CHAPTER III

MATERIALS AND METHODS

This *in vitro* study has been done in the laboratory. This investigation was seperated in two parts:

Part I Elastic limit of elastomeric chains

Part II Force decay of elastomeric chains with simulated tooth movement.

MATERIALS

1. Samples

The Ormco® Grey Power Chain Generation II (Ormco, Glendora, California, U.S.A.) were used as samples in this investigation since they are in popular used on our clinics and the manufacture claims them to provide a more uniform continuous delivering force over a longer period of time (Ormco® catalogue, 1998). They were made from synthetic polymer, thermoplastic polyurethane elastomer, but the exact composition was proprietary information. They were manufactured by a die-punching process so that they were rectangular in cross-section. Chains are produced with different unit forms for using in various clinical situation: closed space, open space and wide space forms. The loops of closed space form adjoin to each other continuously without a solid bar, but the loops of open space and wide space forms are separated by 0.5 millimeters solid bars and 1.5 millimeters solid bars, respectively. All forms of elastomeric chains had the same loop dimension, 0.7 millimeters chain mass width, 1.4 millimeters inside diameter (ID) and 0.5 millimeters thickness. The solid bar area had the same chain mass width and thickness as the loop area. The joint area of the closed form was two times of the solid bar width (1.4 millimeters).

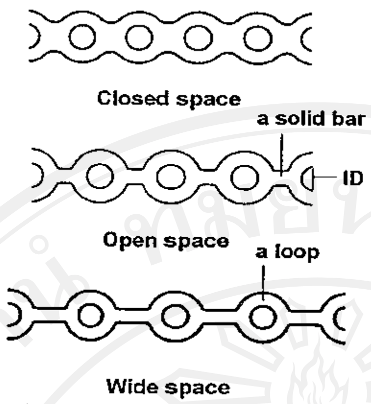


Figure 3.1 Three forms of elastomeric chain, Ormco® Power Chain Generation II

The different module lengths (3 and 4 loops) of each form of Ormco® Grey Power Chain Generation II were used for elastic limit and force decay with simulated tooth movement study. These lengths of elastomeric chain were frequently used in the clinic to retract the canine into an extracted premolar space.

The elastomeric chains were obtained from the local vendor in Thailand. They were remained sealed in their plastic containers throughout the experiment and stored in refrigerator until they were randomly selected for testing. They were left at room condition at least 24 hours prior to test. Each form of orthodontic elastomeric chains was trimmed in three and four loops with an extra half loop at each end to prevent distortion by cutting.

2. Instruments

Part I: Elastic limit of orthodontic elastomeric chains

1. Universal Testing Machine (model 5566, Instron Corporation, England), with 10 kilonewtons load cell and computer connection, was used to obtain force-displacement curves from each group of elastomeric chains and to measure the generated force and displacement at elastic limit. Each upper and lower

crosshead was placed with a designed vertical stainless steel hook, 1.2 millimeters in diameter, for holding the elastomeric chains. The lower crosshead was fixed in place, and the upper crosshead could be moved up vertically at a constant rate of the " crosshead speed " for distracting the elastomeric chains. The output of the transducer attached to the crosshead was fed to an internal X-Y chart recorder that automatically plotted force versus displacement curves which was shown on the monitor. The generated force and displacement were recorded in newton and millimeter, respectively.

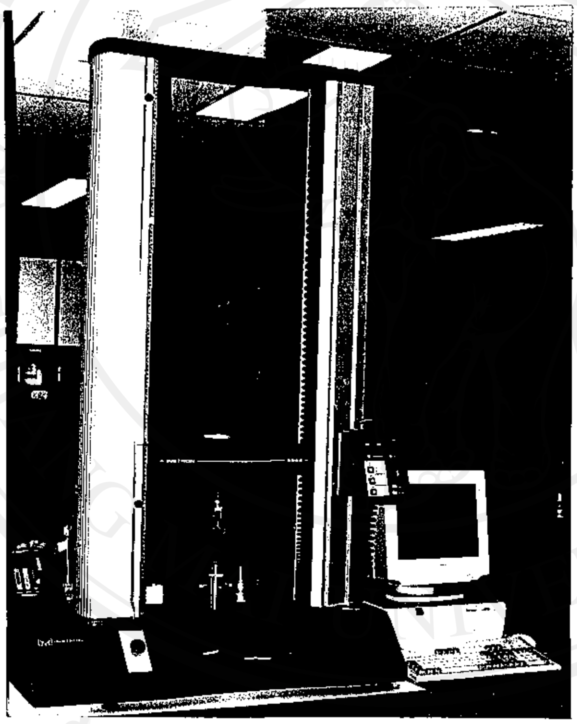


Figure 3.2 Universal testing machine with vertical stainless steel hooks

2. Electronic measuring microscope, model MM-11 (Nikon, Japan), was used for digital measuring the original chain length of elastomeric chains to the nearest of 0.001 millimeter.

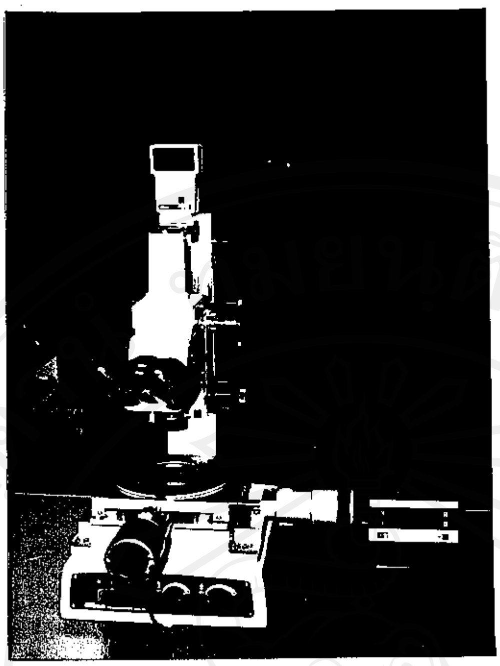


Figure 3.3 Electronic measuring microscope

Part II: Force decay of elastomeric chains with simulated tooth movement

1. Six metal jigs, 5.0 x 17.5 x 1.5 centimeters in closing condition, were constructed for stretching and holding the elastomeric chains. A metal jig composed of 2 half symmetrical metal plates, the left and the right metal plates, which were connected by three standard one-millimeter metal expansion screws, an adjusting screw and two stabilizing screws, and two guiding brass pins. All of them were placed horizontally and parallelly to each others. The adjusting screw was embedded in the middle of both metal plates for varying the distance between two metal plates. The other two stabilizing screws were embedded only in the right metal plate, one-centimeter from each end of the metal plate, and their tips of screws touched at the medial side of the left metal plate. They were activated in the same amount of the adjusting screw for keeping the consistency of metal plate distance. Two guiding brass pins were embedded in both metal plates medially to the stabilizing screws for controlling parallelism of both metal plates (Figure 3.4A).

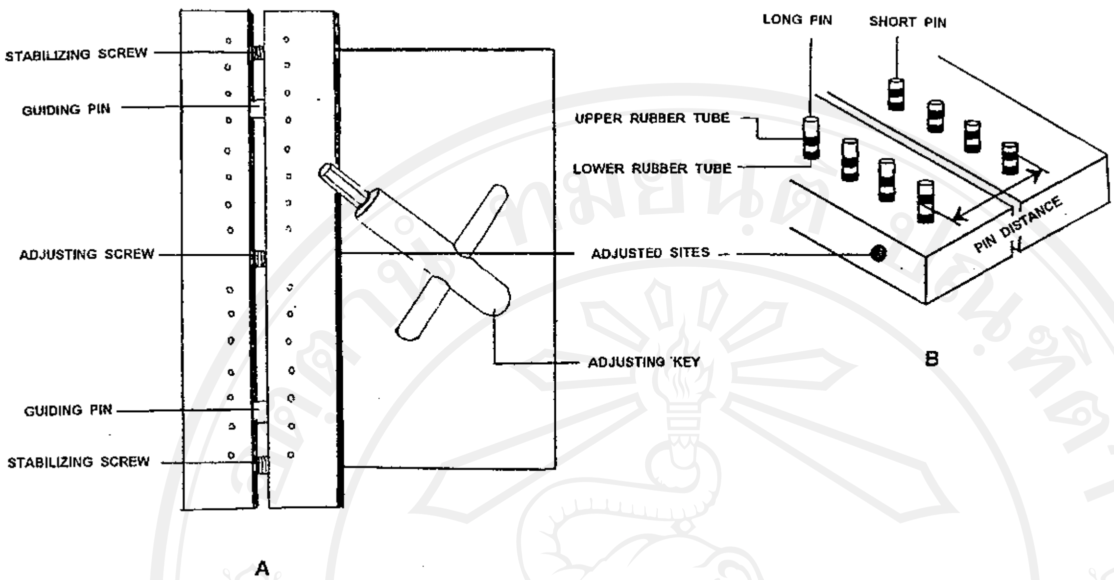


Figure 3.4 Metal jig for providing varied extension of elastomeric chains
A) metal jig and adjusting key B) pin distance measurement

Fifteen vertical stainless steel pins, 1.2 millimeters in diameter, were mounted in a row of one-centimeter apart on either side of the midline of the metal jig in identical fashion (Figure 3.4A). The long pin row was designed on the left metal plate, as fixed pins and the short pin row was on the right metal plate, as stretched pins for convinence of force measurement. Each vertical pin was fixed with a five-millimeter long lower rubber tube at its base for keeping the same height of all samples. Then, a sample of elastomeric chain was following placed, and a similar upper rubber tube was fixed for locking it in place. The distance between each pair of pins was measured from the outer sides of the contralateral pins (Figure 3.4B).

The initial pin distance was 22.5 millimeters that was represented the distance between the mesial wing of the upper canine bracket and mesial of the buccal tube with hook at mid-mesiobuccal cusp of upper first molar, which were placed on the average size of teeth as shown in Figure 3.5 (Modified from Srisopark, 1972; Pattanaporn, 1983).

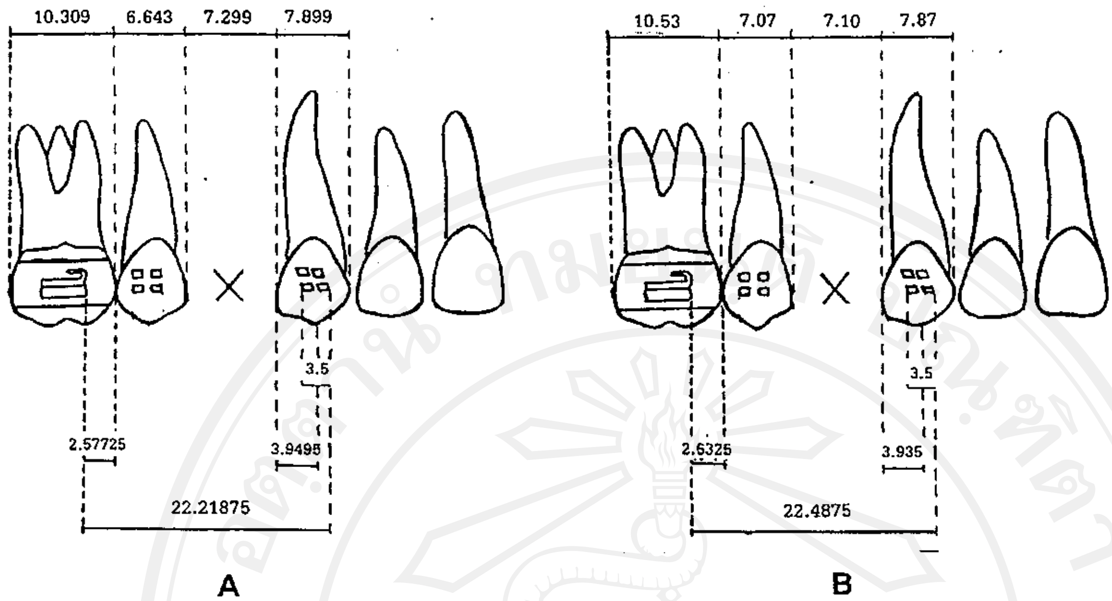


Figure 3.5 The initial pin distance modified from the studies of tooth size analysis A) by Srisopark (1972) and B) by Patanaporn (1983)

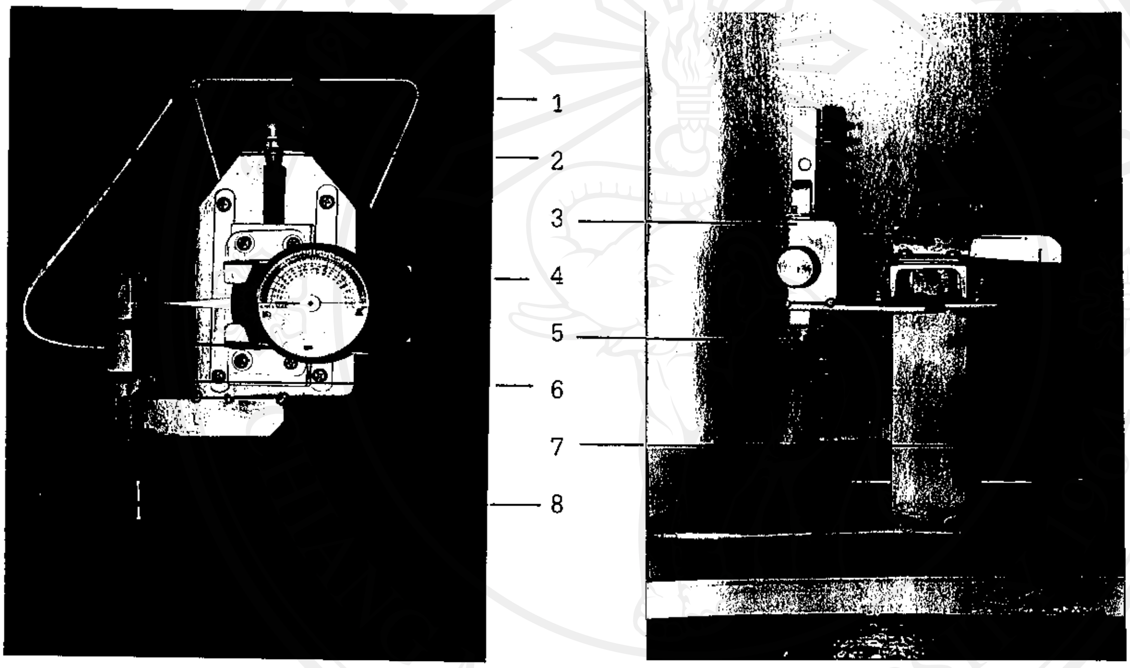
Two metal jigs were marked for each group: closed, open and wide space elastomeric chains. The reliability test of the pin distance of the metal jigs were done two times, before and after this investigation by the same investigator. The distances between each pair of the pins were measured by a digital vernier caliper to evaluate means and standard deviations. The initial and the final pin distance distribution and one-way analysis of variance (ANOVA) among three groups were shown in Appendix A. There was no significant difference among three groups of the metal jigs in both initial and final pin distances.

2. Two sealed clear plastic boxes with 100 percent humidity, which were prepared by lining the bottom of the plastic boxes with one-centimeter thickness of cotton and totally soaking with a liter of distilled water 24 hours before testing and throughout the six-week period. A fit plastic perforated plate was lay over the soaking cotton for supporting the metal jigs. Each plastic box could be stored three metal jigs, a four-loop closed space jig, a three-loop open space jig and a three-loop wide space jig.

3. A Correx gauge (Federwaage 100-500 grams, dial type, stress and tension gauge, Dentaurum, Germany) was used to obtain force magnitude data. Since the original gauge was scaled in 10-gram increments with a range of 100 to 500 grams, so it was necessary to modify the scale for better accuracy in the force magnitude reading. Therefore, a standard 180-degree scale clear plastic sheet that was scaled in one-degree increments, was fixed on the dial of the Correx gauge. The 180 degrees equals to 500 grams, so that one degree of force is approximated to 2.78 grams. The Correx gauge was calibrated by standard weights which were guarantied by the Central Bureau of Weights and Measures, Thailand, before testing and recalibrated after the test to insure the validity of readings. The Correx gauge arm and the scale pointer before using should be at the zero degree scale. The force magnitude was read to the nearest 0.5 degrees. The degrees of force were converted into gram-unit and recorded.

Figure 3.6 The Correx gauge with modified dial

4. The stable aluminium stander composed of a vertical metal jig holder and a horizontal Correx gauge holder (Figure 3.7). Both of them were perpendicular to each other. A straight stainless steel wire pointer, 1.2 millimeters in diameter, was fixed on the left side of the gauge-holder at the same level and paralleling to the Correx gauge arm for setting the gauge position.



A. Top view B. Side view

Figure 3.7 The stable stander with a metal jig and a Correx gauge

1. Gauge-moving screw
2. Correx gauge arm
3. Jig holder part of the stander
4. Horizontal Correx gauge
5. Vertical metal jig
6. Stainless steel wire pointer
7. Gauge-holder part of the stander
8. Jig-holder screw

5. An incubator (Memmert model 200, Memmert Corporation, Germany) was constantly maintained at 37°C ± 1°C and the temperature was checked daily. All samples were stored in this incubator during the six-week period.

Figure 3.8 Incubator, Memmert model 200

6. A digital vernier caliper (Diginatic, Mitutoyo, Japan), which measured to the nearest 0.01 millimeter, was used for measuring the pin distances of metal jigs.

Figure 3.9 Digital vernier caliper

7. An orthodontic elastic forceps was used to hold the elastomeric chains.

METHODS

Part I: Elastic limit of orthodontic elastomeric chains

The samples for tensile testing comprised of three and four loops of three different unit forms, which were closed, open and wide space orthodontic elastomeric chains. There were ten samples for each group that were as follow:

Group 1 Three-loop closed space elastomeric chain (C_3)

Group 2 Four-loop closed space elastomeric chain (C_4)

Group 3 Three-loop open space elastomeric chain (O_3)

Group 4 Four-loop open space elastomeric chain (O_4)

Group 5 Three-loop wide space elastomeric chain (W_3)

Group 6 Four-loop wide space elastomeric chain (W_4)

A total of 60 pieces of chains were tensile tested to determine the force magnitude and displacement required at the elastic limit. The original chain length within each group were measured in passive state from the internal surface of the most distant aspect of one terminal loop to the same location on the other terminal loop (Figure 3.10) by an electronic measuring microscope and recorded in millimeters. The original chain length distribution of six groups were shown in Table 3.1.

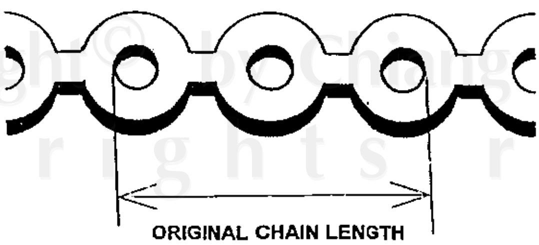


Figure 3.10 The original chain length measurenent of three-loop open space elastomeric chain

Table 3.1 The original chain length distribution of six groups of orthodontic elastomeric chains

Group	Type of E-chain	No. of samples	Original Chain Length (mm.)			
			Min.	Max.	$\overline{X}$	SD
1	C_3	10	6.898	6.988	6.942	0.026
2	C_4	10	9.686	9.760	9.732	0.031
3	O_3	10	8.166	8.463	8.340	0.084
4	O_4	10	11.684	11.928	11.825	0.081
5	W_3	10	10.260	10.384	10.342	0.040
6	W_4	10	14.775	15.017	14.871	0.085

This investigation was performed in the laboratory of Faculty of Dentistry, Mahidol University under the room condition, $25^{o}C \pm 1^{o}C$ and $50 \pm 5\%$ humidity. The tensile testing was performed on an Instron® universal testing machine with a vertical stainless steel hook on each crosshead. The initial distance between the upper and the lower hooks, original gage length, equaled to the mean original chain length of each group, and the Instron® universal testing machine was set as zero.

The American Standard for Testing and Materials (ASTM) D 882 suggested that the crosshead speed (millimeter per minute) was depended on the initial strain rate that must be equal in all groups for comparable values. The strain rate in this experiment was 200% per minute for obtaining optimum load-displacement curve. Therefore, the crosshead speed for each group of elastomeric chain was two times of its mean original gage length per minute as shown in Table 3.2.

Table 3.2 The original gage length and the crosshead speed in tension test

Group	Groups of Elastomeric chains	Original gage length $\bar{X}$ (mm.)	Crosshead speed (mm./min)
1	C_3	6.942	13.884
2	C_4	9.732	19.464
3	O_3	8.340	16.678
4	O_4	11.825	23.650
5	W_3	10.342	20.684
6	W_4	14.871	29.742

The maximum load in this tension test was set at 10 newtons. Following the calibration procedure, a sample was suspended on the vertical hooks of both crossheads of the Instron® universal testing machine and the testing in tension was initiated. The force magnitude and the displacement of elastomeric chain were recorded digitally and graphically. The force-displacement curve which was shown on the computer monitor was selectively marked at the elastic limit point, the greatest stress which a material is capable of sustaining without any permanent strain remaining upon complete release of the stress (ASTM D 1566). The force magnitude (elastic strength) and displacement at elastic limit point were recorded by load transducer and the internal chart recorder.

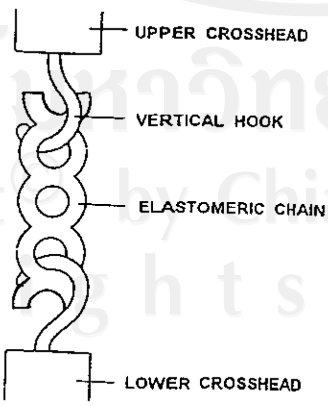


Figure 3.11 Loading an elastomeric chain on hooks of upper and lower crossheads of the Instron® universal testing machine

All groups had been performed with the same manner. It is necessary to set the original gage length and the crosshead speed according to their original chain length of elastomeric chains prior to the tensile test. Then, the percent elongation, the extension of a uniform section of a specimen expressed as percent of the original length (ASTM D 1566), at elastic limit was calculated from the original chain length and the displacement of each sample by the following equation

$$\text{Percent elongation (\%)} = \frac{\text{Displacement at elastic limit (mm.)}}{\text{Original chain length (mm.)}} \times 100$$

The graphs of force versus displacement were obtained, and the generated force, displacement and percent elongation at elastic limit were recorded.

Part II: Force decay of elastomeric chains with simulated tooth movement

This experiment was performed at the Faculty of Dentistry, Chiang Mai University at room temperature, $30^{\circ}C \pm 2^{\circ}C$. Three groups for thirty samples each of orthodontic elastomeric chains were four-loop closed space (C_4), three-loop open space (O_3) and three-loop wide space (W_3) that produced the initial force range 300 grams to 400 grams at 22.5 millimeters extension. Fifteen samples of each group were tested at a time. All samples in each group were measured the initial force magnitudes.

INSTRUMENT SETTING UP

For force measuring, the metal jig and Correx gauge were placed on the stable stander as Figure 3.7. The metal jig was placed in the vertical metal jig holder, and was set and locked at proper position that the tested pair of pins was in the same level to the stainless steel wire pointer. The long pin row of the metal jig was on the left side of the operator. The Correx gauge position was adjusted to place in optimal anteroposterior position perpendicular to the metal jig and parallel to horizontal plane. The Correx gauge could be moved only in horizontal plane by turning the gauge-moving screw to stretch the elastomeric chains, which was measured the force simutaneously. Therefore, the force measurement was performed in the horizontal plane. Then the jig position was readjusted to test another next sample by the same manner.

All metal jigs were initially set at 22.5-millimeter pin distance. Then, the lower rubber tubes were fixed at all pins. All metal jigs were preconditioned as simulated oral environment 24-hour before testing by storing in the seal plastic boxes under 100% humidity condition with constant 37^{o}C in the incubator. For force decay with simulated tooth movement study, the samples were suspended between every pair of pins on the metal jig. A metal jig was removed from the plastic box one by another. One end of loop of fifteen elastomeric chains were placed on each fifteen long pins, one for each, following with the upper rubber tubes. Then, the metal jig with elastomeric chains was placed at metal-jig holder of the stander according to the instrument setting up method. The other end loop of each sample was hold by orthodontic elastic forceps and transfered to the Correx gauge arm which was medially to the short pin tip before the stretching. Then, each sample was slowly stretched for 22.5 millimeters to the short pin by turning the gauge-moving screw until the Correx gauge arm pointed at the tip of short pin. The force magnitude was read immediately and recorded. After that, the free end loop was transfered to the short pin in stretching condition, and fixed with an upper rubber tube. After all fifteen samples were measured the force and recorded, they were placed in the 37^{o}C preheated incubator.

Throughout six-week period all samples were removed from the incubator for a few minutes necessary to measure force magnitudes thereafter at the first day, and at the end of 1^{st}, 2^{nd}, 3^{rd}, 4^{th}, 5^{th} and 6^{th} week respectively. Then they were quickly returned to the testing condition.

The force measurement at each specific time was obtained almost the same manner as previous description, except of the manipulation of each sample prior to force measurement. The upper rubber tubes at the short pin were removed and repositioned the upper rubber tubes at the long pin to the pin tip. Then, the last loop at the short pin of each sample was transfered to the Correx gauge arm and the force measurement was proceeded as previous.

Since this study would evaluate the force decay rates and patterns of elastomeric chains during six weeks with simulated tooth movement. Following the force measurements of each time period, the pin distance was decreased 0.5 millimeters per week for simulated tooth movement.

After the exerted forces at each time period were obtained, they were calculated for the following variables:

1. Percent remaining force (%) $= \dfrac{\text{Remaining force in each time period (gm.)} \times 100}{\text{Initial force (gm.)}}$
2. Force decay (gm.) = Force at the end of the prior period (gm.) - Force at the end of that period (gm.)
3. Percent Force decay (%) $= \dfrac{\text{Force decay (gm.)} \times 100}{\text{Initial force (gm.)}}$
4. Force decay rate (gm/day) $= \dfrac{\text{Force decay in each time interval (gm.)}}{\text{Number of days in that time interval (days)}}$
5. Percent force decay rate (%/day) $= \dfrac{\text{Percent force decay in the time interval (\%)}}{\text{Number of days in that time interval (days)}}$

Then, all variables were statistical tested for comparing their differences by SPSS for Window Release 6.0 program.

STATISTICAL ANALYSES

The data were proceeded by the SPSS for Window Release 6.0 program (SPSS Inc., Chicago, 1989-1993) as following:

1. Descriptive analysis was used for determining means and standard deviations of;

1.1 generated force at elastic limit for both different length of three forms of elastomeric chains,

1.2 displacement at elastic limit for both different length of three forms of elastomeric chains,

1.3 percent elongation at elastic limit for both different length of three forms of elastomeric chains,

1.4 the remaining force and percent remaining force at each time period of three forms of elastomeric chains,

1.5 the force decay and percent force decay at each time interval of three forms of elastomeric chains, and

1.6 the force decay rate and percent force decay rate at each time interval of three forms of elastomeric chains.

2. Two way analysis of variance (ANOVA) was used for comparing the generated force and percent elongation at elastic limit among three-loop and four-loop groups of three forms of elastomeric chains. Therefore, null hypotheses, H_o, were:

2.1 H_o: There was no significant differences of generated force at elastic limit among three-loop and four-loop groups of three forms of elastomeric chains,

2.2 H_o: There was no significant differences of percent elongation at elastic limit among three-loop and four-loop groups of three forms of elastomeric chains.

3. One way analysis of variance (ANOVA), was used for testing the percent remaining force and the percent force decay rate among three forms of elastomeric chains in the same time. If there were significant differences, the

multiple comparisons were following tested. Therefore, null hypothesis, H_o, were:

3.1 H_o: There were no significant difference of the percent remaining force among three forms of elastomeric chains in each period.

3.2 H_o: There were no significant difference of the percent force decay rate among three forms of elastomeric chains in each time interval.

4. One way analysis of variance (ANOVA) with repeated one factor, was used for testing the percent remaining force and the percent force decay rate among different times in the same form of elastomeric chains. If there were significant differences, the multiple comparisons were following tested. Therefore, null hypothesis, H_o, were:

4.1 H_o: There were no significant difference of the percent remaining force of three forms of elastomeric chains among eight periods.

4.2 H_o: There were no significant difference of the percent force decay rate of three forms of elastomeric chains among seven time intervals.

The probability of significance was denoted as * for $p< 0.05$, ** for $p< 0.01$ and *** for $p< 0.001$.