ANNUAL REPORT OF

AN EVALUATION OF A NOVEL MOUTH RINSE TO CONTROL DENTAL CARIES IN CHILDREN

2009

1.0 DOCUMENT CONTROL

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2.0 CONTENTS

3.0 INTRODUCTION

This annual report presents descriptive and analytical statistics from first year data of a three year clinical trial of a novel mouth rinse to control dental caries in school children commenced in October 2007. This report should not be released for public comment or cited until accepted and endorsed by the respective parties listed on page 2 and authorised by the author.

4.0 STUDY AIM AND DESIGN

The aim of this research project is to evaluate a chair side bacterial detection and treatment protocol, known commercially as CariFree™, in 5-10 year old school children attending the Musgrave Hill (Gold Coast) and Vincent (Townsville) school dental clinics. A double blind randomised clinical trial study design using a treatment and placebo arm was employed for this purpose. Both the placebo and treatment mouth rinse were administered to children using the same clinical protocol, with the only difference being the active component of the treatment mouth rinse, which is not known.

A risk assessment protocol was used to determine caries risk of each child participant by completion of a validated oral health questionnaire, assessment of oral bacterial biofilm activity[1], measured in relative light units (RLU) using an adenosine triphosphate (ATP) bioluminescence meter, CariScreen™ and Mutans streptococci (MS) counts[2] using a chair side culture system, CariCult™. RLU's were recorded directly from the meter (Range 0-9999) and MS counts were recorded after 48 hours incubation by counting the highest density of colony forming units per square centimetre and allocation to the appropriate descriptive category (Low, Medium and High). Each child was examined at baseline and their current caries activity recorded using WHO recognised caries scores of decayed, missing and filled primary and permanent teeth surfaces (termed dmfs and DMFS respectively)[3]. Measures of caries experience were expressed as the caries index to allow for the changing

number of teeth present in each child's mouth due to primary tooth shedding and permanent tooth emergence during normal physiological dental growth and development. A periodic caries index for each child was determined by dividing the summation of the dmfs and DMFS scores by the number of tooth surfaces present at time of examination. Caries increment was calculated by subtracting the periodic caries index from the baseline caries index at the completion of each year of the study to express relative change (positive, negative or no change) of caries index across time.

Baseline and progressive ATP activity and microbiological levels of MS were recorded after each mouth rinse cycle to assess the effectiveness of the treatment and placebo mouth rinse in reducing oral bacterial biofilm activity and MS counts in caries active children. Test results for each child's ATP levels measured in RLU and MS counts were compared at each time interval for significant change between the mouth rinse groups.

5.0 DATA MANAGEMENT

All clinical and questionnaire information was electronically transferred to an Excel data spreadsheet and saved on a data memory stick prior to further data management. The data were cross checked and changed when corresponding fields were not correctly matched. An example of data entry error was when the sum of the d, m and f components did not match the recorded dmfs score. Decayed or filled tooth surfaces were double checked against previous records to ensure all diseased surfaces were recorded correctly. When computational errors were identified, the total number of diseased tooth surfaces per child was adjusted down to balance with the total score. All changes made to the received data files and a copy file of the changed data was then saved on a password protected laptop database. This database file also recorded any potential errors such as missing data and any consistency adjustments that were made to ensure all dental examinations had corresponding matched records in the child's questionnaire file.

The data management and statistical analysis were performed using the SPSS (V.17) program and saved as a data output file on the laptop database. Updates to the statistical analysis were saved manually under several sub-file headings and saved to the original database.

Descriptive statistics using frequency and descriptive functions were calculated to determine the percentage of RLU and CFU groups, mean caries indices and standard deviation for each treatment group and each school. Progressive and baseline caries indices for each child were compared for trends using the paired Student T test. Cross tabulations of selected categorical variables and comparison of annual mean caries increments for each study and school group were performed using a Pearson Chi Square and analysis of variance procedure at the 5% level of significance.[4]

6.0 RESULTS

Treatment groups:

The number of enrolled participants at the Musgrave Hill (MH) site was 214 and 92 at Vincent (V) site with 22 (10%) and 1 (1%) dropouts by the end of 2008 respectively. Children were randomly assigned to two treatment groups (termed Galah and Kookaburra) as shown in Table 1 and 2. The identity of the treatment and placebo groups remains unknown at this stage.

Table 1. Musgrave Hill mouth rinse group

	Frequency	Percent	Valid Percent	Cumulative Percent
Galah	**103**	**48.1**	**48.1**	**48.1**
Kookaburra	**111**	**51.9**	**51.9**	**100.0**
Total	**214**	**100.0**	**100.0**	

Table 2. Vincent mouth rinse group

	Frequency	Percent	Valid Percent	Cumulative Percent
Galah	44	47.8	47.8	47.8
Kookaburra	48	52.2	52.2	100.0
Total	92	100.0	100.0	

Treatment cycles:

Each group was given the placebo or treatment mouth rinse cycles four times during the year to coincide with the Education Queensland term schedule. The total number of daily mouth rinses given to each child participant varied during each four to six weekly cycle, allowing for staff ADO's and other school events. Each child was offered the mouth rinse according to their group designation and asked to hold and swish in their mouth for 30 seconds before expectoration. The number of treatment mouth rinses were recorded in the treatment register for Musgrave Hill and Vincent are shown in Tables 3 and 4.

Table 3. Musgrave Hill treatment cycles

Mouth Rinse Cycles						
	N	Range	Minimum	Maximum	Mean	Std. Deviation
1 Tx Cycle	194	22	1	23	12.4	3.4
2 Tx Cycle	186	27	1	28	13.5	5.0
3 Tx Cycle	171	26	1	27	19.1	4.1
4 Tx Cycle	125	13	8	21	16.9	2.8

Table 4. Vincent treatment cycles

Mouth Rinse Cycles						
	N	Range	Minimum	Maximum	Mean	Std. Deviation
1 Tx Cycle	88	16	1	17	10.4	3.2
2 Tx Cycle	82	14	1	15	11.5	2.6
3 Tx Cycle	81	12	6	18	14.7	3.2
4 Tx Cycle	78	12	7	19	16.3	2.7

Caries groups:

The number of caries active children at the Musgrave Hill site was 148 (69%) and at the Vincent site was 72 (77%). Dental examinations were conducted at the beginning (1), mid year (2) and end of the year (3). A caries index for each child was calculated by dividing the number of decayed, filled or missing tooth surfaces by the total number of tooth surfaces in the child's mouth at each examination. One case at Vincent school with multiple extracted teeth was excluded from the statistical analysis due to the potential outlier effect. Descriptive statistics of caries index for children with active disease at each school are shown in Tables 5 and 6 and Figures 1 and 2 respectively.

Table 5. Musgrave Hill caries index

Descriptive Statistics

		Caries Index 1	Caries Index 2	Caries Index 3
N	Valid	**148**	**129**	**117**
	Missing	**0**	**19**	**31**

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Caries Index 1	**148**	**.01**	**.69**	**.098**	**.095**
Caries Index 2	**129**	**.00**	**.44**	**.089**	**.087**
Caries Index 3	**117**	**.00**	**.39**	**.089**	**.078**

Table 6. Vincent caries index

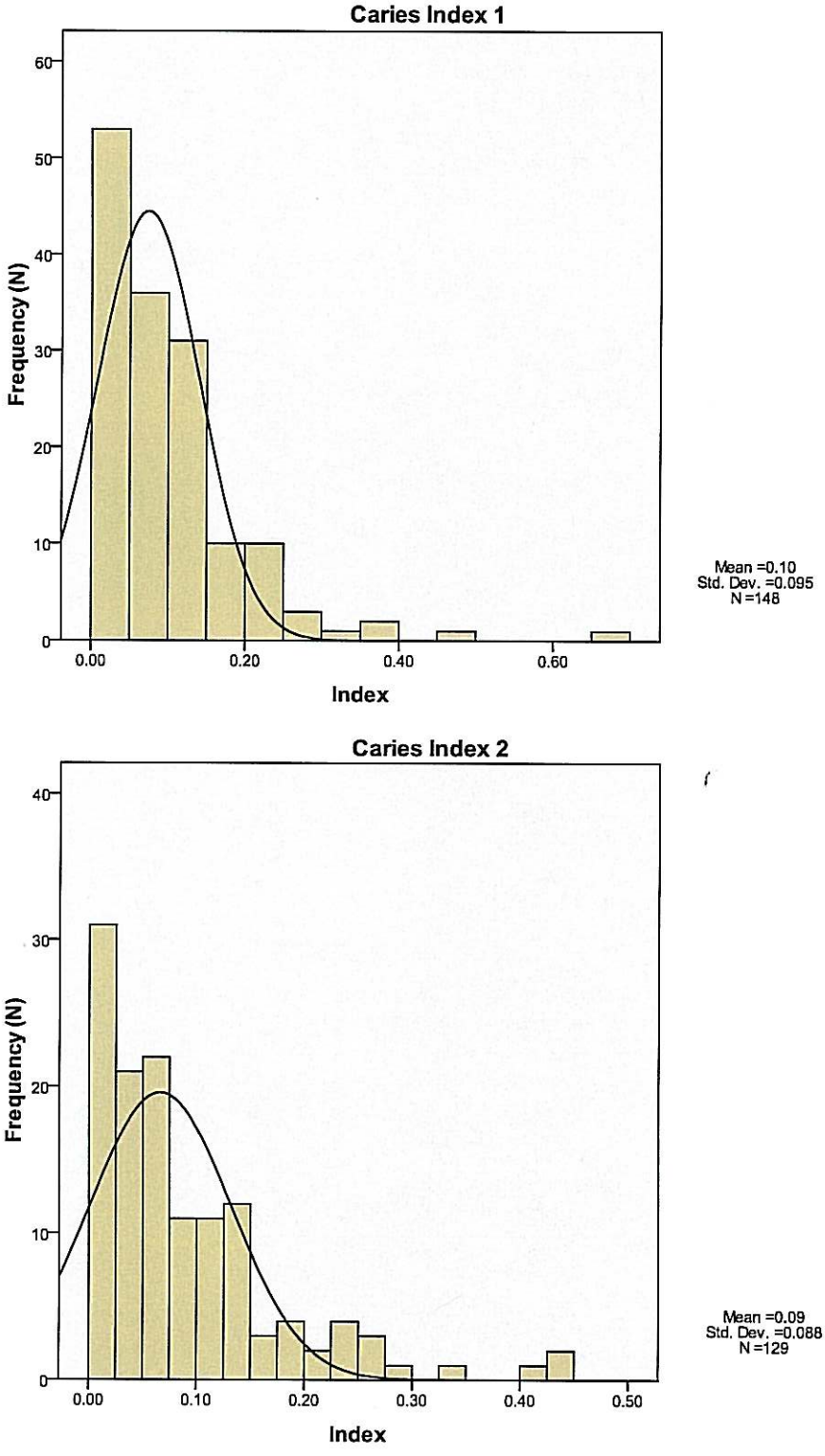
Descriptive Statistics

		Caries Index 1	Caries Index 2	Caries Index 3
N	Valid	**71**	**64**	**58**
	Missing	**0**	**7**	**13**

Descriptive Statistics

	N	Minimum	Maximum	Mean	Std. Deviation
Caries Index 1	71	.01	.41	.098	.094
Caries Index 2	64	.00	.40	.101	.099
Caries Index 3	58	.00	.42	.091	.094

Figure 1. Musgrave Hill caries index at each examination



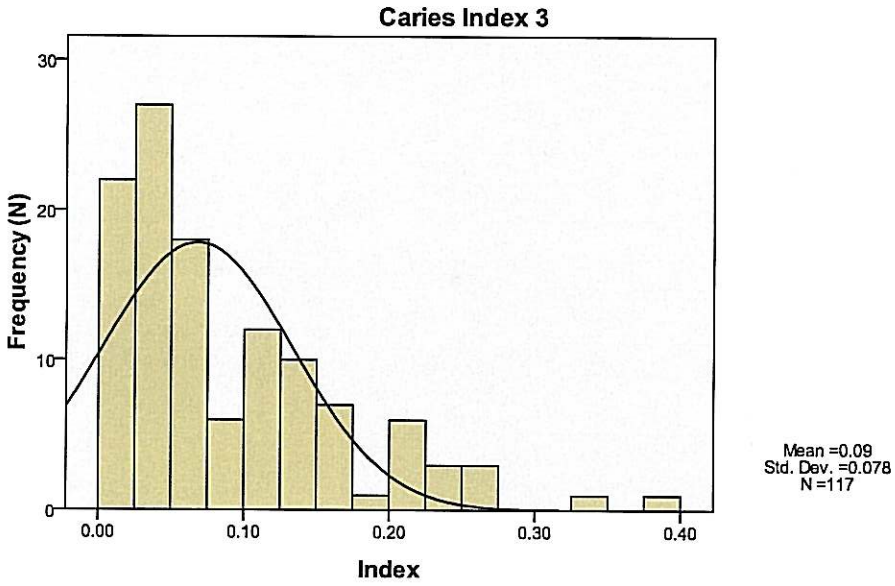
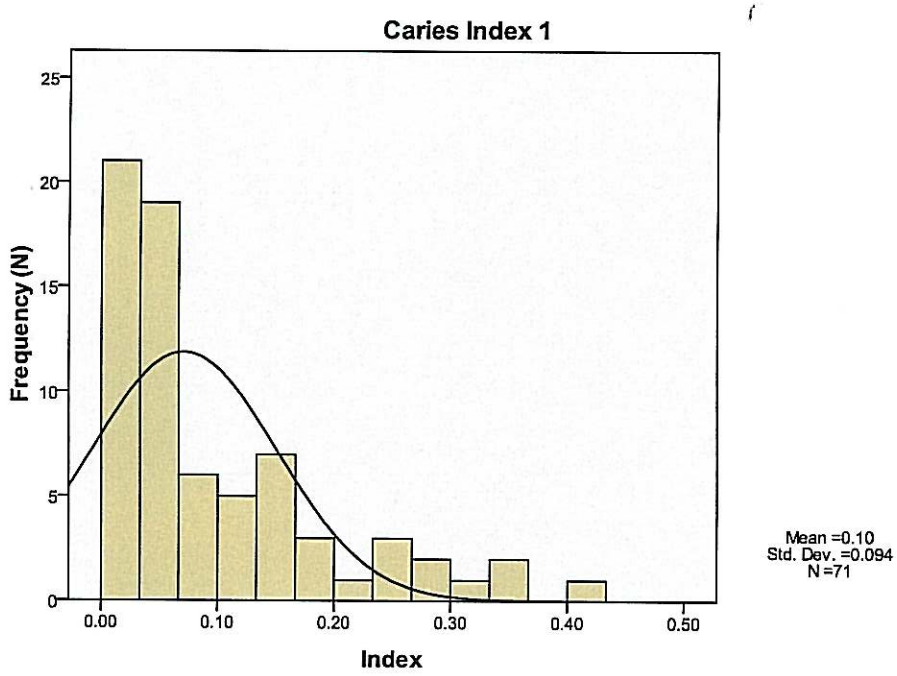
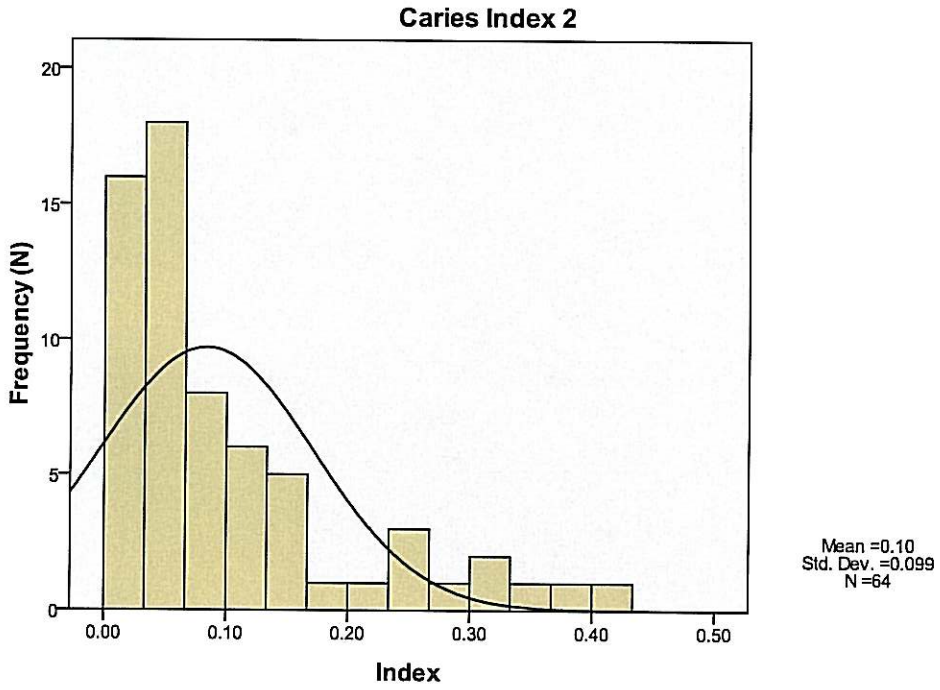
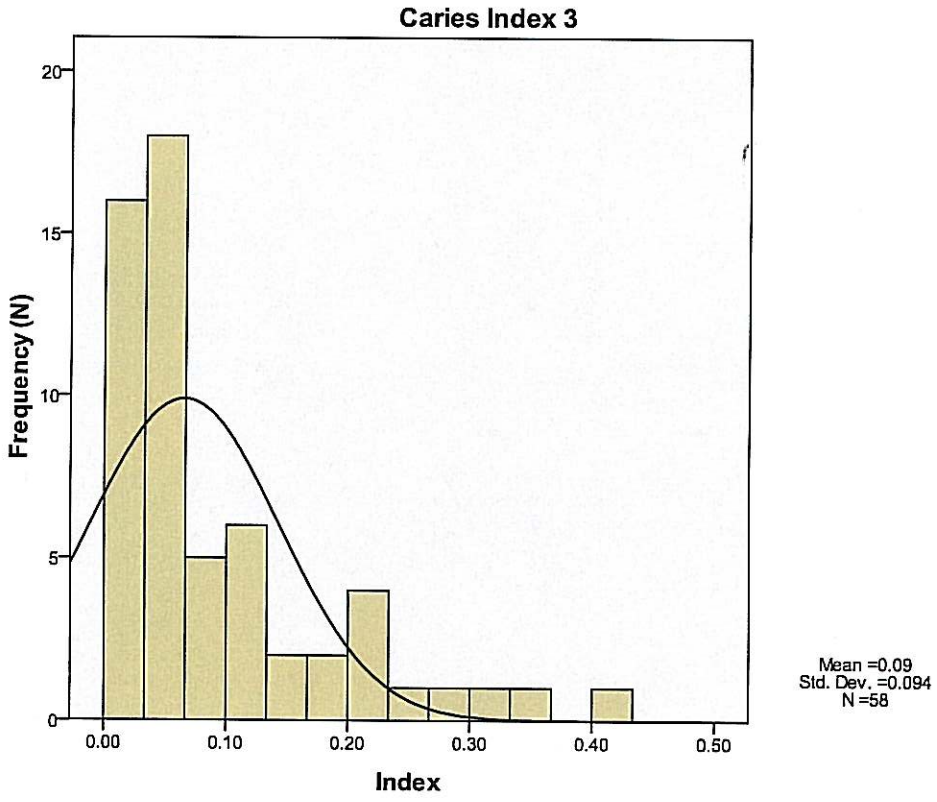


Figure 2. Vincent caries index at each examination




Caries Index 2
Frequency (N)
20
15
10
5
0
0.00
0.10
0.20
0.30
0.40
0.50
Index
Mean =0.10
Std. Dev. =0.099
N =64


Caries Index 3
Frequency (N)
20
15
10
5
0
0.00
0.10
0.20
0.30
0.40
0.50
Index
Mean =0.09
Std. Dev. =0.094
N =58

Mean caries index 1, 2 and 3 was compared between schools using ANOVA and was found to be not significant.

ANOVA

Caries index 1

	Sum of Squares	df	Mean Square	F	Sig.
Between Schools	**3.702**	**1**	**3.702**	**2.056**	**.153**
Within Schools	**392.548**	**218**	**1.801**		
Total	**396.250**	**219**			

ANOVA

Caries index 2

	Sum of Squares	df	Mean Square	F	Sig.
Between Schools	**1.155**	**1**	**1.155**	**2.261**	**.134**
Within Schools	**98.077**	**192**	**.511**		
Total	**99.232**	**193**			

ANOVA

Caries index 3

	Sum of Squares	df	Mean Square	F	Sig.
Between Schools	**.186**	**1**	**.186**	**1.992**	**.160**
Within Schools	**16.229**	**174**	**.093**		
Total	**16.414**	**175**			

Mean caries index 1, 2 and 3 were also compared between treatment groups and found to be not significant.

ANOVA

Caries index 1

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	**8.943**	**3**	**2.981**	**1.663**	**.176**
Within Groups	**387.307**	**216**	**1.793**		
Total	**396.250**	**219**			

ANOVA

Caries index 2

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	**2.299**	**3**	**.766**	**1.502**	**.215**
Within Groups	**96.933**	**190**	**.510**		
Total	**99.232**	**193**			

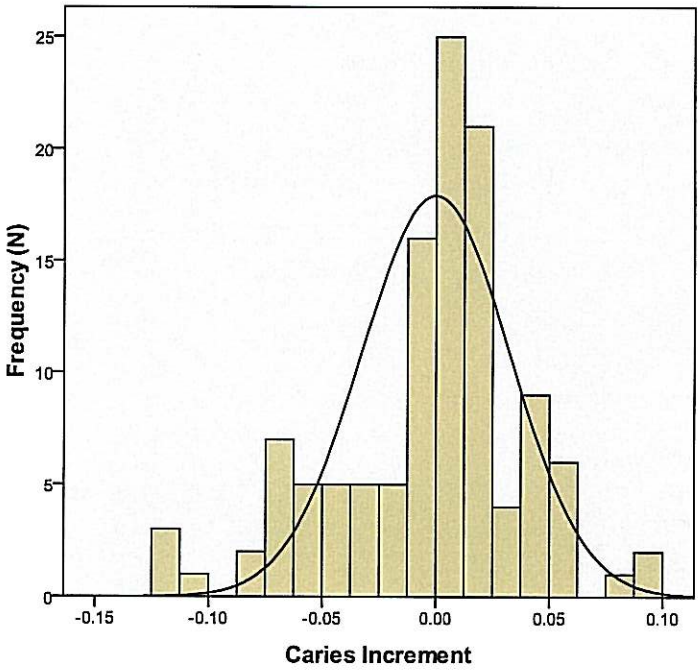
ANOVA

Caries index 3

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	**.390**	**3**	**.130**	**1.394**	**.246**
Within Groups	**16.025**	**172**	**.093**		
Total	**16.414**	**175**			

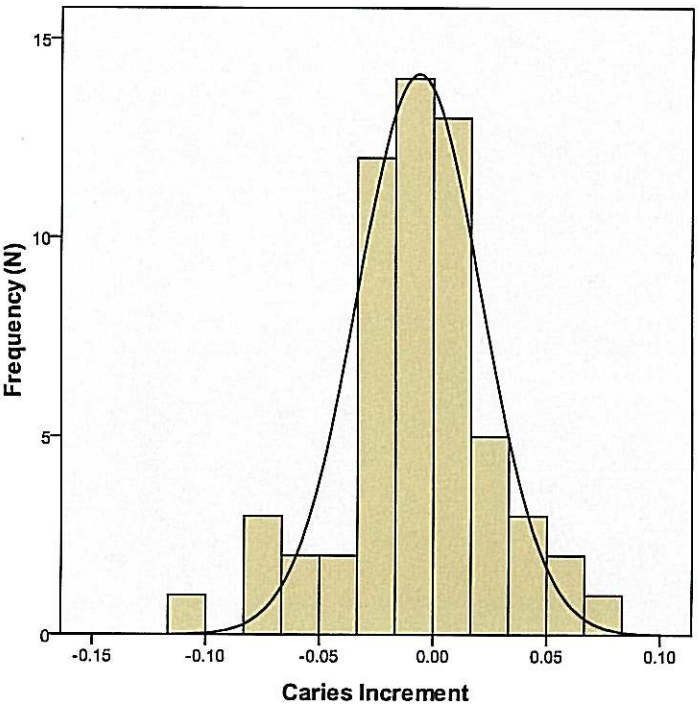
Caries increment was determined for each school by subtracting the calculated caries index at the end of the year (caries index 3) from the index at the beginning of the year (caries index 1). A positive value indicates a reduction of caries experience, zero indicates no change, and a negative value demonstrates an increase of caries experience during the year. The caries increment for Musgrave Hill is shown in Figure 3 and for Vincent in Figure 4 respectively.

Figure 3. Musgrave Hill Caries Increment 2008



Caries Increment 08		
Number	Valid	117
	Missing	31
Mean		0.0027
Median		0.00
Std. Deviation		0.043

Figure 4. Vincent Caries Increment 2008



Caries Increment 08		
Number	Valid	58
	Missing	13
Mean		0.0081
Median		0.01
Std. Deviation		0.033

Mean caries increment was compared between schools and treatment groups by ANOVA. The mean caries increment was not significantly different between either schools or treatment groups.

ANOVA

Caries increment 08

	Sum of Squares	df	Mean Square	F	Sig.
Between Schools	.001	1	.001	.849	.358
Within Schools	.265	173	.002		
Total	.267	174			

ANOVA

Caries increment 08

	Sum of Squares	df	Mean Square	F	Sig.
Between Groups	.004	3	.001	.920	.433
Within Groups	.262	171	.002		
Total	.267	174			

The combined caries increment for both schools during 2008 was determined and descriptive statistics are shown in Table 7. The mean caries increment was 0.004 or the equivalent of four less carious lesions for every 1000 tooth surfaces.

Table 7. Combined Schools Caries Increment 2008

Caries increment 08

Number	Valid	175
	Missing	45
Mean		0.0045
Median		0.0000
Std. Deviation		0.0399

CariScreen™ groups:

CariScreen™ tests were performed at the beginning and after completion of each treatment cycle of mouth rinse, a total of five tests for each child during the year. The Musgrave Hill CariScreen™ test results at each examination are shown in Table 8.

Table 8. Musgrave Hill CariScreen™ values (RLU's) at each examination

Cariscreen 1

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	101	47.2	47.2	47.2
	9000-9500	49	22.9	22.9	70.1
	>9500	64	29.9	29.9	100.0
	Total	214	100.0	100.0	

Cariscreen 2

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	65	30.4	31.4	31.4
	9000-9500	40	18.7	19.3	50.7
	>9500	102	47.7	49.3	100.0
	Total	207	96.7	100.0	
Missing	System	7	3.3		
Total		214	100.0		

Cariscreen 3

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	63	29.4	33.3	33.3
	9000-9500	31	14.5	16.4	49.7
	>9500	95	44.4	50.3	100.0
	Total	189	88.3	100.0	
Missing	System	25	11.7		
Total		214	100.0		

Cariscreen 4

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	64	29.9	35.2	35.2
	9000-9500	52	24.3	28.6	63.7
	>9500	66	30.8	36.3	100.0
	Total	182	85.0	100.0	
Missing	System	32	15.0		
Total		214	100.0		

Cariscreen 5

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	55	25.7	33.1	33.1
	9000-9500	41	19.2	24.7	57.8
	>9500	70	32.7	42.2	100.0
	Total	166	77.6	100.0	
Missing	System	48	22.4		
Total		214	100.0		

The Vincent CariScreen™ test results at each examination are shown in Table 9.

Table 9. Vincent CariScreen™ values (RLU's) at each examination

Cariscreen 1

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	45	48.9	48.9	48.9
	9000-9500	10	10.9	10.9	59.8
	>9500	37	40.2	40.2	100.0
	Total	92	100.0	100.0	

Cariscreen 2

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	30	32.6	34.1	34.1
	9000-9500	6	6.5	6.8	40.9
	>9500	52	56.5	59.1	100.0
	Total	88	95.7	100.0	
Missing	System	4	4.3		
Total		92	100.0		

Cariscreen 3

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	74	80.4	90.2	90.2
	9000-9500	5	5.4	6.1	96.3
	>9500	3	3.3	3.7	100.0
	Total	82	89.1	100.0	
Missing	System	10	10.9		
Total		92	100.0		

Cariscreen 4

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	71	77.2	89.9	89.9
	9000-9500	7	7.6	8.9	98.7
	>9500	1	1.1	1.3	100.0
	Total	79	85.9	100.0	
Missing	System	13	14.1		
Total		92	100.0		

Cariscreen 5

		Frequency	Percent	Valid Percent	Cumulative Percent
RLU	<9000	70	76.1	95.9	95.9
	9000-9500	2	2.2	2.7	98.6
	>9500	1	1.1	1.4	100.0
	Total	73	79.3	100.0	
Missing	System	19	20.7		
Total		92	100.0		

CariCult™ groups:

CariCult™ testing was performed in a similar manner and recorded at the beginning and after each treatment cycle of mouth rinse. The Musgrave Hill CariCult™ test results at each examination are shown in Table 10.

Table 10. Musgrave Hill CariCult™ values (CFU's) at each examination

Caricult 1

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	78	36.4	36.4	36.4
	Moderate	73	34.1	34.1	70.6
	Low	63	29.4	29.4	100.0
	Total	214	100.0	100.0	

Caricult 2

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	58	27.1	28.0	28.0
	Moderate	55	25.7	26.6	54.6
	Low	94	43.9	45.4	100.0
	Total	207	96.7	100.0	
Missing	System	7	3.3		
Total		214	100.0		

Caricult 3

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	91	42.5	48.1	48.1
	Moderate	70	32.7	37.0	85.2
	Low	28	13.1	14.8	100.0
	Total	189	88.3	100.0	
Missing	System	25	11.7		
Total		214	100.0		

Caricult 4

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	76	35.5	41.8	41.8
	Moderate	76	35.5	41.8	83.5
	Low	30	14.0	16.5	100.0
	Total	182	85.0	100.0	
Missing	System	32	15.0		
Total		214	100.0		

Caricult 5

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	89	41.6	53.9	53.9
	Moderate	57	26.6	34.5	88.5
	Low	19	8.9	11.5	100.0
	Total	165	77.1	100.0	
Missing	System	49	22.9		
Total		214	100.0		

The Vincent CariCult™ test results at each examination are shown in Table 11.

Table 11. Vincent CariCult™ values (CFU's) at each examination

Caricult 1

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	89	96.7	96.7	96.7
	Moderate	2	2.2	2.2	98.9
	Low	1	1.1	1.1	100.0
	Total	92	100.0	100.0	

Caricult 2

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	55	59.8	62.5	62.5
	Moderate	31	33.7	35.2	97.7
	Low	2	2.2	2.3	100.0
	Total	88	95.7	100.0	
Missing	System	4	4.3		
Total		92	100.0		

Caricult 3

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	21	22.8	25.9	25.9
	Moderate	52	56.5	64.2	90.1
	Low	8	8.7	9.9	100.0
	Total	81	88.0	100.0	
Missing	System	11	12.0		
Total		92	100.0		

Caricult 4

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	32	34.8	40.5	40.5
	Moderate	38	41.3	48.1	88.6
	Low	9	9.8	11.4	100.0
	Total	79	85.9	100.0	
Missing	System	13	14.1		
Total		92	100.0		

Caricult 5

		Frequency	Percent	Valid Percent	Cumulative Percent
CFU	High	13	14.1	17.8	17.8
	Moderate	51	55.4	69.9	87.7
	Low	9	9.8	12.3	100.0
	Total	73	79.3	100.0	
Missing	System	19	20.7		
Total		92	100.0		

School groups:

CariScreen™ and CariCult™ data were grouped by nominal categories to facilitate statistical analysis using a Pearson Chi-square test. Cross tabulations were performed between schools and these results are shown in shown in Tables 12 and 13. Pearson Chi Square analysis and level of significance are shown for each cross tabulation.

Table 12. Musgrave Hill and Vincent school CariScreen™ categories (RLU's) at each examination

Cariscreen 1 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Cariscreen 1 category	<9000	Count	101	45	146
		% within school	47.2%	48.9%	47.7%
	9000-9500	Count	49	10	59
		% within school	22.9%	10.9%	19.3%
	>9500	Count	64	37	101
		% within school	29.9%	40.2%	33.0%
Total		Count	214	92	306
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	6.939[a]	2	.031
N of Valid Cases	306		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 17.74.

Cariscreen 2 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Cariscreen 2 category	<9000	Count	65	30	95
		% within school	31.4%	34.1%	32.2%
	9000-9500	Count	40	6	46
		% within school	19.3%	6.8%	15.6%
	>9500	Count	102	52	154
		% within school	49.3%	59.1%	52.2%
Total		Count	207	88	295
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	7.471[a]	2	.024
N of Valid Cases	295		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 13.72.

Cariscreen 3 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Cariscreen 3 category	<9000	Count	63	74	137
		% within school	33.3%	90.2%	50.6%
	9000-9500	Count	31	5	36
		% within school	16.4%	6.1%	13.3%
	>9500	Count	95	3	98
		% within school	50.3%	3.7%	36.2%
Total		Count	189	82	271
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	75.561[a]	2	.000
N of Valid Cases	271		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 10.89.

Cariscreen 4 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Cariscreen 4 category	<9000	Count	64	71	135
		% within school	35.2%	89.9%	51.7%
	9000-9500	Count	52	7	59
		% within school	28.6%	8.9%	22.6%
	>9500	Count	66	1	67
		% within school	36.3%	1.3%	25.7%
Total		Count	182	79	261
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	67.630[a]	2	.000
N of Valid Cases	261		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 17.86.

Cariscreen 5 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Cariscreen 5 category	<9000	Count	55	70	125
		% within school	33.1%	95.9%	52.3%
	9000-9500	Count	41	2	43
		% within school	24.7%	2.7%	18.0%
	>9500	Count	70	1	71
		% within school	42.2%	1.4%	29.7%
Total		Count	166	73	239
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	80.181[a]	2	.000
N of Valid Cases	239		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 13.13.

Table 13. Musgrave Hill and Vincent school CariCult™ categories (CFU's) at each examination

Caricult 1 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Caricult 1 category	High	Count	78	89	167
		% within school	36.4%	96.7%	54.6%
	Moderate	Count	73	2	75
		% within school	34.1%	2.2%	24.5%
	Low	Count	63	1	64
		% within school	29.4%	1.1%	20.9%
Total		Count	214	92	306
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	94.359[a]	2	.000
N of Valid Cases	306		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 19.24.

Caricult 2 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Caricult 2 category	High	Count	58	55	113
		% within school	28.0%	62.5%	38.3%
	Moderate	Count	55	31	86
		% within school	26.6%	35.2%	29.2%
	Low	Count	94	2	96
		% within school	45.4%	2.3%	32.5%
Total		Count	207	88	295
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	56.063[a]	2	.000
N of Valid Cases	295		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 25.65.

Caricult 3 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Caricult 3 category	High	Count	91	21	112
		% within school	48.1%	25.9%	41.5%
	Moderate	Count	70	52	122
		% within school	37.0%	64.2%	45.2%
	Low	Count	28	8	36
		% within school	14.8%	9.9%	13.3%
Total		Count	189	81	270
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	**17.044**[a]	**2**	**.000**
N of Valid Cases	**270**		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 10.80.

Caricult 4 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Caricult 4 category	High	Count	**76**	**32**	**108**
		% within school	**41.8%**	**40.5%**	**41.4%**
	Moderate	Count	**76**	**38**	**114**
		% within school	**41.8%**	**48.1%**	**43.7%**
	Low	Count	**30**	**9**	**39**
		% within school	**16.5%**	**11.4%**	**14.9%**
Total		Count	**182**	**79**	**261**
		% within school	**100.0%**	**100.0%**	**100.0%**

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	**1.484**[a]	**2**	**.476**
N of Valid Cases	**261**		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 11.80.

Caricult 5 Cross tabulation between Schools

			School		Total
			Musgrave Hill	Vincent	
Caricult 5 category	High	Count	89	13	102
		% within school	53.9%	17.8%	42.9%
	Moderate	Count	57	51	108
		% within school	34.5%	69.9%	45.4%
	Low	Count	19	9	28
		% within school	11.5%	12.3%	11.8%
Total		Count	165	73	238
		% within school	100.0%	100.0%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	29.356[a]	2	.000
N of Valid Cases	238		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 8.59.

Treatment groups:

CariScreen™ and CariCult™ data were again grouped by nominal categories to facilitate statistical analysis using Pearson Chi-square test. Cross tabulations were performed between treatment groups with schools and these results are shown in shown in Tables 14 and 15. Pearson Chi Square analysis and level of significance are shown for each cross tabulation.

Table 14. CariScreen™ categories (RLU's) at each examination by Kookaburra (K) and Galah (G) groups within Musgrave Hill (MH) and Vincent (V) schools

Cariscreen 1 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Cariscreen 1 category	<9000	Count	53	22	48	23	146
		% within CS1cat	36.3%	15.1%	32.9%	15.8%	100.0%
	9000-9500	Count	22	7	27	3	59
		% within CS1cat	37.3%	11.9%	45.8%	5.1%	100.0%
	>9500	Count	28	15	36	22	101
		% within CS1cat	27.7%	14.9%	35.6%	21.8%	100.0%
Total		Count	103	44	111	48	306
		% within CS1cat	33.7%	14.4%	36.3%	15.7%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	10.475[a]	6	.106
N of Valid Cases	306		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 8.48.

Cariscreen 2 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Cariscreen 2 category	<9000	Count	30	14	35	16	95
		% within CS2cat	31.6%	14.7%	36.8%	16.8%	100.0%
	9000-9500	Count	15	2	25	4	46
		% within CS2cat	32.6%	4.3%	54.3%	8.7%	100.0%
	>9500	Count	54	27	48	25	154
		% within CS2cat	35.1%	17.5%	31.2%	16.2%	100.0%
Total		Count	99	43	108	45	295
		% within CS2cat	33.6%	14.6%	36.6%	15.3%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	11.205[a]	6	.082
N of Valid Cases	295		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 6.71.

Cariscreen 3 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Cariscreen 3 category	<9000	Count	28	38	35	36	137
		% within CS3cat	20.4%	27.7%	25.5%	26.3%	100.0%
	9000-9500	Count	15	1	16	4	36
		% within CS3cat	41.7%	2.8%	44.4%	11.1%	100.0%
	>9500	Count	48	2	47	1	98
		% within CS3cat	49.0%	2.0%	48.0%	1.0%	100.0%
Total		Count	91	41	98	41	271
		% within CS3cat	33.6%	15.1%	36.2%	15.1%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	77.045[a]	6	.000
N of Valid Cases	271		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 5.45.

Cariscreen 4 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Cariscreen 4 category	<9000	Count	31	36	33	35	135
		% within CS4cat	23.0%	26.7%	24.4%	25.9%	100.0%
	9000-9500	Count	26	3	26	4	59
		% within CS4cat	44.1%	5.1%	44.1%	6.8%	100.0%
	>9500	Count	31	0	35	1	67
		% within CS4cat	46.3%	.0%	52.2%	1.5%	100.0%
Total		Count	88	39	94	40	261
		% within CS4cat	33.7%	14.9%	36.0%	15.3%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	67.955[a]	6	.000
N of Valid Cases	261		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 8.82.

Cariscreen 5 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Cariscreen 5 category	<9000	Count	26	32	29	38	125
		% within CS5cat	20.8%	25.6%	23.2%	30.4%	100.0%
	9000-9500	Count	21	1	20	1	43
		% within CS5cat	48.8%	2.3%	46.5%	2.3%	100.0%
	>9500	Count	36	0	34	1	71
		% within CS5cat	50.7%	.0%	47.9%	1.4%	100.0%
Total		Count	83	33	83	40	239
		% within CS5cat	34.7%	13.8%	34.7%	16.7%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	80.453[a]	6	.000
N of Valid Cases	239		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 5.94.

Table 15. CariCult™ categories (CFU's) at each examination by Kookaburra (K) and Galah (G) groups within Musgrave Hill (MH) and Vincent (V) schools

Caricult 1 Cross tabulation by Groups with Schools

			Group				Total
			GMH	GV	KMH	KV	
Caricult 1 category	High	Count	38	43	40	46	167
		% within CC1	22.8%	25.7%	24.0%	27.5%	100.0%
	Moderate	Count	39	1	34	1	75
		% within CC1	52.0%	1.3%	45.3%	1.3%	100.0%
	Low	Count	26	0	37	1	64
		% within CC1	40.6%	.0%	57.8%	1.6%	100.0%
Total		Count	103	44	111	48	306
		% within CC1	33.7%	14.4%	36.3%	15.7%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	97.242[a]	6	.000
N of Valid Cases	306		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 9.20.

Caricult 2 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Caricult 2 category	High	Count	30	32	28	23	113
		% within CC2	26.5%	28.3%	24.8%	20.4%	100.0%
	Moderate	Count	25	10	30	21	86
		% within CC2	29.1%	11.6%	34.9%	24.4%	100.0%
	Low	Count	44	1	50	1	96
		% within CC2	45.8%	1.0%	52.1%	1.0%	100.0%
Total		Count	99	43	108	45	295
		% within CC2	33.6%	14.6%	36.6%	15.3%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	63.742[a]	6	.000
N of Valid Cases	295		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 12.54.

Caricult 3 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Caricult 3 category	High	Count	43	9	48	12	112
		% within CC3	38.4%	8.0%	42.9%	10.7%	100.0%
	Moderate	Count	34	27	36	25	122
		% within CC3	27.9%	22.1%	29.5%	20.5%	100.0%
	Low	Count	14	5	14	3	36
		% within CC3	38.9%	13.9%	38.9%	8.3%	100.0%
Total		Count	91	41	98	40	270
		% within CC3	33.7%	15.2%	36.3%	14.8%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	**17.826**[a]	**6**	**.007**
N of Valid Cases	**270**		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 5.33.

Caricult 4 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Caricult 4 category	High	Count	**38**	**17**	**38**	**15**	**108**
		% within CC4	**35.2%**	**15.7%**	**35.2%**	**13.9%**	**100.0%**
	Moderate	Count	**35**	**18**	**41**	**20**	**114**
		% within CC4	**30.7%**	**15.8%**	**36.0%**	**17.5%**	**100.0%**
	Low	Count	**15**	**4**	**15**	**5**	**39**
		% within CC4	**38.5%**	**10.3%**	**38.5%**	**12.8%**	**100.0%**
Total		Count	**88**	**39**	**94**	**40**	**261**
		% within CC4	**33.7%**	**14.9%**	**36.0%**	**15.3%**	**100.0%**

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	**2.067**[a]	**6**	**.913**
N of Valid Cases	**261**		

a. 0 cells (.0%) have expected count less than 5. The minimum expected count is 5.83.

Caricult 5 Cross tabulation by Groups within Schools

			Group				Total
			GMH	GV	KMH	KV	
Caricult 5 category	High	Count	43	5	46	8	102
		% within CC5	42.2%	4.9%	45.1%	7.8%	100.0%
	Moderate	Count	30	21	27	30	108
		% within CC5	27.8%	19.4%	25.0%	27.8%	100.0%
	Low	Count	9	7	10	2	28
		% within CC5	32.1%	25.0%	35.7%	7.1%	100.0%
Total		Count	82	33	83	40	238
		% within CC5	34.5%	13.9%	34.9%	16.8%	100.0%

Chi-Square Tests

	Value	df	Asymp. Sig. (2-sided)
Pearson Chi-Square	34.285[a]	6	.000
N of Valid Cases	238		

a. 2 cells (16.7%) have expected count less than 5. The minimum expected count is 3.88.

7.0 DISCUSSION

This clinical trial has now been underway for 20 to 23 months at the Vincent and Musgrave Hill research sites respectively. A significant reduction in recorded oral bacterial activity and culture following two mouth rinse cycles, particularly at the Vincent school, is very pleasing. Although a significant reduction of caries experience, measured by a positive or zero caries increment, is yet to be realised statistically, the reducing caries increment trend at both trial sites remains encouraging. Overall, the combined mean caries increment by the end of 2008 at the both sites was 0.0045, or translated into clinical terms, represents a reduction of four carious lesions for every 1000 tooth surfaces (i.e. approximately the equivalent of eight

primary school children with 120 tooth surfaces each) in one year. Applied across the spectrum of approximately 40,000 high risk children living in socially disadvantaged circumstances in Queensland in 2006[5], the potential therapeutic benefits of a school mouth rinsing program conducted in these communities are considerable.

The potential therapeutic benefit of the CariFree™ treatment protocol becomes more evident when the yearly increment for caries active children during 2008 at Musgrave Hill school (0.003) is compared with that of the previous year's increment (-0.01) based on 123 available patient records for 2007. The therapeutic benefit is calculated to be 13 carious lesions (-0.01-0.003 = -0.013) for every 1000 at risk tooth surfaces or eight children. Unfortunately, there are no previous records available at the Vincent site to determine the potential benefit gained at this site. The improvement of mean caries increment during 2008 at Vincent school is 0.008 or eight carious lesions for every 1000 at risk tooth surfaces or the equivalent of eight primary school children, suggesting that the treatment protocol may be more effective when the initial caries index is higher.

Both placebo and treatment mouth rinses seem to have comparable efficacy with no demonstrated advantage over one another in reducing biofilm activity or Mutans streptococci counts. They both seem to be equally effective in reducing the oral biofilm activity, particularly when the baseline measurements were high, as occurred at the Vincent site. However, it remains uncertain as to whether the current reduction in oral biofilm activity and Mutans streptococci counts will be sustainable and lead to a long term reduction of caries experience in susceptible children over time.

The fact that 64 children with no caries at the commencement of the clinical trial have remained decay free after four cycles of a mouth rinse protocol is also significant. Only eight (10%) children (three Galah and five Kookaburra) from Musgrave Hill school and five (6%) children (two Galah and three Kookaburra) from Vincent school have developed active dental caries to date from an initial disease free state.

Both research sites have sampled children with higher caries prevalence (69% at Musgrave Hill and 77% at Vincent) and caries severity (mean dmfs Musgrave Hill was 5.7 and 7.4 at Vincent) compared with the most recent Queensland state data (49% and 2.3 respectively) from 2001[6]. Similar cross sectional surveys[7] undertaken in the north Brisbane region in 1998-2002 also report lower caries experience data in this region (prevalence 35% and mean dmfs 2.9) compared with the trial sites. The inclusion of a greater proportion of six to ten year old children in the current research project compared with the previous surveys could partly explain the higher disease experience in this study given the time dependant nature of caries progression. However, the likelihood that these samples over-represent children from higher risk communities is the most probable explanation.

Enrolment and dropout rates at both sites have been commensurate with similar trials conducted in other centres. Despite some initial resistance, the participants have now accepted the mouth rinsing as part of their daily school routine. Surprisingly, the younger children have been more accepting of the program than the older children and are often quite keen to participate. Compliance with the maintenance mouth rinse during the school vacation periods is uncertain as no quantitative measures of compliance have been undertaken.

8.0 CONCLUSIONS

The aim of the research project is to evaluate a non-surgical intervention to control dental caries experience in disease susceptible children by modulation of the bacterial ecology on the tooth surface[8]. Results to date confirm that the project is on track to achieve this aim at both trial sites by demonstration of a decreasing trend of caries increment and a concomitant significant reduction in oral biofilm activity and Mutans streptococci counts after two mouth rinse cycles, particularly at the Vincent school. In addition, there appears to be no significant difference between the treatment and placebo mouth rinses in terms of clinical efficacy. However, the need to control the disease progression by continued antibacterial therapy prior

to and following surgical intervention is clearly required if a sustainable long term reduction of caries experience in young children is to be achieved.

9.0 RECOMMENDATIONS

It is recommended that the project continue without change to the current research protocol. Consideration may be given to dropping the maintenance rinse component next year due to the perceived poor compliance rate.

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