glass loading with a constant Bis-GMA/TEGDMA ratio.

3.6 Fumed Silica Loading

The effect of increasing fumed silica loading in each formulation is reported in Table 6. While the monomer loading decreased, the monomer ratio of 50/50 Bis-GMA/TEGDMA was held constant. Glass and microcapsule loading remained at 70 w/w% and 5 w/w% respectively, for each formulation. This data only considers average flexural strength and standard deviation for each monomer and fumed silica loading percentages as reported in Table 6.

% Fumed Silica	Flexural Strength	SD
0	104.5	17.7
0.2	104.2	19.9
0.5	115.4	20.4
0.6	98.9	19
0.8	107.8	18.1
1	99.2	18.6
1.4	99.6	15.8
1.6	92.5	12.8
1.8	116.6	14.8
2	107	15.7
2.1	110.1	16.6
2.2	123.5	17.7
2.3	108.4	12.5
2.4	104.7	18.9
2.5	92.1	17.6
2.6	105.9	18.9
2.7	104	18
2.8	104	17.4

Table 6: The average flexural strength for orthodontic resin cements as a function of fumed silica loading.

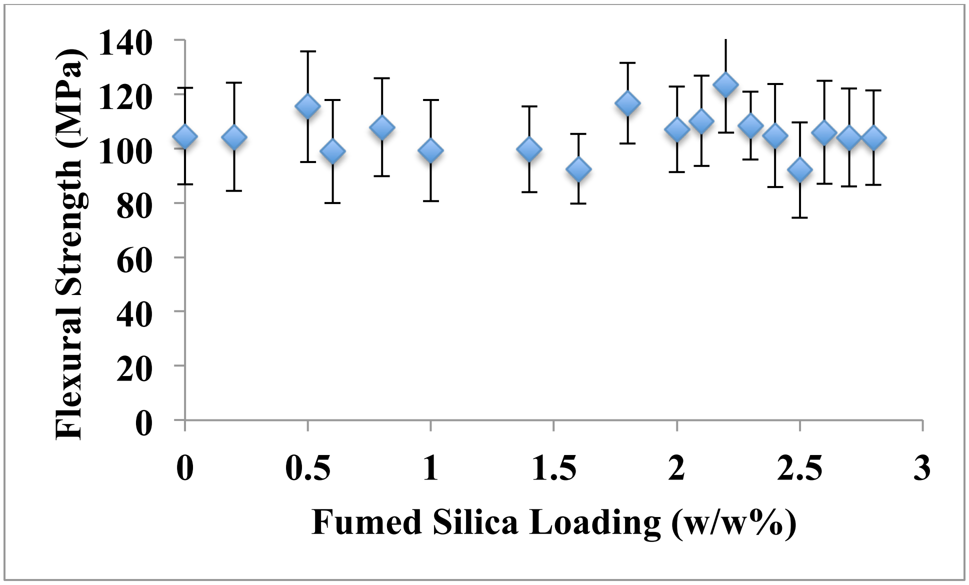

Figure 5. The flexural strength (MPa) was measured as a function of the fumed silica loading. This was done in order to measure the effect of fumed silica loading had on the flexural strength when all other formulation variables were constant.

3.7 Microcapsule Loading

The effect of increasing microcapsule loading in the formulation on the flexural strength is reported in Table 7. While the monomer loading was decreased, the monomer ratio of 50/50 Bis-GMA/TEGDMA was held constant. Glass and fumed silica loading remained at 70 w/w% and 5 w/w% respectively, for each formulation. This data only considers the average flexural strength and microcapsule loading percentages as reported in Table 7.

% Microcapsules	Flexural Strength	SD
6	114.2	20.2
7	113.4	23.6
8	92.3	25.9
9	84.2	15.7
10	67.7	15.6

Table 7: The average flexural strength for orthodontic resin cements as a function of microcapsule loading.

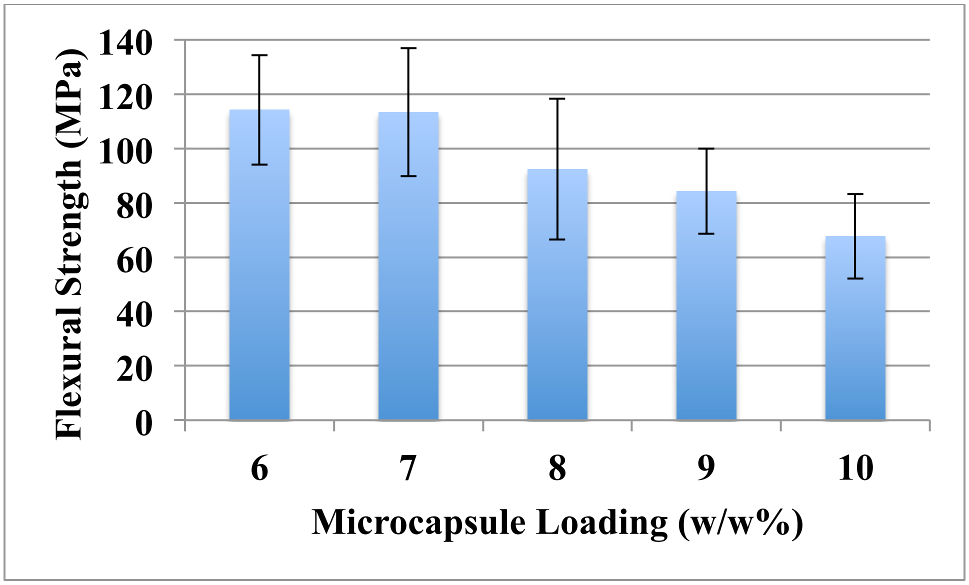

Figure 6. The flexural strength (MPa) as a function of microcapsule loading (w/w%) was measured. This was done in order to compare the impact of microcapsule loading on flexural strength when all other formulation variables were constant.

3.8 Commercial Orthodontic Cements

The flexural strength (MPa) was measured for commercially available orthodontic resin cements. These values are reported in Table 8.

Formulation	Flexural Strength	SD
3M Transbond XT	106.6	19.2
Ormco Grengloo	134.8	14.8
Reliance Assure	115.4	26.1

Table 8: The average flexural strength for commercial orthodontic resin cement formulations.

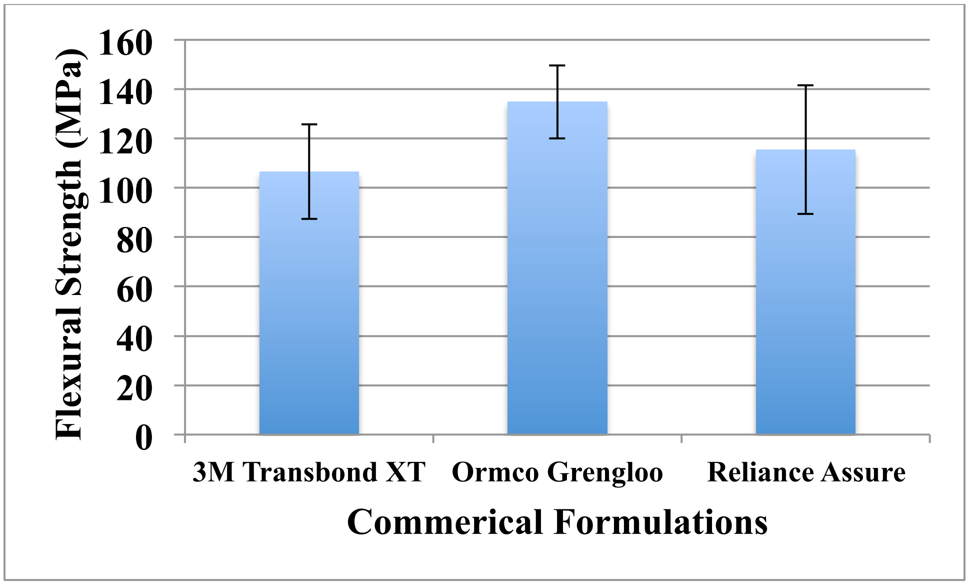

Figure 7. The flexural strength (MPa) as a function of commercial formulations was measured. This was done to compare our formulations to the commercially available orthodontic resin cements on the market.

Section 4

Discussion

The purpose of this study was to develop a new approach to promote remineralization by incorporating ion permeable microcapsules as a filler in orthodontic resin cement. To promote remineralization and prevent demineralization of the enamel in the oral environment, the resin cement would release remineralizing ions (e.g. fluoride, calcium, phosphate) contained within the microcapsules. This formulation could then be used in orthodontic treatment to combat early stages of dental caries, known as white spot lesion development. Furthermore, inclusion of microcapsules in orthodontic resin cements enhanced the uptake and release of additional fluoride ions due to tooth brushing [34]. If this does not adversely impact flexural strength, improvement could be potentially achieved. The variables of Bis-GMA, TEDGMA, glass, fumed silica, and microcapsules all can be manipulated to produce differing effects on the physical properties in an orthodontic resin cement. These variables were prepared in over 65 formulations with the goal of measuring and analyzing the flexural strength. While understood that each independent variable of the continuous phase directly affects one another, the data was systemically analyzed for the effect of each variable.

The first variable considered was the role of the monomer ratio. Across all the formulations prepared, the flexural strength at different monomer ratios were averaged and divided into four categories. This study used Bis-GMA as the toughening monomer and TEGDMA as the diluent monomer. As seen in both Table 2 and Figure 1 the role Bis-GMA/TEGDMA ratio had on flexural strength was observed over the range of 4:1 through 1:1. Multiple variations of in the continuous phase existed with regards to monomer, glass, fumed silica, and microcapsule loading. When flexural strength was measured, a slight trend downward of the mean was observed from 50/50 to 80/20. But this trend was not statistically significant in the context of standard deviations. With the ratio of Bis-GMA/TEGDMA monomers taken into account, the percent monomer loading was the next variable of interest. Again a multitude of variations existed for glass, fumed silica, and microcapsule loading in addition to different monomer ratios. In Table 3 and Figure 2 the role of monomer loading on the flexural strength was examined. In this data table and graph no significant trend was observed.

The next variable that was considered is the effect of barium boroaluminosilicate glass loading. Although the expected result would be higher flexural strength with increased glass content, the trend was examined if that were true in the presence of microcapsules. The selected formulations had different variations in the continuous phase of monomer, fumed silica, and microcapsule loading. The flexural strength and standard deviation for glass loadings of 45-70 w/w% can be seen in Table 4 and Figure 3. Some multiple trials of the same glass loadings were averaged together to produce their flexural strength. When looking through this particular table and graph no discernable trend was observed. To further test this observation, a subset of formulations was prepared where all the variables were held constant. In Table 5 and Figure 4 the flexural strength was measured for formulations where the monomer ratio was constant at 80/20 Bis-GMA/TEGDMA with 5 w/w% fumed silica and 5 w/w% microcapsule loadings. No statistical significance was found when taking these flexural strength results into account.

Fumed silica loading was the next variable considered. In Table 6 and Figure 5 the flexural strength and standard deviation were reported. The percent fumed silica loading range of 0-2.8 w/w% was thought to produce higher flexural strength with each increased loading of fumed silica. Once again in the presence of microcapsules, no distinguishable trend of additive loading on the flexural strength was apparent.

The final variable considered was the microcapsule loading in the formulations. In most all of the formulations 5 w/w% total microcapsules [2 w/w% 0.8 M NaF, 2 w/w% 5.0 M $Ca(NO_3)_2$, and 1 w/w% 2.0 M K_2HPO_4] was used in combination of the continuous phase variations of monomer, glass, and fumed silica loading. Ideally, the fluoride would continue to be released at higher concentrations for longer periods of time. To test the varying degrees of increased microcapsule loading, Table 1 illustrates how the formulations were changed to incorporate the increased percentage of fluoride microcapsules. Both Table 7 and Figure 6 report the effect microcapsule loading (6-10 w/w%) had on flexural strength. The trend of increased microcapsule loading leads to a decrease in the mean value of the flexural strength above 7 w/w% was observed.

These experiments have shown that the incorporation of microcapsules into orthodontic resin cement is a potential way to manage white spot lesions. The polyurethane based semipermeable membrane of the microcapsules act as a filler within the resin cement that is capable of potentially promoting remineralization. This study has demonstrated that there are potential formulations of orthodontic resin cement with microcapsules that can meet the standard for flexural strength of an orthodontic bracket cement.

REFERENCES

1. Dale, B. A., & Fredericks, L. P. (2005). Antimicrobial Peptides in the Oral Environment: Expression and Function in Health and Disease. *Current Issues in Molecular Biology*, *7*(2), 119–133. http://doi.org/10.1093/jac/dki103
2. Kudo, Y., Tada, H., Fujiwara, N., Tada, Y., Tsunematsu, T., Miyake, Y., & Ishimaru, N. (2016). Oral environment and cancer. *Genes and Environment*, *38*, 13. http://doi.org/10.1186/s41021-016-0042-z
3. Sampaio-Maia, B., & Monteiro-Silva, F. (2014). Acquisition and maturation of oral microbiome throughout childhood: An update. *Dental Research Journal*, *11*(3), 291–301.
4. Ross, Michael H., Kaye, G.I. and Pawlina, W. (2006) *Histology: a text and atlas*, 5th ed., Philadelphia; London: Lippincott Williams & Wilkins, ISBN 0-7817-7221-4
5. Fincham AG, Moradian-Oldak J, Simmer JP, "The Structural Biology of the Developing Dental Enamel Matrix" J Struct Biol M 1991; 126:270-299
6. Newman, Michael G. Carranza's Clinical Periodontology, 10th Edition. Saunders Book Company, 072006. 9.4.2.
7. Darby M L, Walsh M M. *Dental Hygiene Theory and Practice*. 2010.
8. Ten Cate J, Featherstone JD. "Mechanistic Aspects of the Interactions Between Fluoride and Dental Enamel" Crit Rev Oral Biol M 1991; 2: 283-296.
9. Barira Islam, Shahper N Khan, Asad U Khan "Dental caries: From infection to prevention" Med Sci Monit 2007; 13(11): RA196-203.
10. Yeolekar TS, Chowdhary NR, Mukunda K, Kiran N. Evaluation of Microleakage and Marginal Ridge Fracture Resistance of Primary Molars Restored with Three Restorative Materials: A Comparative *in vitro* Study. *International Journal of Clinical Pediatric Dentistry*. 2015;8(2):108-113. doi:10.5005/jp-journals-10005-1294.
11. "How does fluoride protect my teeth and make them strong?". *UCSB Science Line*. Regents of the University of California. Retrieved 3 June 2016.
12. Marsh PD and Martin M. Oral Microbiology. 5th ed. Churchill Livingstone Elsevier, London, UK. 2009.
13. A. Wiegand, W. Buchalla, T. Attin, Review on fluoride-releasing restorative materials-fluoride release and uptake characteristics, antibacterial activity and influence on caries formation, Dent. Mater. 23 (2007) 343-362
14. Srivastava K, Tikku T, Khanna R, Sachan K. Risk factors and management of white spot lesions in orthodontics. *Journal of Orthodontic Science*. 2013;2(2):43-49. doi:10.4103/2278-0203.115081.
15. J Orthod Sci. 2013 Apr-Jun; 2(2): 43–49. doi: 10.4103/2278-0203.115081
16. Alkis H, Turkkahraman H, Adanir N. Microleakage under orthodontic brackets bonded with different adhesive systems. *European Journal of Dentistry*. 2015;9(1):117-121. doi:10.4103/1305-7456.149656.
17. St-Pierre L, Bergeron C, Qian F, et al: Effect of polishing direction on the marginal adaptation of composite resin restorations. J Esthet Restor Dent 25:25-38, 2013
18. Lockyer, MWG; Holland D; Dupree R (1995). "NMR investigation of the structure of some bioactive and related glasses". J. Non-Crys. Sol. 188 (3): 207–219. doi:10.1016/0022-3093(95)00188-3

19. N. Zhang, M. D. Weir, C. Chen, M. Melo, Y. Bai, H. K. Xu, Orthodontic cement with protein-repellent and antibacterial properties and the release of calcium and phosphate ions, Journal of Dentistry 50 (2016) 51-59.
20. Ali, Saqib, Imran Farooq, and Kefi Iqbal. "A Review of the Effect of Various Ions on the Properties and the Clinical Applications of Novel Bioactive Glasses in Medicine and Dentistry." *The Saudi Dental Journal* 26.1 (2014): 1–5. *PMC*. Web. 20 Jan. 2016.
21. Davidson MT, Greving TA, McHale WA, Latta MA, Gross SM. "Ion Permeable Microcapsules for the release of biologically available ions for remineralization" J Biomed Res A 2012; 100: 665-672.
22. Summit J. Fundamentals of Operative Dentistry: A Contemporary Approach. 2006 Quintessence Publishing Co.
23. Ewoldsen N, Demke R. A Review of Orthodontic Cements and Adhesives. American Journal of Orthodontics and Dentofacial Orthopdeics. 2001;120:45-48
24. Davidson MT, Greving TA, McHale WA, Latta MA, Gross SM. "Ion Permeable Microcapsules for the release of biologically available ions for remineralization" J Biomed Res A 2012; 100: 665-672.
25.Falbo, M. (2012) The release of calcium, fluoride, and phosphate ions from ion permeable microcapsules in a rosin varnish. Published dissertation. Creighton University.
26. Fulton, J (2013) The Effect of Continuous Phase and Glass Content on Fracture toughness and Calcium and Phosphate Ion Release from Ion Permeable Microcapsules in Glaze Formulations Published dissertation. Creighton University
27. Cooper, R. (2013) Encapsulation of Calcium and Phosphate Ions in a Toothpaste Formulation, Published dissertation. Creighton University
28. Murphy J, Riley JP. Single solution method for the determination of phosphate in natural waters. *Anal Chim Acta* 1962; 27:31-36. (Molybdenum Blue Method Source)
29. J.M. Hodgkinson (2000), *Mechanical Testing of Advanced Fibre Composites*, Cambridge: Woodhead Publishing, Ltd., p. 132–133.
30. William D. Callister, Jr., *Materials Science and Engineering*, Hoken: John Wiley & Sons, Inc., 2003.
31. International Bureau of Weights and Measures (2006), *The International System of Units (SI)* (PDF) (8th ed.), p. 118, ISBN 92-822-2213-6
32. Degarmo, Black & Kohser 2003, p. 31
33. M.T. Davidson, T.A. Greving, W.A. McHale, M.A. Latta, S.M. Gross, The control of phosphate ion release from ion permeable microcapsules formulated into rosin varnish and resin glaze, Dental Mater. 29 (2013) 804-813.
34. B.D. Burbank et. al, Ion release, fluoride charge of and adhesion of an orthodontic cement paste containing microcapsules, Journal of Dentistry. 45 (2016) 32-38.
35. M. Slater, (2014) Fluoride, Calcium, and Phosphate Ion Release From Ion Permeable Microcapsules Incorporated Into A Two-Part Orthodontic Adhesive System, Creighton University.