



BPS3061 BWX Sukin Body Wash Development Report

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1. Introduction

1.1 Objects

The following development report illustrates the formation of the Sukin body wash authorised by BWX Ltd. This body wash contains isethionate as active ingredient, and anionic surfactant system thickening with amphoteric surfactant.

1.2 Context

BWX is a global leading natural beauty and wellness business. The goal of BWX is to accelerate the natural revolution, creating amazing products and experiences to make customers feel better. There were two global offices, located in Melbourne, Australia and California, USA, leading six personal care categories, including Sukin, Andalou, Mineral Fusion, Nourished Life, Flora & Fauna and Go-To.

Sukin is consistent with the value of Australian made vegan products with natural ingredients, and cruelty free and carbon natural manufacturing process. Sukin defines natural and has become the No.1 natural skincare brand in Australian Pharmacy. Sukin made the choices to enhance the environmental stability by producing vegan products, using recyclable packaging, being carbon neutral and cruelty free. As one of the most popular body wash brands in Australia, Sukin Natural body washes aim to avoid using harmful ingredients, including sulphates, parabens, artificial colours and animal derivatives, but instead, using natural ingredients such as Leon, Tea Tree and Lime. Besides the natural ingredients, the affordable price also helps Sukin to become the first choice of Australian daily care.

Table 0: Table of factors

Name of Factor	API amount (Isethionate) (%)	pH adjuster (NaCl) amount (%)	Amphoteric surfactant amount
Level -1	4%	0%	15%
Level 0	5%	2.5%	17.5%
Level +1	6%	5%	20%

2. Formulation Development Study 1

2.1 Quality Target Product Profile

The quality target product profile (QTPP) summarises the properties of body wash, critical quality attributes (CQA), closure system and labelling.

Table 1. Quality Target Product Profile for BWX Sukin body wash, 200 g

QTPP Elements	Target	Justification
Dosage form	Solution	As required by the industrial partner
Dosage design	Colourless and viscous solution with no suspended solids	This general target is to enable appearance and to improve appeal of the product
Route of administration	Topical (skin and hair)	Topical is the most general and common route for cleansing skins
Dosage strength	4 - 6 % (of 200 g body wash formulation)	As required by the industrial partner
Pharmacokinetics	No epidermal diffusion	Topical use only and epidermal diffusion is undesirable
Stability	At least 2-4 years shelf life at room temperature under dark environment and in an unopened condition	Equivalence requirement as cosmetic product
Product Critical Quality Attribute	Physical appearance	Cosmetic industrial equivalence requirement and satisfaction of the specification by industrial partner
	Wetting time	
	Dirt dispersion	
	Cleaning capability	
	Percentage of solid content	
	Surface tension	
	Foaming ability	
	Foaming stability	
	pH	
	Viscosity	
Container closure system	Plastic bottle with pump or refillable package	Needed to achieve the target shelf-life and to ensure tablet integrity during shipping
Administration with labelling	Dosage form, direction of use, specific advice on storage, dispensing date, dispensing	Clear and precise label required
Alternative methods of administration	None	Exclusive administration route of tropic as body wash

2.2 Critical Quality Attributes

Critical quality attributes (CQA) were then identified based on the safety and efficacy aspects of the product.

Table 2. Critical Quality Attributes (CQAs) for BWX Sukin body wash, 200 g.

Quality Attributes of the Product		Target	Is this a CQA	Justification
Physical Attributes	Appearance	Colourless, viscous solution with no precipitation	Yes	Appearance is the major determining criteria for customers. Therefore, they are critical. The target should satisfy the requirement of the industrial partners.
	Odour	No unpleasant odour	Yes	Odour largely affects the customer's choice of the product; therefore, it is critical. And noticeable odour should be covered by the addition of fragrance. The target is set to ensure customers' acceptability.
Wetting time		NMT 3 sec	No	Wetting time is a measurement of body wash to maintain contact with a solid surface. It is not critical as viscosity is a more direct measurement.
Dirt dispersion (cleaning capability)		Meet the designed range	Yes	Dirt dispersion is the measure of cleaning capability of body wash. To meet the cleaning purpose, it is critical, and a designed range should be achieved.
Surface tension		Reduction in surface tension ranging from 31.68 to 38.72 dyn/cm	No	Only dramatic increase in tension that affects the production of cosmetic formulations such as body wash is caused by the comprehensive aggregation of active ingredients, so it is not critical.
Foaming ability and stability		Meet the designed range	Yes	Foaming ability and stability affect both cleaning capability and customers' experience. Therefore, it is critical.
Percentage of Solid content		NLT 14.9%	No	Solid content is not a critical parameter as it does not affect the cleaning ability of body wash and therefore the target is to lower than 14.9%
pH		4.5 to 7.5 will be ideal for skin and 5.5 for scalp	Yes	pH is a critical attribute as right pH prevents unwanted breakage, maintains moisture and natural oils.
Viscosity		NMT Absolute viscosity cP 3000	Yes	Viscosity is the major determining point for customers. Affecting appearance and wetting time. Therefore, it is critical.

2.3 Initial Risk Assessment of the Formulation Variables

Risk assessment was used throughout the manufacturing process to determine the potentially high-risk variables and their corresponding studies. The outcomes were summarised in the table below. For most responses, the goal is to achieve an appropriate value rather than a maximum or minimum, provided the value minimises the risk throughout the manufacturing, packing, transporting, storage and administration.

The levels of the risk were ranked as High, Medium and Low. High Risk means further investigation is needed whereas low risk means that no further investigation is needed.

Table 3. Overview of Relative Risk Ranking System

Low	The risk is acceptable. No further investigation needed.			
Medium	The risk is acceptable. Further investigation may be needed to reduce the risk.			
High	The risk is unacceptable. Further investigation is needed to reduce the risk.			

Formulation Factors	Product CQA				
	Physical appearance	Odour	Foaming stability	pH	Viscosity
API amount (%)	Medium	Low	Medium	Low	Medium
pH adjuster amount (%)	Low	Low	Medium	High	Medium
Amphoteric surfactant amount (%)	Low	Low	High	Low	High

Finally, design of experiments (DoE) was conducted to investigate the impact of isethionate, sodium chloride and amphoteric surfactant amounts (presented in percentage, %) on CQAs, and samples were stored in 4, 25 and 50 °C for two weeks to investigate the stability.

3. Alteration from Protocols

3.1 Viscometer Spindle

In the viscosity test where Brookfield DV1 Viscometer Spindle-05 was initially indicated to use in protocols, changed to Spindle-04 in the experiment due to its greater surface area and higher sensitivity resulting in improving torque (accuracy) in low spindle speed. Therefore, improving the accuracy of results by eliminating the effect of high spindle speed on the viscosity. However, Spindle-04 was used from week 1 hence the stability of the body wash may change hence the viscosity may be less accurate.

3.2 Storage Temperature

To do the stability analysis, the body wash needs to be kept at 4°C, 25°C (room temperature), and 50°C for 2 weeks and return to the 25°C to do the analysis. However, samples were kept in a 7°C fridge and room temperature were significantly lower than 25°C. Additionally, sample temperatures were not controlled at the same level during the measurements. Therefore, the accuracy and precision of tests can be significantly decreased.

4. Formulation Development Study 2

4.1 Factorial DoE method and Results

Each trial sample was equally divided into three samples, which were stored for 4 °C, 25 °C and 50 °C weeks respectively

4.1.1 Viscosity

	API amount (Isethionate) (%)	pH adjuster (NaCl) amount (%)	Amphoteric surfactant amount (%)	Viscosity(4, Week 0)	Viscosity(4, Week 2)	Viscosity(25, Week 0)	Viscosity(25, Week 2)	Viscosity(50, Week 0)	Viscosity(50, Week 2)
Level -1	4	0	15						
Level 0	5	2.5	17.5						
Level 1	6	5	20						
EXP1	-1	-1	-1	1584	2012	1498	1566	1584	1616
EXP2	1	-1	-1	1300	1034	711	729	1300	1400
EXP3	-1	1	-1	1450	4591	3690	3890	1450	2150
EXP4	1	1	-1	1510	7287	6800	6880	1510	1554
EXP5	-1	-1	1	3400	8949	9000	7192	3400	3450
EXP6	1	-1	1	2000	8199	8000	7872	2000	2480
EXP7	-1	1	1	3613	20993	18290	18298	3613	3651
EXP8	1	1	1	2355	16483	15900	15940	2355	2445
CP1	0	0	0	2800	23499	19000	19484	2800	3160
CP2	0	0	0	3098	20389	19010	20164	3098	3102
CP3	0	0	0	2890	15388	20001	21895	2890	2850

Figure 1. The DoE result table of Viscosity over 2 weeks at 4°C, 25°C and 50°C

4.1.2 pH

	API amount (Isethionate) (%)	pH adjuster (NaCl) amount (%)	Amphoteric surfactant amount (%)	pH (4, Week 0)	pH (4, Week 2)	pH (25, Week 0)	pH (25, Week 2)	pH (50, Week 0)	pH (50, Week 2)
Level -1	4	0	15						
Level 0	5	2.5	17.5						
Level 1	6	5	20						
EXP1	-1	-1	-1	6.26	6.26	6.47	6.46	6.33	6.33
EXP2	1	-1	-1	6.35	6.35	6.53	6.56	6.38	6.38
EXP3	-1	1	-1	6.37	6.37	6.08	6.08	6.37	6.37
EXP4	1	1	-1	5.99	5.93	6.25	6.22	5.9	5.92
EXP5	-1	-1	1	6.22	6.22	6.28	6.32	6.14	6.17
EXP6	1	-1	1	6.38	6.29	6.41	6.35	6.26	6.23
EXP7	-1	1	1	6.16	6.21	6.23	6.25	5.94	6.03
EXP8	1	1	1	6.02	6	6.15	6.13	5.9	5.81
CP1	0	0	0	6.3	6.1	6.4	6.38	6.1	6.17
CP2	0	0	0	6.02	6.28	6.42	6.34	6.02	6.03
CP3	0	0	0	5.97	6.43	6.39	6.37	6.02	6.02

Figure 2. The DoE result table of pH over 2 weeks at 4°C, 25°C and 50°C

4.1.3 Others

	API amount (Isethionate) (%)	pH adjuster (NaCl) amount (%)	Amphoteric surfactant amount (%)	Foam Stability (4, Week 0)	Foam Stability (4, Week 2)	Foam Stability (25, Week 0)	Foam Stability (25, Week 2)	Foam Stability (50, Week 0)	Foam Stability (50, Week 2)	Odour (4°C)	Odour (25°C)	Odour (50°C)
Level -1	4	0	15									
Level 0	5	2.5	17.5									
Level 1	6	5	20									
EXP1	-1	-1	-1	13.79	12.73	12.73	15.76	15.76	0	0	2.67	0.77
EXP2	1	-1	-1	1.64	1.62	1.23	1.21	6.92	1.64	0	0.72	1.43
EXP3	-1	1	-1	1.92	1.92	1.34	1.31	1.5	1.92	0	0	0.18
EXP4	1	1	-1	2.67	2.51	1.62	1.59	2.61	2.67	0.91	0.91	1.88
EXP5	-1	-1	1	2.33	2.31	2.31	2.34	2.16	2.33	15	14.955	1.82
EXP6	1	-1	1	1.67	1.65	1.47	1.2	1.24	1.67	4.05	4.05	5.16
EXP7	-1	1	1	1.87	1.86	1.74	1.21	1.58	1.87	3.36	3.36	6.25
EXP8	1	1	1	0.65	0.55	0.42	0.41	0.96	0.65	6.01	8.65	8.06
CP1	0	0	0	0	0	1.43	1.43	0	0	1.96	0	1.15
CP2	0	0	0	0	0	0.1	0	0.56	0	0.98	0	0.6
CP3	0	0	0	1.67	1.23	0	0	1.7	1.67	2.67	0	1.99

Figure 3. The DoE result table of Foam stability and odour over 2 weeks at 4°C, 25°C and 50°C

4.2 Viscosity

4.2.1 Evaluation

The raw data graph, also known as “song graphs”, of viscosity were developed after correctly transcribing data from a table of results, all data was accurate and relevant.

Song graphs with error bars were present in viscosity (cP) vs trial (Exp/CP) below, including week 0 and 2, and three different temperatures (4, 25 and 50°C) respectively. They were arranged in pairs with the same temperature, e.g., week 0 4°C and week 2 4°C, to investigate the effect of different storage conditions on viscosity in a two-week period.

The upper line graphs further demonstrated the significant factor or interaction on viscosity. There was 0 significant figure for viscosity.

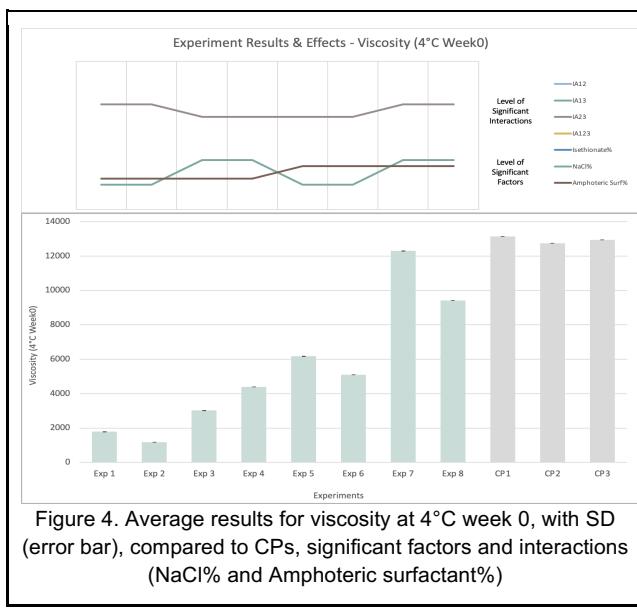


Figure 4. Average results for viscosity at 4°C week 0, with SD (error bar), compared to CPs, significant factors and interactions (NaCl% and Amphoteric surfactant%)

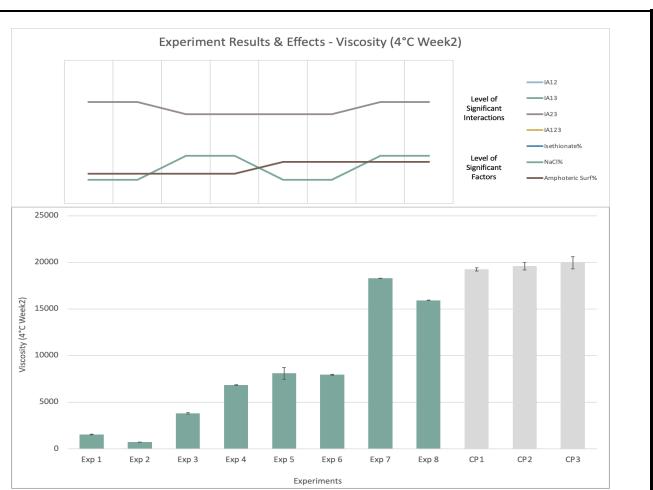


Figure 5. Results for viscosity at 4°C week 2, with SD (error bar), compared to CPs, significant factors and interactions (Amphoteric Surf% and NaCl%)

In the song graphs of both week 0 and 2 (4°C), NaCl% (Factor 2) and Amphoteric surfactant % (Factor 3) were significant factors, CPs were maintained in a consistent manner without presence of outlier.

When the level of Factor 2 (NaCl%) moved from -1 (Exp1+2, Exp5+6) to +1 (Exp3+4, Exp 7+8), the Response - viscosity increased, positively affecting the viscosity.

For Factor 3 (Amphoteric surfactant %) level increased from -1 to +1, viscosity increased, positively affecting the viscosity.

The similar significant factors indicated that the factors' effects on viscosity were stable but the large variations in viscosity (around 2000 cP) demonstrated that samples were unstable and changed significantly over time.

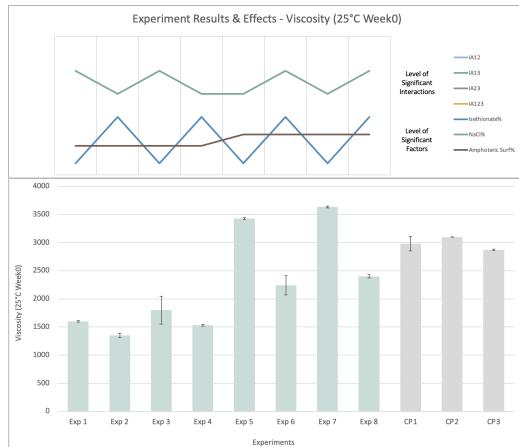


Figure 6. Average results for viscosity at 25°C week 0, with SD (error bar), compared to CPs, significant factors and interactions (Isethionate%, Amphoteric surfactant% and IA12)

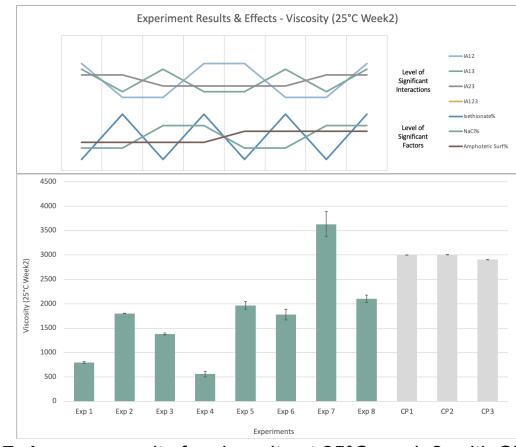


Figure 7. Average results for viscosity at 25°C week 2, with SD (error bar), compared to CPs, significant factors and interactions (Isethionate%, NaCl%, Amphoteric surfactant%, IA12 and IA13)

In the song graphs of both week 0 and 2 (25°C), isethionate% (Factor 1) and Amphoteric surfactant % (Factor 3) were significant factors, CPs were maintained in a consistent manner without presence of outlier. In week 0, viscosity at Exp1-4 was generally lower than Exp5-8, indicating that viscosity increased at Amphoteric surfactant level at +1, which was a positive effect. and viscosity decreased when isethionate% increased from -1 to +1, negatively affecting the response.

Interaction between those factors (IA12) also had a significant effect on viscosity, when it's at its highest level (+1), viscosity increased. In week 2, NaCl (Factor 2) became a significant factor which could not be directly read from the raw result graph due to the existence of IA12 and IA23. Similar to week 0, IA13 also significantly affects response in week 0.

The similar significant factors and interactions indicated that the factors' effects on viscosity were stable but NaCl% and IA13 effects became significant after 2 weeks. Additionally, the overall viscosity was stable except for Exp 1, 4, and 5.

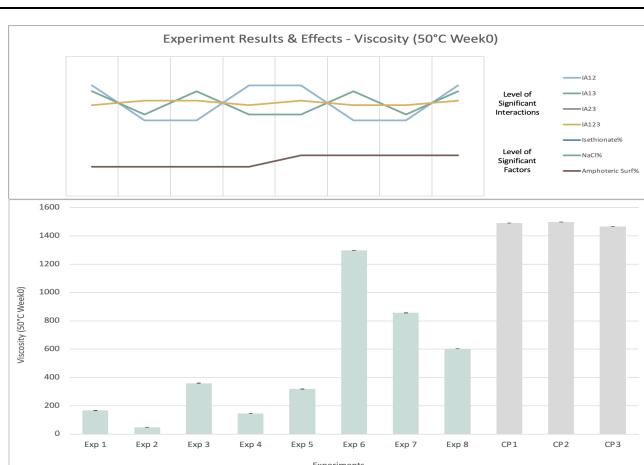


Figure 8. Average results for viscosity at 50°C week 0, with SD (error bar), compared to CPs, significant factors and interactions (Amphoteric surfactant%)

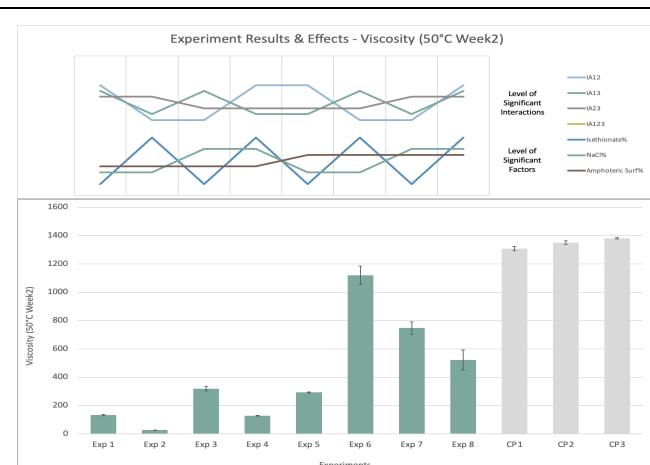


Figure 9. Average results for viscosity at 50°C week 2, with SD (error bar), compared to CPs, significant factors and interactions (Isethionate%, NaCl%)

In 50°C, amphoteric surfactant (Factor 3) was the only significant factor in week 0, while all three factors became significant in week 2. The raw result graphs were similar in both weeks, where Exp1-4 had a lower viscosity than Exp5-8, demonstrating the positive effect of amphoteric surfactant. In week 2, isethionate% (Factor 1) and NaCl (Factor 2) significantly negatively affected viscosity, viscosity decreased when isethionate and NaCl moved from higher level of +1.

The significant factors were completely different between these two groups viscosity. Demonstrated that the factors' effects on viscosity were not stable. However, the overall viscosity was stable with a minor difference hence samples were stable at 50°C over 2 weeks. In summary, different factors and interactions were detected to be significant under different temperatures implied that the temperature had a significant effect on samples' stability. However, at 50°C, samples were most stable in the viscosity aspect.

4.2.2 Analysis

4.2.2.1 Effect of factor

4.2.2.1.1 4°C

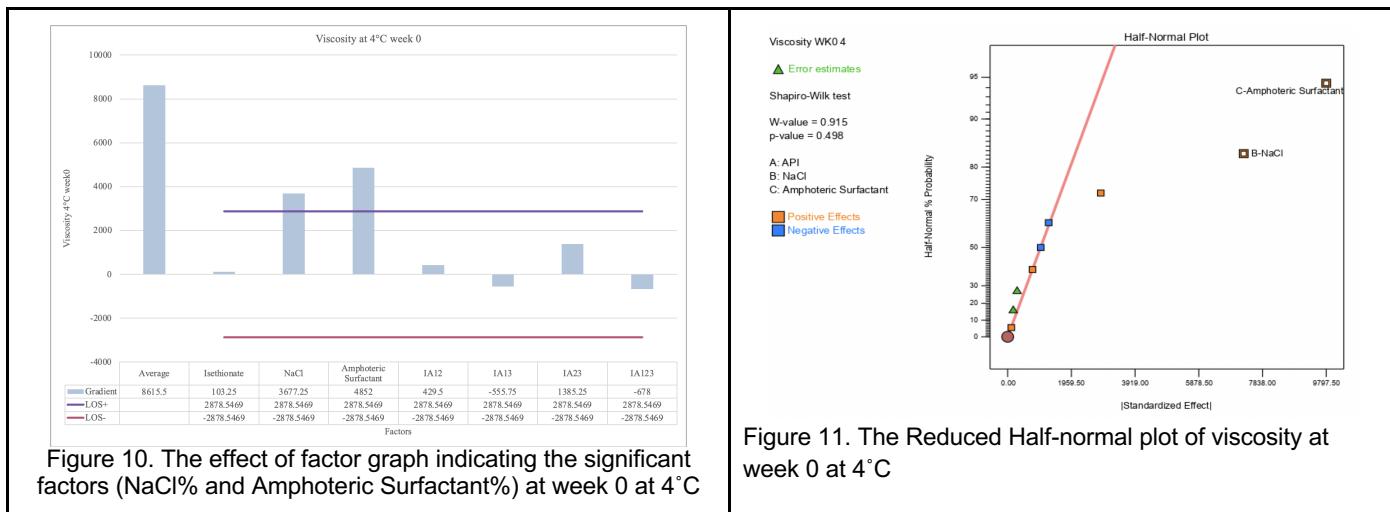


Figure 10. The effect of factor graph indicating the significant factors (NaCl% and Amphoteric Surfactant%) at week 0 at 4°C

In the week 0 4°C model, both NaCl% (Factor 2) and amphoteric surfactant% (Factor 3) were positively significant factors, having a positive effect on viscosity. As the Level of NaCl% increased from -1 to +1, viscosity increased by 7355 cP. Amphoteric surfactant% with similar effect, as its level increased from -1 to +1, viscosity increased by 9704 cP. To maximise the viscosity, higher levels of NaCl and Amphoteric surfactant were used. This result was further confirmed by the half-normal plot graph at the right, where both positive effect factors (labelled in orange colour), NaCl and Amphoteric surfactant were far away from the red line, indicating the magnitude of significance, and other factors layed on the red line. No significant interaction presented in the graph above.

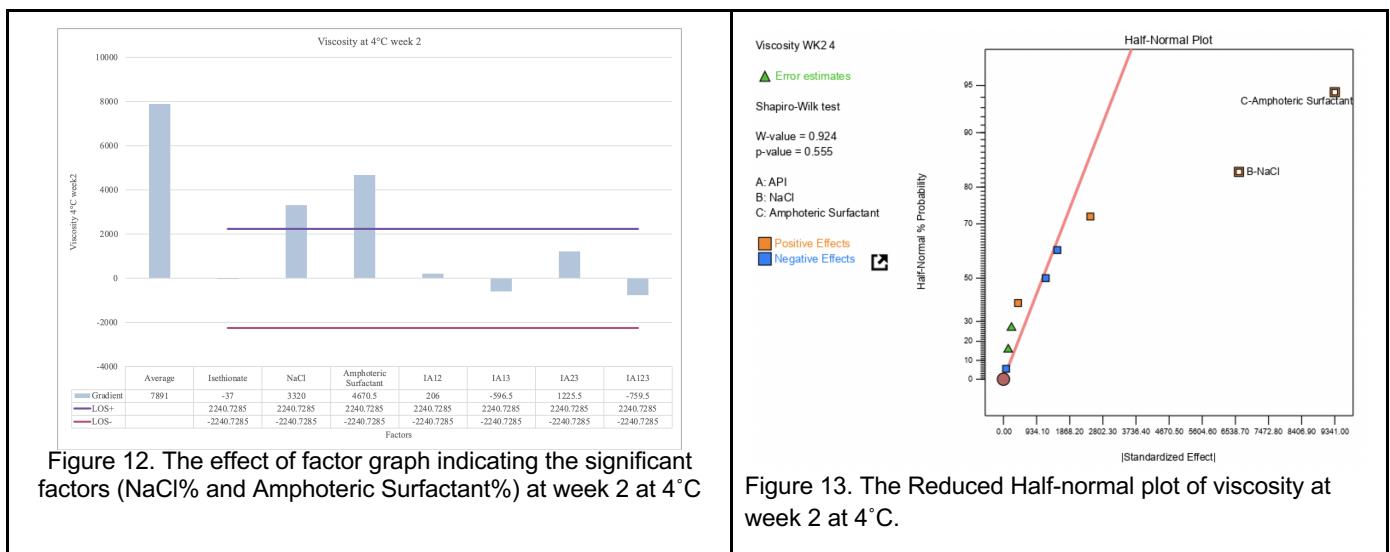
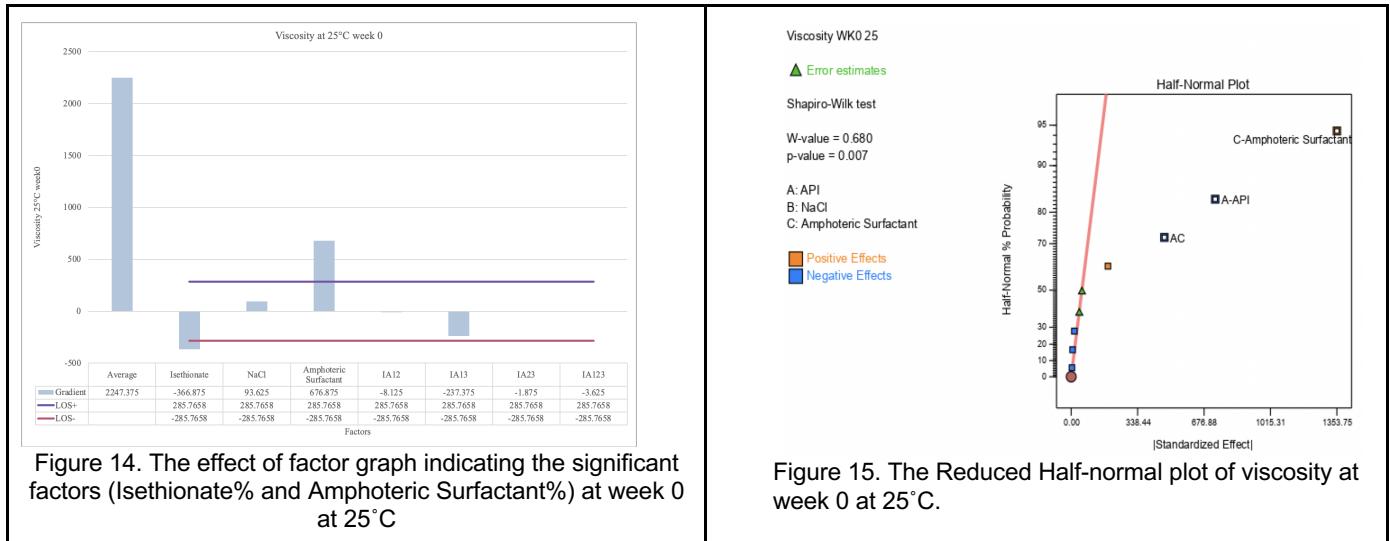


Figure 12. The effect of factor graph indicating the significant factors (NaCl% and Amphoteric Surfactant%) at week 2 at 4°C

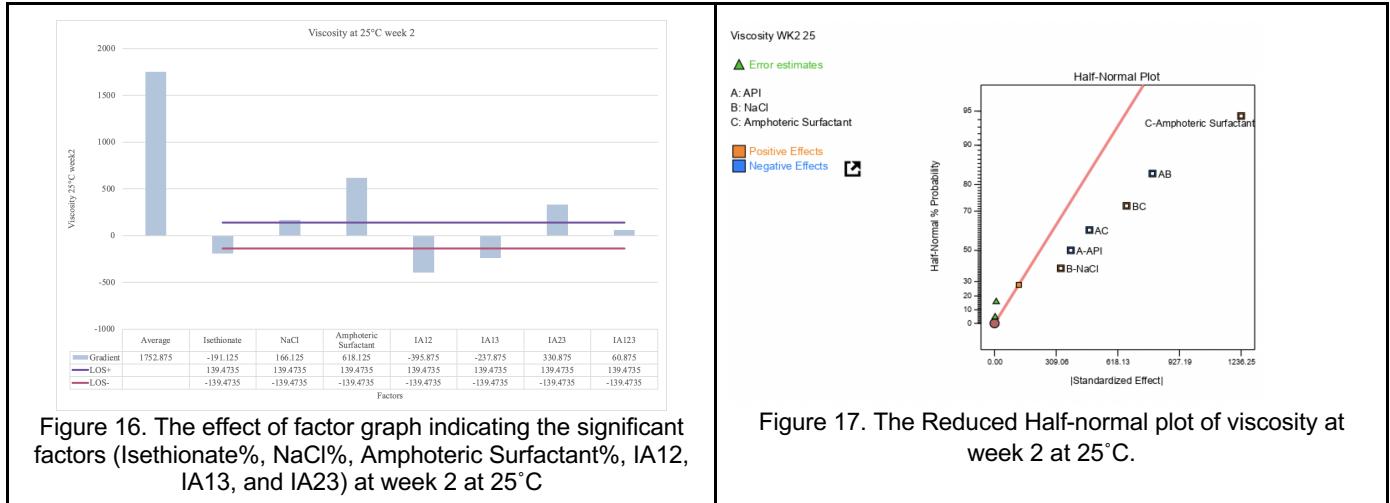
Similar to the week 0, NaCl% (Factor 2) and amphoteric surfactant% (Factor 3) were positively significant factors in the week 2 model, positively affecting the viscosity. As the Levels of NaCl% and Amphoteric surfactant% increased from -1 to +1, viscosity increased by 6640 cP and 9341 cP respectively. Half-normal further verified the results in the effect graph, where positive effects of NaCl and Amphoteric surfactant layed far away from the red line, corresponding to greater magnitude of significance.

4.2.2.1.2 25°C



In the week 0 25°C model, both isethionate% (Factor 1) and amphoteric surfactant% (Factor 3) were significant factors, having negative and positive effect on viscosity respectively. As the Level of isethionate% increased from -1 to +1, viscosity decreased by 734 cP. Amphoteric surfactant% with oppose effect, as its level increased from -1 to +1, viscosity increased by 1354 cP. To maximise the viscosity, lower level of isethionate and higher level of Amphoteric surfactant were used. This result was further confirmed by the half-normal plot graph at the right, where the negative effect factor (labelled in blue colour), isethionate, and positive effect factor (labelled in orange colour) - Amphoteric surfactant were far away from the red line, indicating the magnitude of significance, and other factors layed on the red line.

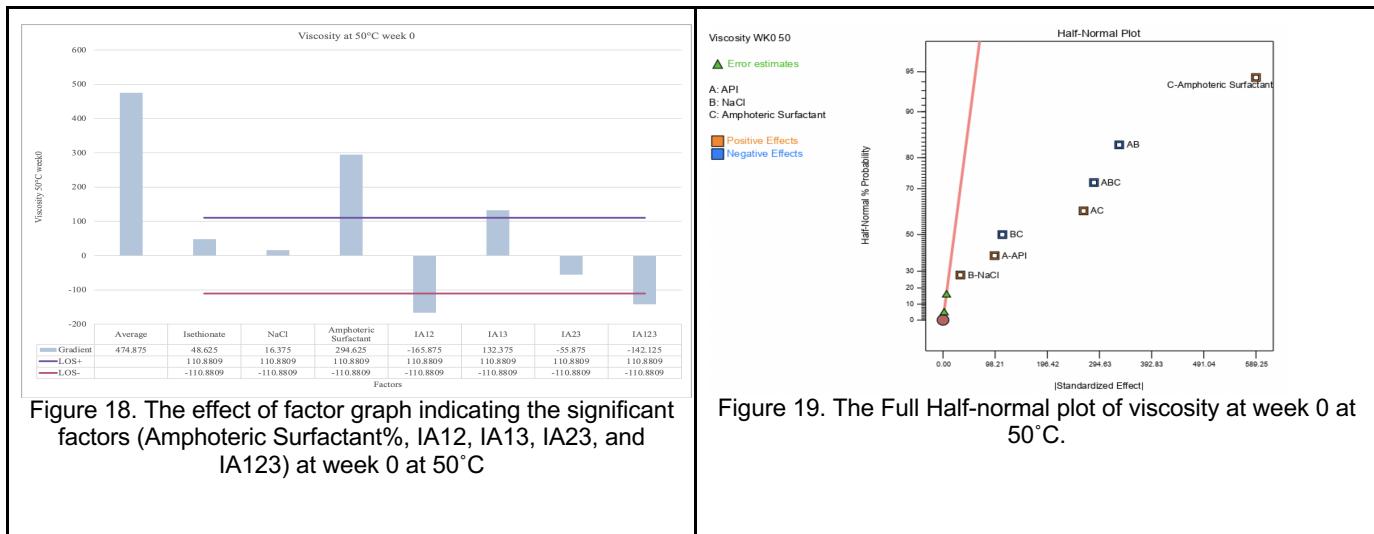
No significant interaction presented in the graph above.



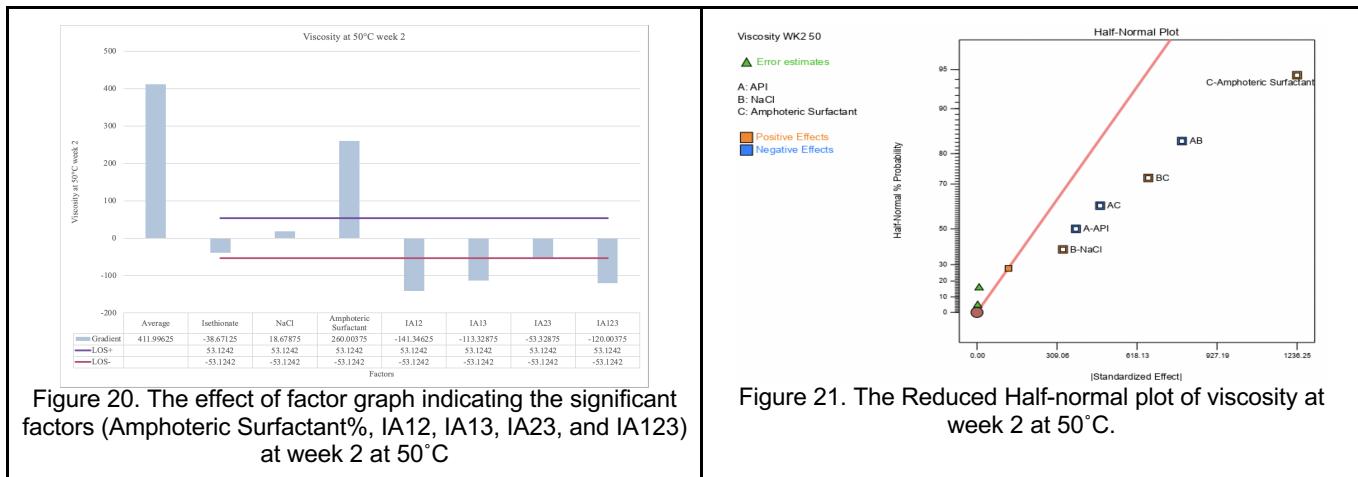
In week 2, significant factor isethionate% (Factor 1) and amphoteric surfactant% (Factor 3) behaved in the similar manner as week 0. As their levels increased from -1 to +1, viscosity decreased by 383 cP (significantly negative isethionate) and increased by 1236 cP (significantly positive amphoteric surfactant). Moreover, NaCl, IA12, IA13 and IA23 were significant factors in this model. NaCl and IA23 were positively affected, when their levels moved from -1 to +1, viscosity increased by 332 cP and 662 cP respectively. For the negative effects of IA12 and IA13, when their levels moved from -1 to +1, viscosity decreased by 792 cP and 476 cP respectively.

Half-normal plots present similar results, all significant factors layed on the right side of the graph with negative ones labelled with blue colour and positive ones labelled with orange colour.

4.2.2.1.3 50°C



In week 0 50°C model, amphoteric surfactant% (Factor 3) significantly increased viscosity by 589 cP when its level moved from -1 to +1, confirming by the half-normal plot as it layed further away from the red line. Both IA12 and 123 were significant interactions, having a negative effect on viscosity. When their levels increased from -1 to +1, viscosity decreased by 332 cP and 284 cP respectively. Oppositely, IA13 affected viscosity in a positive way, as its level increased from -1 to +1, viscosity increased by 265 cP. No reduced models can be applied due to all significant interactions related to all three factors, thus, non-significant factor NaCl was also depicted in the half-normal plot at the right.



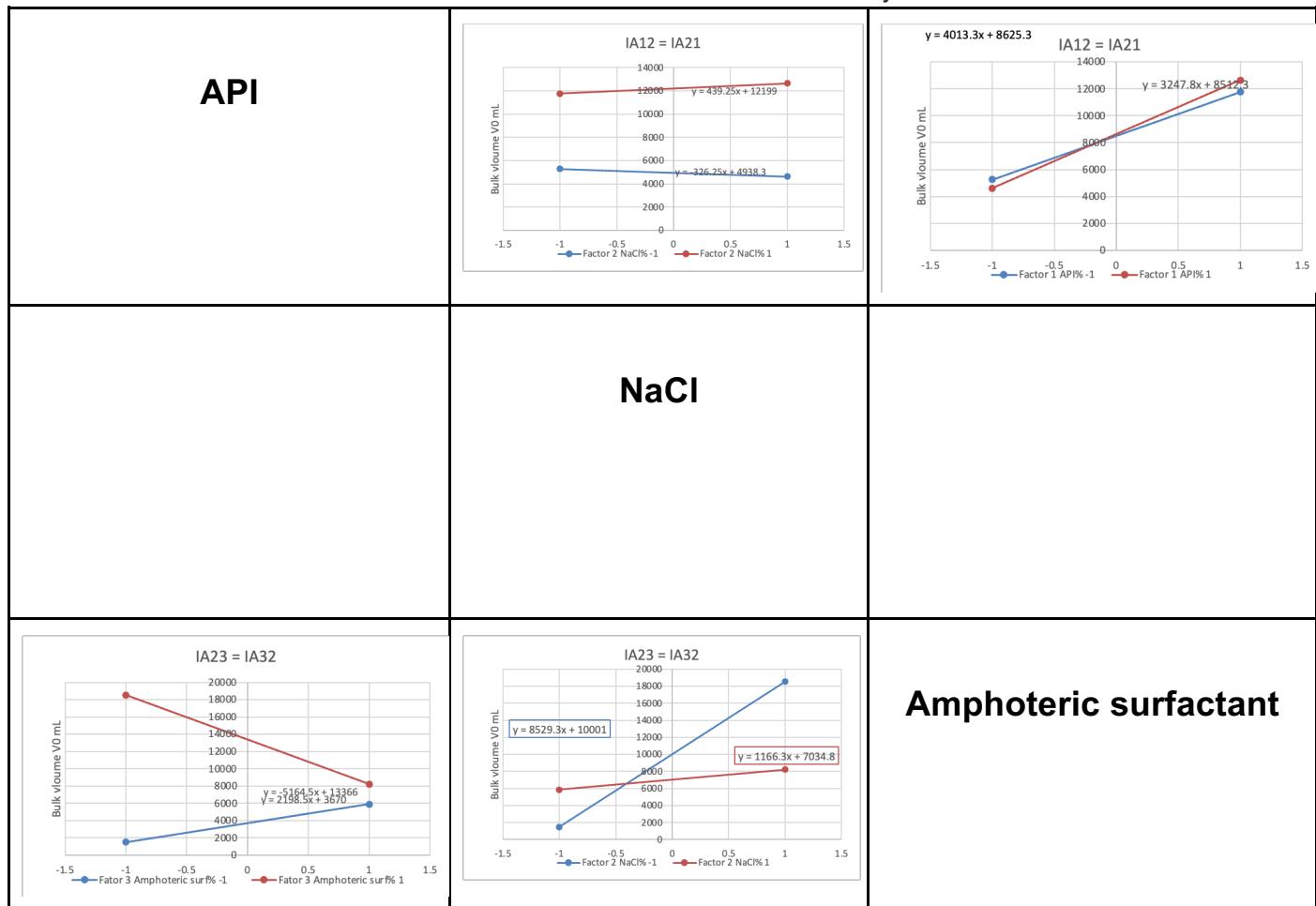
Week 2 50°C model was quite like the week 0 model while isethionate% (Factor 1) and IA13 had opposite effects to week 0 50°C models, these results were revealed in both gradient effect graph and half-normal plot.

4.2.2.2 Interactions

A detailed interaction analysis was performed in “effect of factor” section

4.2.2.2.1 Week 0 4°C

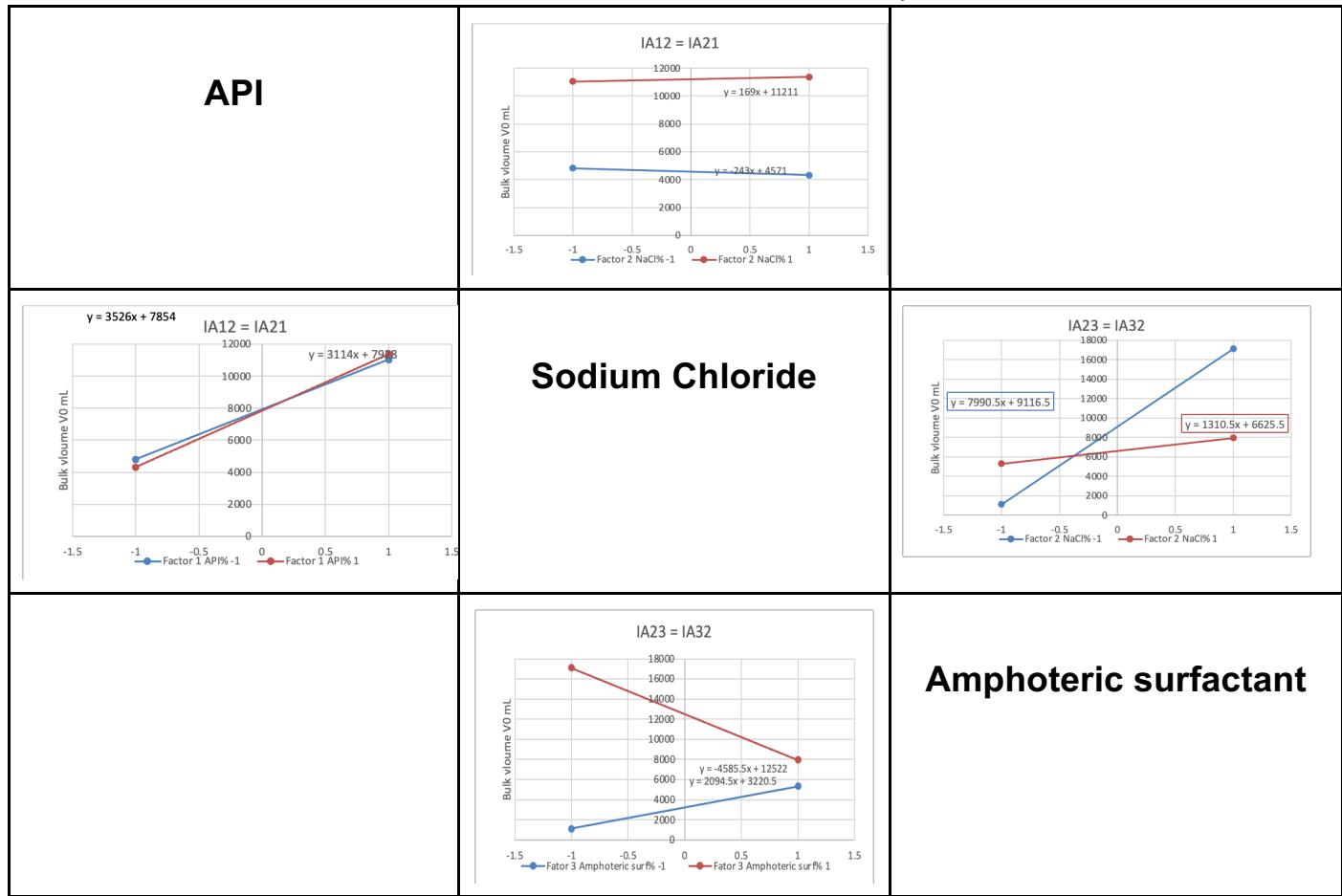
Table 4. The interactions between three factors in viscosity at week 0 at 4°C



In week 0 4°C, IA 12 and IA23 presented as there were interaction points between factor levels of -1 and +1, while none of them was significant.

4.2.2.2 Week 2 4°C

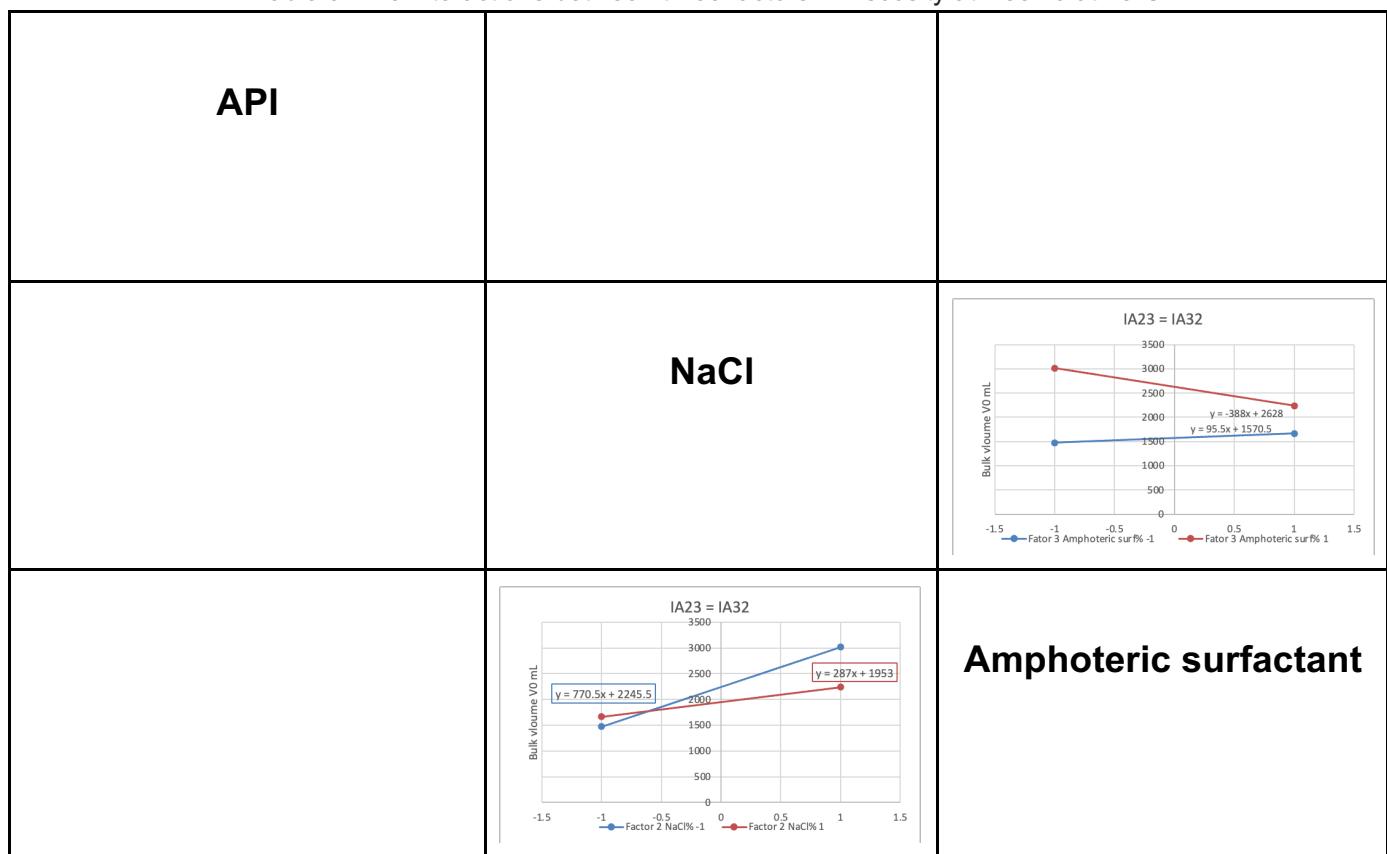
Table 5. The interactions between three factors in viscosity at week 2 at 4°C



Like week 0, non-significant IA12 and 13 presented in week 2, and no interaction between API and Amphoteric surfactant (IA23)

4.2.2.2.3 Week 0 25°C

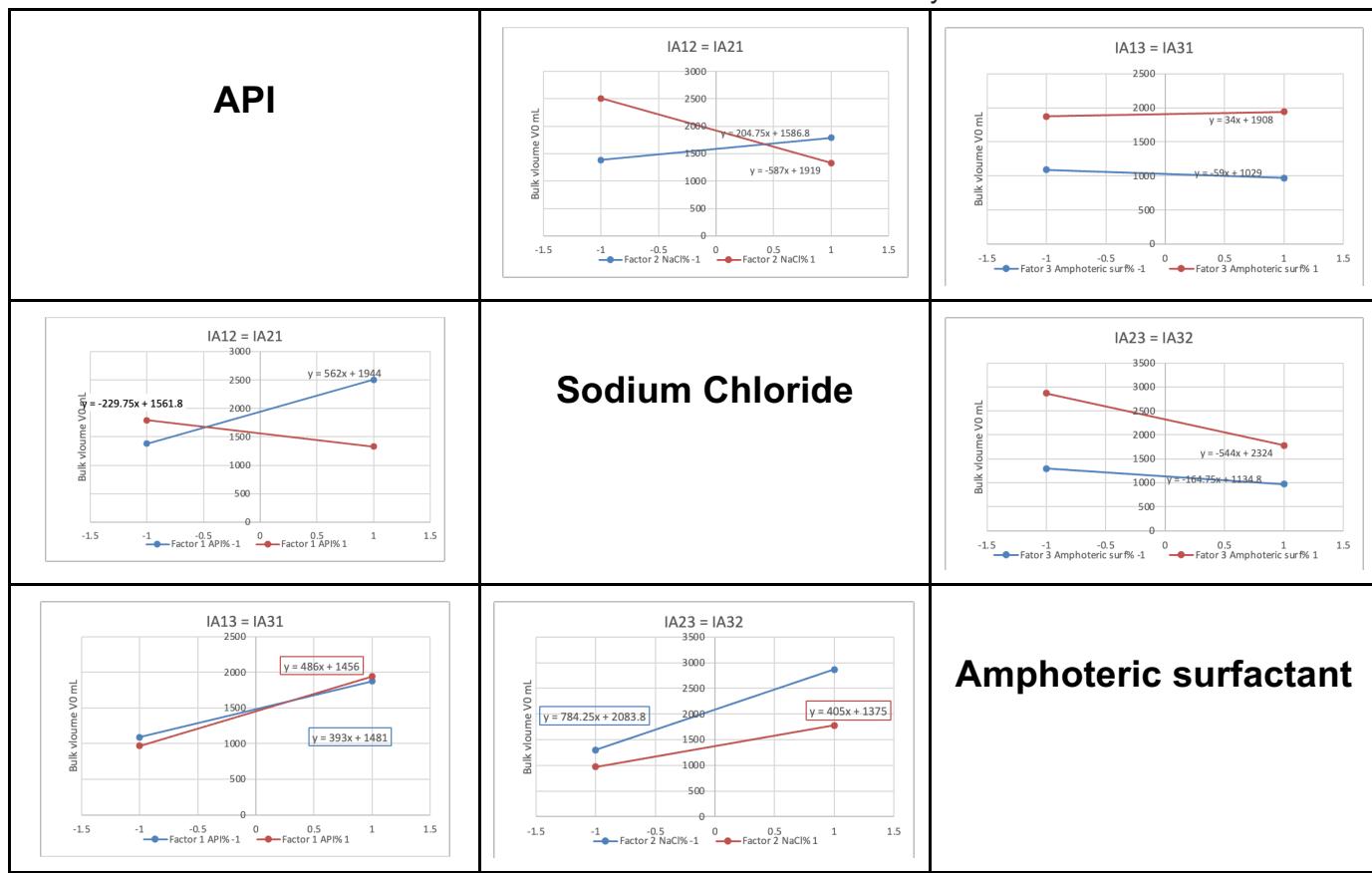
Table 6. The interactions between three factors in viscosity at week 0 at 25°C



No interaction between API (Factor 1) and NaCl (Factor 2) - IA12, or API and amphoteric surfactant - IA23 in week 0 25°C. IA13 existed and was a significant factor, they interacted at about a factor level of -0.55.

4.2.2.2.4 Week 2 25°C

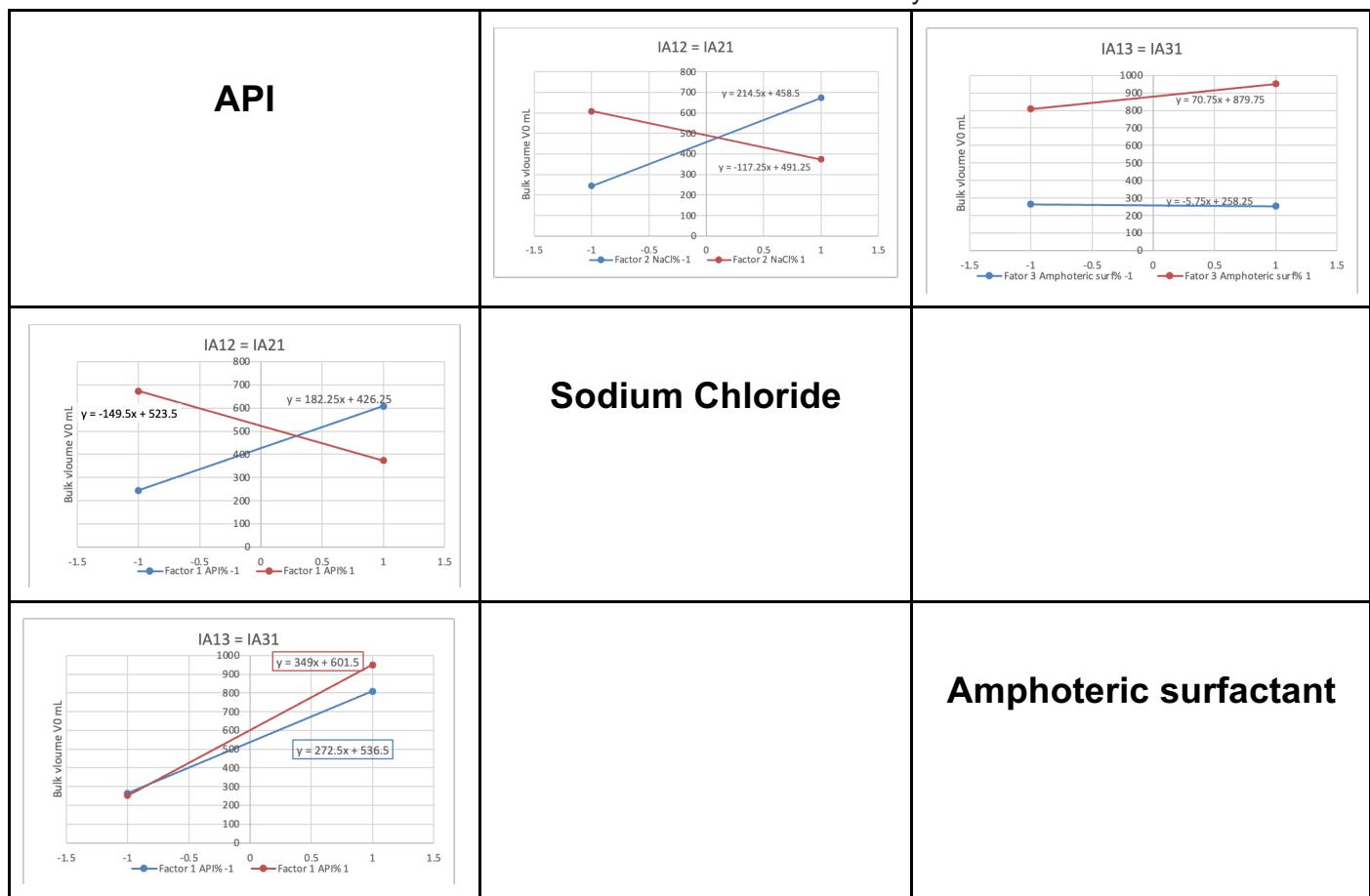
Table 7. The interactions between three factors in viscosity at week 2 at 25°C



No interaction between NaCl and surfactant (IA23), while IA12 and 13 significantly affected viscosity.

4.2.2.2.5 Week 0 50°C

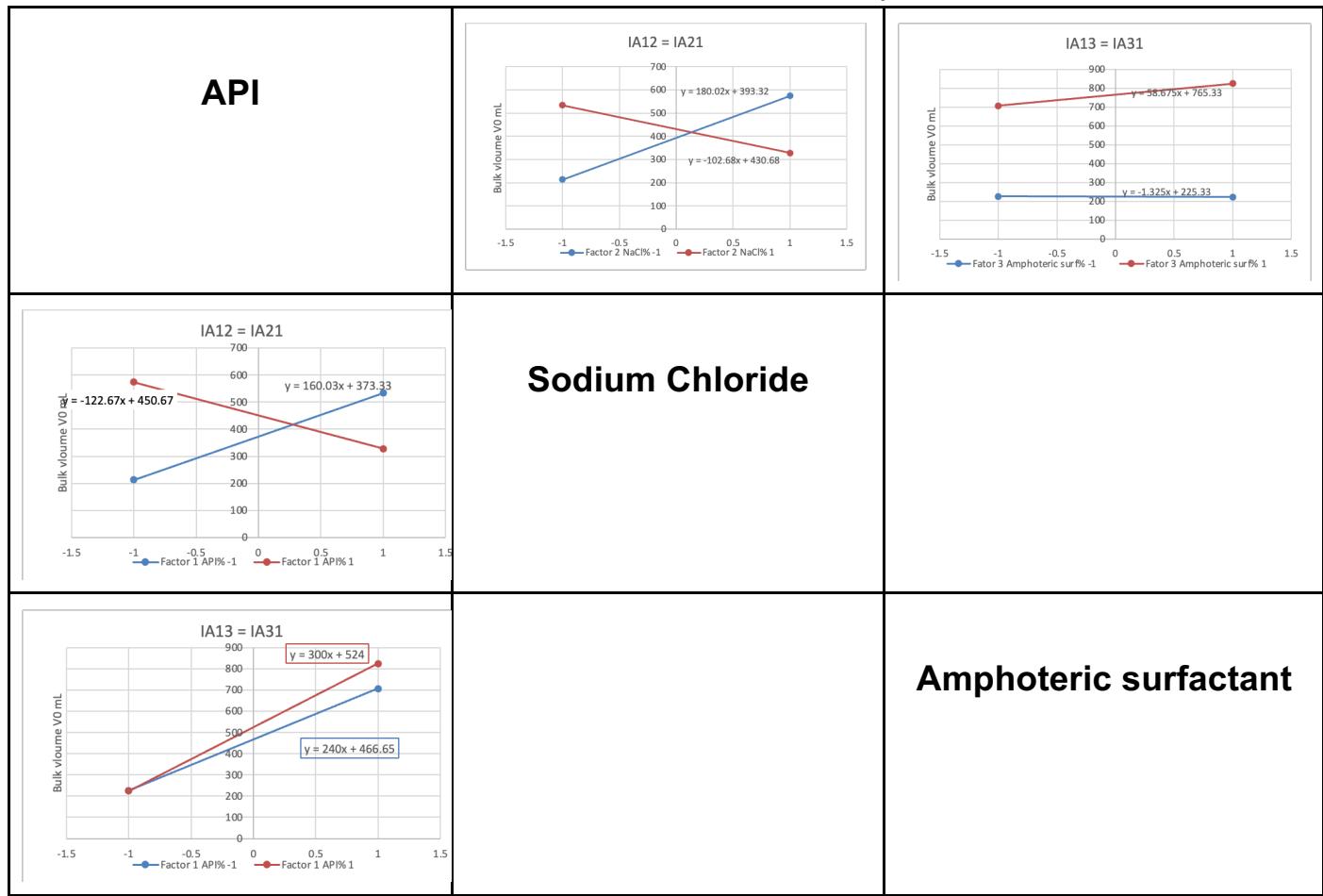
Table 8. The interactions between three factors in viscosity at week 0 at 50°C



No interaction between NaCl and surfactant as not point of interaction presented within -1 to +1, both IA12 and 13 were significant factors.

4.2.2.2.6 Week 2 50°C

Table 9. The interactions between three factors in viscosity at week 2 at 50°C



Similar results obtained from week 2, both IA12 and 13 were significant factors.

4.2.2.3 ANOVA

4.2.2.3.1 Reduced model for average results of viscosity at 4°C week 0 and week 2

Table 10. Reduced ANOVA model of Viscosity at week 0 and 2, 4°C.

Response 1: Viscosity WK0 4°C							Response 4: Viscosity WK2 4°C						
Source	Sum of Squares	df	Mean Square	F-value	p-value		Sum of Squares	df	Mean Square	F-value	p-value		
Model	2.97E+08	2	1.49E+08	40.75	0.0001	significant	2.63E+08	2	1.31E+08	42.86	0.0001	significant	
B-NaCl	1.05E+08	1	1.05E+08	28.89	0.001		8.82E+07	1	8.82E+07	28.77	0.001		
C-Amphoteric Surfactant	1.92E+08	1	1.92E+08	52.60	0.0002		1.75E+08	1	1.75E+08	56.94	0.0001		
Curvature	3.24E+08	1	3.24E+08	88.68	< 0.0001		3.16E+08	1	3.16E+08	103.11	< 0.0001		
Residual	2.56E+07	7	3.65E+06				2.15E+07	7	3.07E+06				
Lack of Fit	2.29E+07	5	4.57E+06	3.41	0.2424	not significant	1.98E+07	5	3.97E+06	4.87	0.1790	not significant	
Pure Error	2.69E+06	2	1.34E+06				1.63E+06	2	8.14E+05				
Cor Total	6.47E+08	10					6.00E+08	10					

The reduced 4°C viscosity models were significant as p-values <0.05. By eliminating insignificant factors (Factor 1 - isethionate%) and interactions, lack of fit (LOF) were not significant due to p-values of 0.2424 and 0.179 (both > 0.05). The p-values of curvature < 0.001 determined the significant curvatures existed, resulting in unpredictability of the reduced models.

4.2.2.3.2 Reduced model for average results of viscosity at 25°C week 0 and week 2

Table 11. Reduced ANOVA model of Viscosity at week 0 and 2, 25°C.

Response 2: Viscosity WK0 25						Response 5: Viscosity WK2 25							
Source	Sum of Squares	df	Mean Square	F-value	p-value	Source	Sum of Squares	df	Mean Square	F-value	p-value		
Model	5.19E+06	3	1.73E+06	106.79	< 0.0001	significant	Model	6.15E+06	6	1.03E+06	85.56	0.0019	significant
A-API	1.08E+06	1	1.08E+06	66.43	0.0002		A-API	2.92E+05	1	2.92E+05	24.39	0.0159	
C-Amphoteric Surfactant	3.67E+06	1	3.67E+06	226.13	< 0.0001		B-NaCl	2.21E+05	1	2.21E+05	18.42	0.0233	
AC	4.51E+05	1	4.51E+05	27.81	0.0019		C-Amphoteric Surfactant	3.06E+06	1	3.06E+06	255.07	0.0005	
Curvature	1.18E+06	1	1.18E+06	72.91	0.0001		AB	1.25E+06	1	1.25E+06	104.62	0.0020	
Residual	97253.17	6	16208.86				AC	4.53E+05	1	4.53E+05	37.77	0.0087	
Lack of Fit	70786.5	4	17696.63	1.34	0.4702	not significant	BC	8.76E+05	1	8.76E+05	73.09	0.0034	
Pure Error	26466.67	2	13233.33				Curvature	3.24E+06	1	3.24E+06	270.45	0.0005	
Cor Total	6.47E+06	10					Residual	35950.79	3	11983.6			
							Lack of Fit	29646.12	1	29646.12	9.40	0.0919	not significant
							Pure Error	6304.67	2	3152.33			
							Cor Total	9.43E+06	10				

Again, in the reduced 25°C viscosity models, both p-values <0.05, indicating the significance of models. By eliminating insignificant factors (Factor 2 - NaCl%) and all interactions except IA13 in week 0, and IA123 in week 2, resulted in not significant lack of fit ($p > 0.05$), significant curvature ($P < 0.01$), demonstrating that 25°C models were unpredictable.

4.2.2.3.3 Full model for average results of viscosity at 50°C week 0 and week 2

Table 12. Full ANOVA model of Viscosity at week 0 and 2,50°C.

Response 3: Viscosity WK0 50°C							Response 6: Viscosity WK2 50°C						
Source	Sum of Squares	df	Mean Square	F-value	p-value		Sum of Squares	df	Mean Square	F-value	p-value		
Model	1.26E+06	7	1.80E+05	90.52	0.011	significant	9.56E+05	7	1.37E+05	298.66	0.0033	significant	
A-API	18915.13	1	18915.13	9.49	0.0912		1.20E+04	1	1.20E+04	26.16	0.0362		
B-NaCl	2145.12	1	2145.12	1.08	0.4084		2.79E+03	1	2.79E+03	6.10	0.1321		
C-Amphoteric Surfactant	6.94E+05	1	6.94E+05	348.55	0.0029		5.41E+05	1	5.41E+05	1182.54	0.0008		
AB	2.20E+05	1	2.20E+05	110.48	0.0089		1.60E+05	1	1.60E+05	349.48	0.0028		
AC	1.40E+05	1	1.40E+05	70.36	0.0139		1.03E+05	1	1.03E+05	224.67	0.0044		
BC	24976.12	1	24976.12	12.54	0.0713		22751.64	1	22751.64	49.75	0.0195		
ABC	1.62E+05	1	1.62E+05	81.11	0.0121		1.15E+05	1	1.15E+05	251.91	0.0039		
Curvature	2.11E+06	1	2.11E+06	1059.18	0.0009		1.84E+06	1	1.84E+06	4032.14	0.0002		
Pure Error	3984.67	2	1992.33				9.15E+02	2	457.33				
Cor Total	3.38E+06	10					2.80E+06	10					

In the full ANOVA Table 14 above, both 50°C viscosity models were significant as model ANOVA p-values < 0.05. In week 0, single significant factor of Amphoteric surfactant% (F3) presented due to p-value of 0.0029 (p < 0.05), and significant interactions, IA12, IA13, IA123 present indicated by their p-values (p < 0.05), curvature with a p-value of 0.009 revealed the non-linearity and unpredictability of this models. Non-significant factor 2 (NaCl%) contributed to the significant interaction, therefore, no reduced model could be used. In week 2, both API% (F1) and Amphoteric surfactant% (F3) were significant (p = 0.0362 and 0.0008 < 0.05), and all interactions were significant, no reduced models could be applied either in 50°C. Significant curvature led to the non-linear property, as well as unpredictability of those models.

4.2.2.4 Interpretation

Based on all reduced ANOVA tables, the amphoteric surfactant was a significant factor and has a positive effect on the viscosity at all 4°C, 25°C and 50°C conditions. The amphoteric surfactant helped to screen the charge carried by the anionic surfactant (isethionate) at the air interface, leading to closer surfactant packing. The closer packing of surfactant molecules at the interface will cause the increase in the viscosity (Stephanie K. Clendennen, Neil W. Boaz, 2019).

At 4°C, the sodium chloride showed a significant and positive effect on the viscosity because it can reduce the micelle charge density, resulting in the conversion of spherical micelles to rod-shaped micelles, thus the viscosity can be increased (George Deckner. 2015). But the effect of sodium chloride on the viscosity will be reduced with increased temperature, which was related to the suspended particles being hard to be stabilised due to faster Brownian motion. Sometimes, when the body wash was placed at room temperature for two weeks, the sodium chloride became significant. The suspended particles may become easier to be stabilised because of a lone time stand still. Conclusively, that can explain why the sodium chloride becomes not significant with the temperature increase.

Theoretically, the isethionate shows a positive effect on the viscosity, which meant the viscosity should grow rapidly with surfactant concentration due to the considerable cross links among micelles and polymers (transient network) (Wang SC, Wei TC, Chen WB, Tsao HK. 2004). However, from the analysed result, the isethionate showed a negative effect on the viscosity. The isethionate was initially added to a calculated quantity of water over 80°C in the manufacturing process, but it was found the solubility of isethionate was unfavourable in terms of enthalpy of solvation (Sun JZ, Parr JW, Erickson MC. 2003). The insufficient solving of isethionate was possible to affect the result of viscosity that had not reached the desired value, which can result in a wrong determination of the effect of isethionate on viscosity.

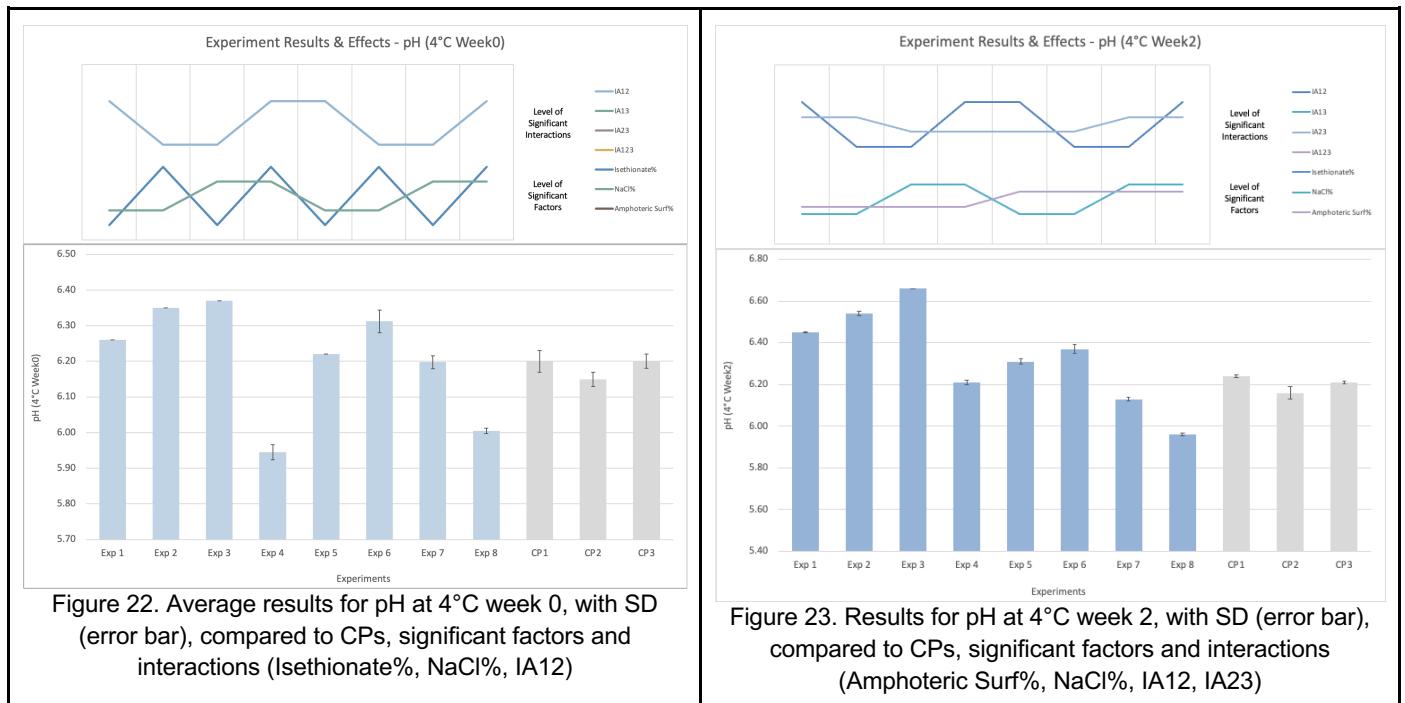
4.2.2.5 Conclusion

In conclusion, the amphoteric surfactant always showed a significant and positive effect on the viscosity because of its physicochemical properties. The sodium chloride only showed a significant and positive effect when the body wash is placed in 4°C. With the temperature increase, the significance of the effect of sodium chloride depends on the micro change in the body wash. The effect of isethionate on the viscosity is not predictable as the result was totally opposite to the ideal one probably due to the insufficient solving of isethionate.

4.3 pH

4.3.1 Evaluation

The graphs of raw data of pH were presented in pH VS trials (Experiment/CP) below with the standard deviation showing at the top of each column (error bar) for both week 0 and week 2 with three different temperatures (4°C, 25°C, 50°C). The “song graph” only showed the trend of significant factors or interactions and was developed after correctly transferring data from the table of results, all data is accurate and relevant. The graphs were arranged in pairs to determine and compare the effect of factors under different temperature conditions.



From the song graphs of pH at 4°C, no outliers were observed among CPs for both week0 and week2. The significant factors for week 0 were Isethionate%, NaCl%, and IA12. Amphoteric Surf%, NaCl%, IA12, IA23 had significant effect on pH for week2.

Different factors and interactions were determined to have significant effects on the pH at 4°C. Most of the trials illustrated a stable pH over 2 weeks except Exp 4.

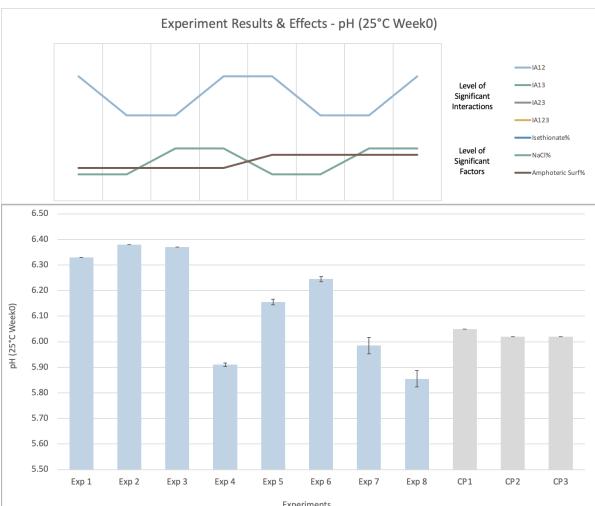


Figure 24. Results for pH at 25°C week 0, with SD (error bar), compared to CPs, significant factors and interactions (NaCl%, Amphoteric Surf%, IA12)

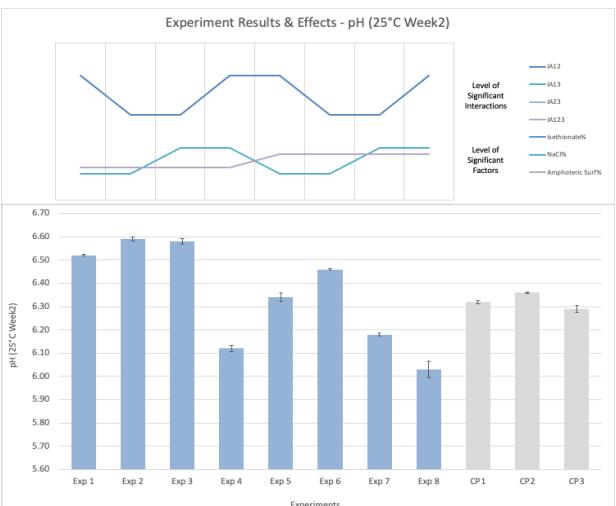


Figure 25. Results for pH at 25°C week 2, with SD (error bar), compared to CPs, significant factors and interactions (Amphoteric Surf%, NaCl%, IA12)

As shown in the song graph at pH 25°C, NaCl%, Amphoteric Surf% and IA12 were significant factors on pH for both week 0 and week 2. CPs remained consistent without outliers appearing. Same factors and interactions were determined to have significant effects on the pH at 25°C hence factors' effects were stable under 25°C. The overall pH was constant over 2 weeks hence samples were stable at 25°C in the pH aspect.

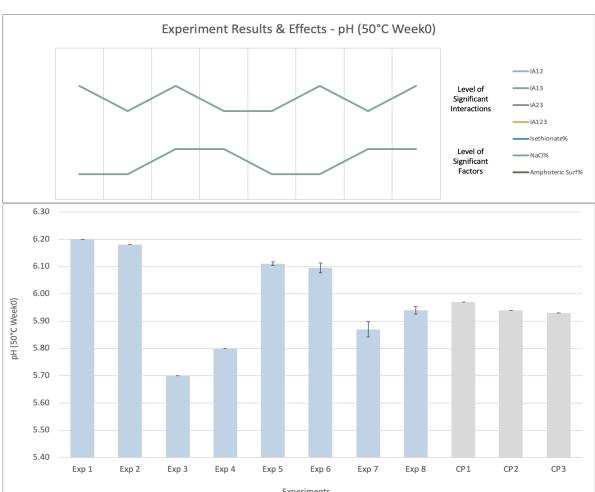


Figure 26. Results for pH at 50°C week 0, with SD (error bar), compared to CPs, significant factors and interactions (NaCl%, IA13)

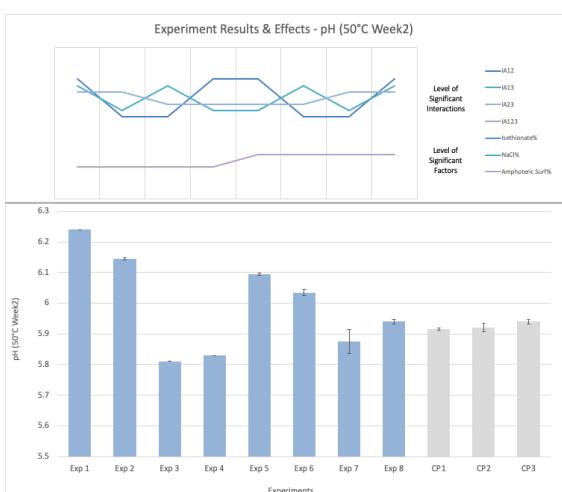


Figure 27. Results for pH at 50°C week 2, with SD (error bar), compared to CPs, significant factors and interactions (Amphoteric Surf%, IA12, IA13, IA23)

For pH at 50°C week 0, NaCl% and IA13 were the significant factors on pH. For pH at 50°C week 2, Amphoteric Surf%, IA12, IA13, IA23 were the significant factors on pH. No outliers between CPs were observed.

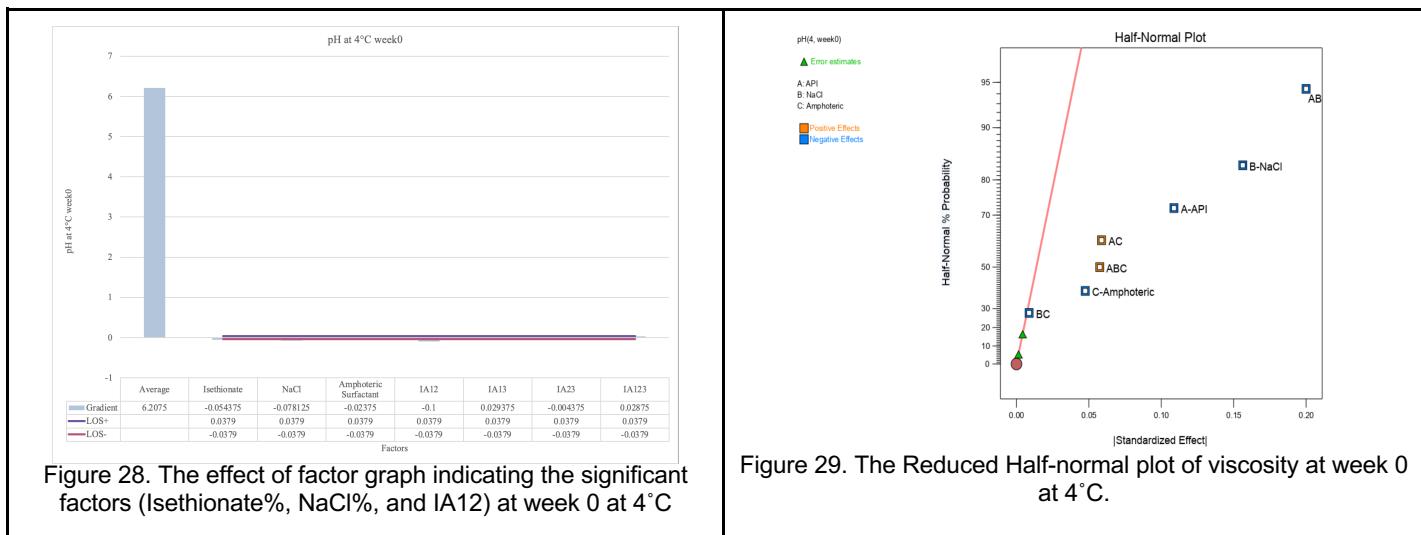
Different factors and interactions were determined to have significant effects on the pH at 50°C hence samples were viewed as unstable. Most of the trials illustrated a stable pH over 2 weeks except Exp 4.

In summary, samples' pH was considered most stable at 25°C over 2 weeks. Similar factors and interactions were observed having significant effects hence can be concluded that amphoteric surfactant%, NaCl% and IA12 significantly affected the pH.

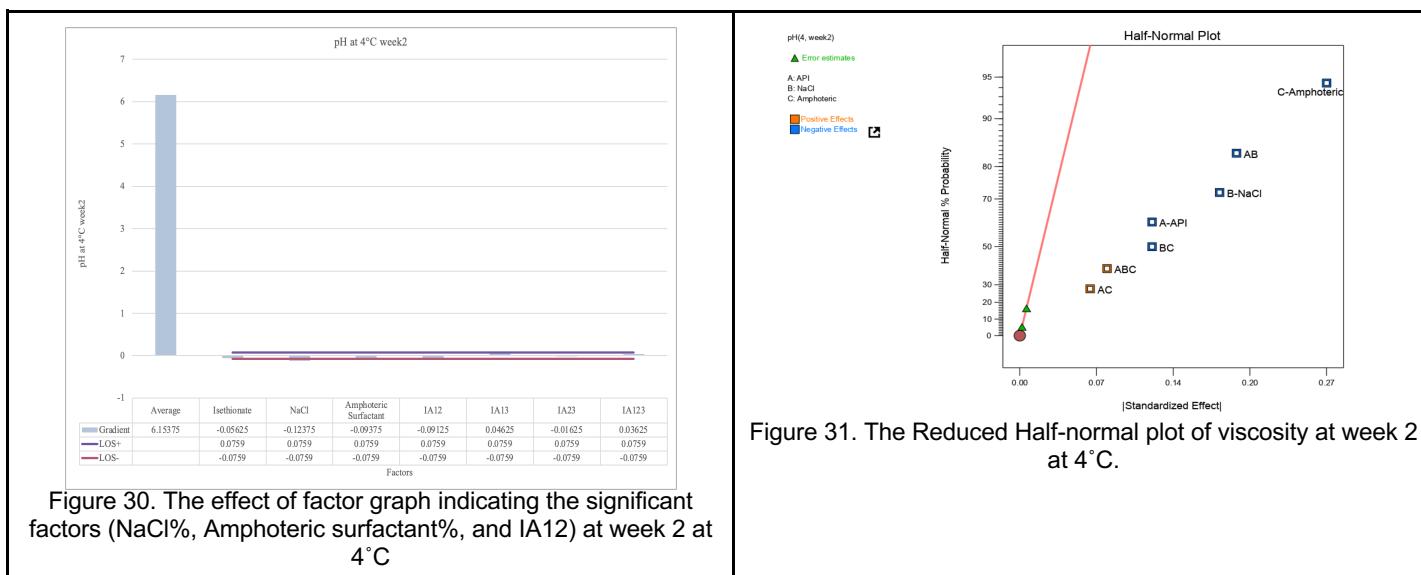
4.3.2 Analysis

4.3.2.1 Effect of factor

4.3.2.1.1 4°C

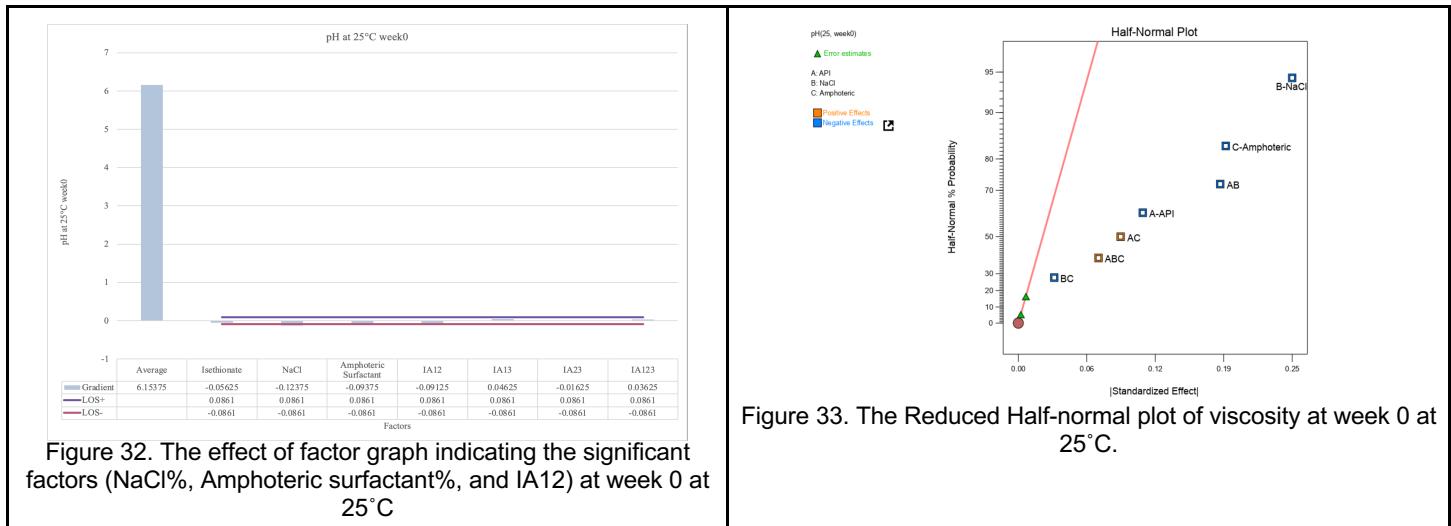


In the week 0 4°C model, the Isethionate% (factor1), NaCl% (factor2), and IA12 were the significant factors on pH as shown in the graph above. Isethionate% had a negative effect that when it changed from 4% (LEVEL-1) to 6% (LEVEL+1), pH decreased by 0.1. Similar for NaCl% and IA12, as NaCl increased from 0% to 5%, pH reduced by 0.16, as IA12 increased, pH reduced by 0.2. The half normal plot showed similar results that AB, B, A were posited away from the pink line. As pH was not the focus parameter for this product, and it was in between the pH range suggested by the company, the pH value was not necessarily to be maximised or minimised.

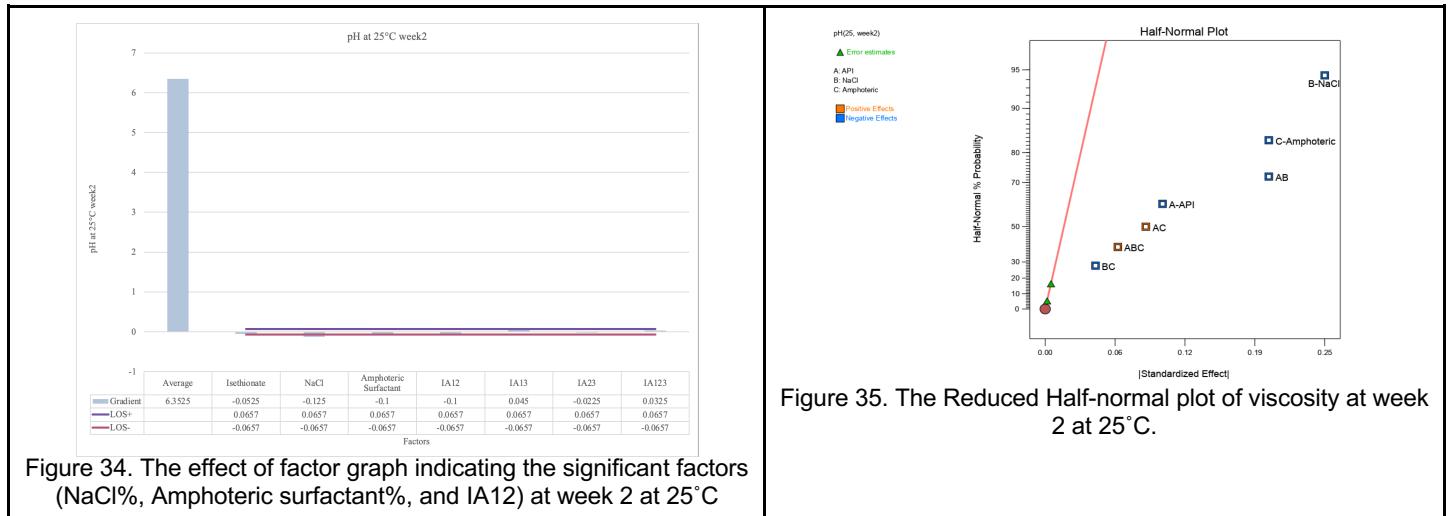


In the model of pH 4°C week2, NaCl%, Amphoteric surfactant% and IA12 had negative effects which when these factors moved from LEVEL-1 (0%, 15%) to level+1(5%, 20%), pH decreased by 0.12, 0.09, and 0.09 respectively. It was further proved by the half normal plot that these factors were far away from the pink line.

4.3.2.1.2 25°C

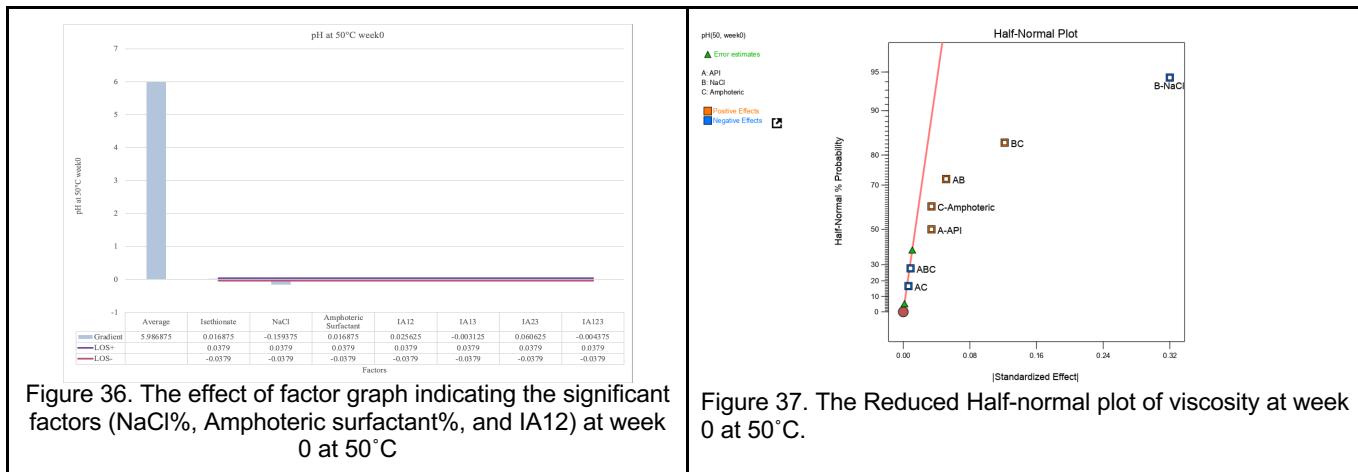


For model pH 25°C week 0, based on the graph of effect and half normal plot, the significant factors for pH were NaCl%, amphoteric surfactant% and IA12. NaCl had a negative effect that when it moved from 0% to 5%, pH decreased by 0.24. Similar for amphoteric surfactant and IA12, when they moved from level-1 to level +1, pH decreased by 0.18 and 0.18 respectively.

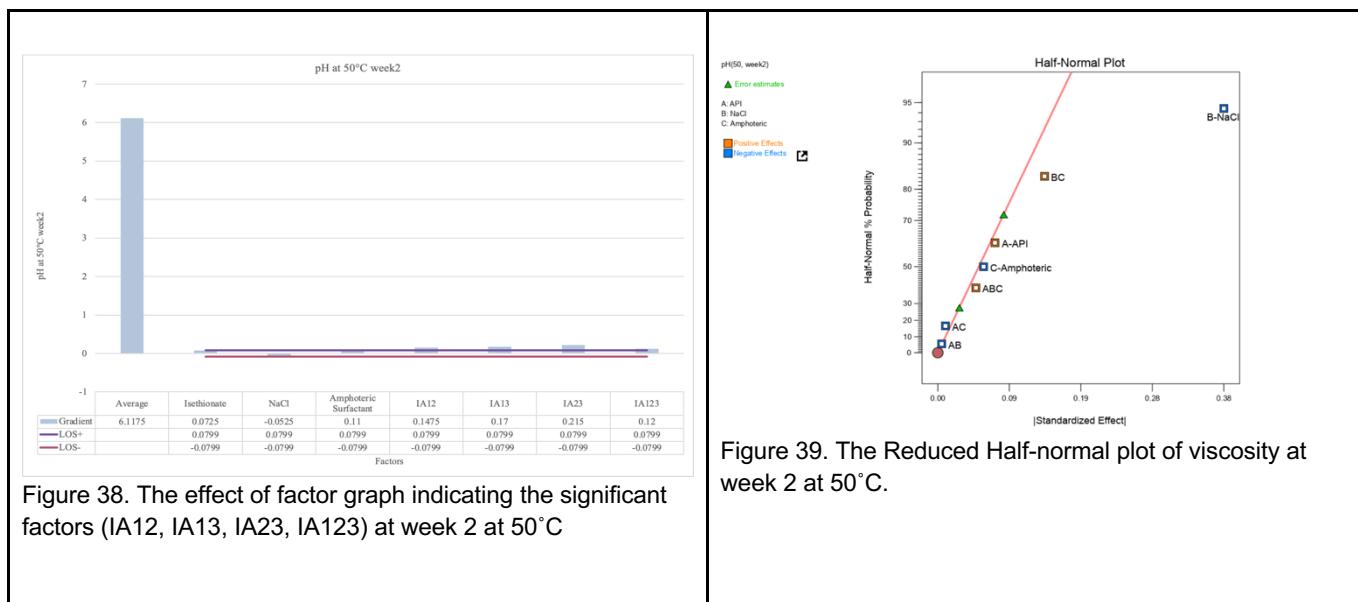


Similar for week 2, the significant factors were again NaCl%, amphoteric surfactant% and IA12. As NaCl moved from 0% to 5%, pH decreased by 0.26. Similar for amphoteric surfactant and IA12, when they moved from level-1 to level +1, pH decreased by 0.2 and 0.2 respectively. In the half normal plot, B, C,AB were shown in blue, indicating that they had a negative effect on pH.

4.3.2.1.3 50°C



According to the graph of effects at the left, NaCl had the most significant negative effect on pH that as it changed from 0% to 5%, pH decreases by 0.3, it was then confirmed by the half normal plot where factor 2 was very far from the pink line. The interaction between NaCl and amphoteric surfactant was also a significant factor in this model. When IA23 changed from LEVEL-1 to LEVEL+1, pH decreased by 0.12.



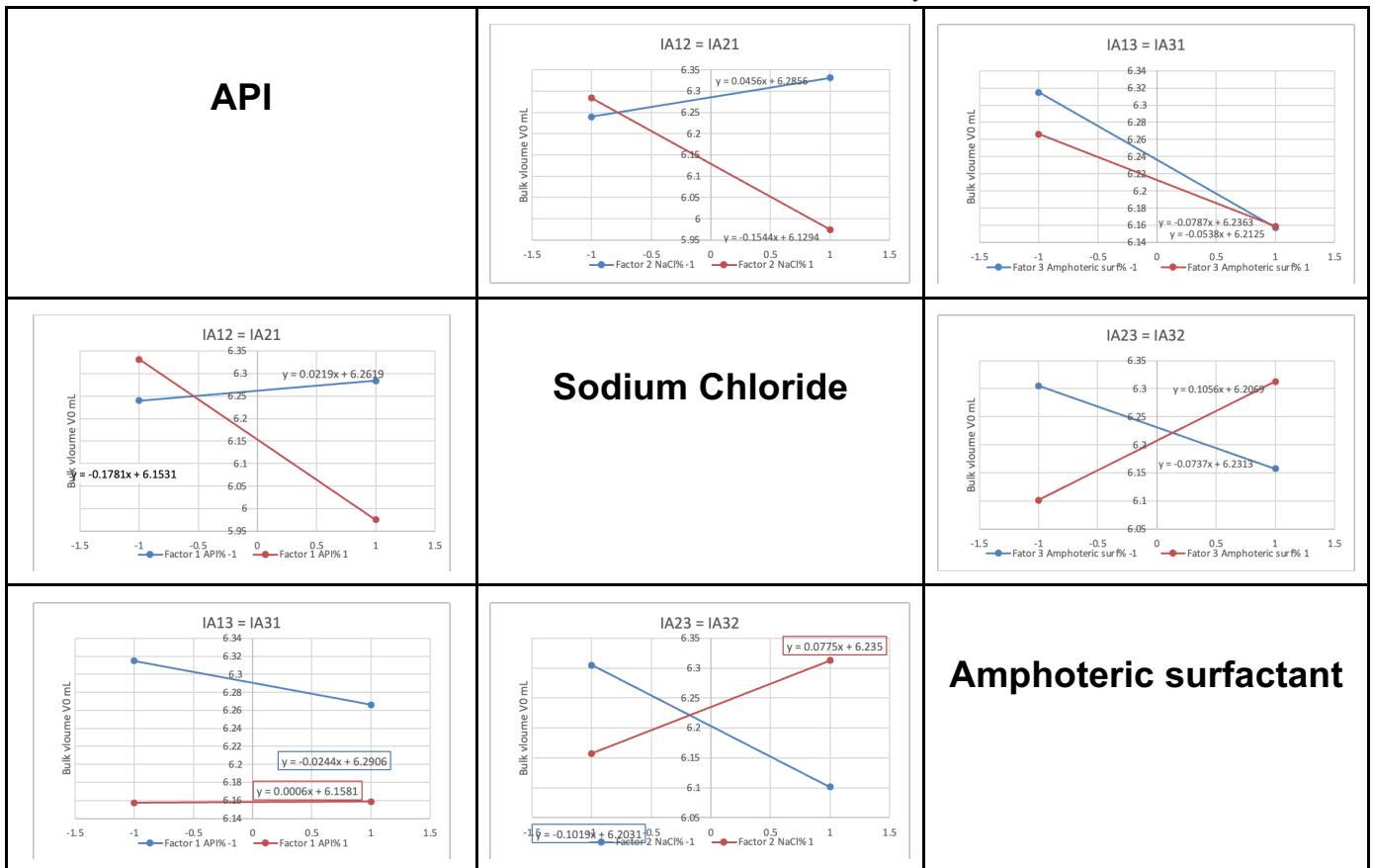
For the effects of pH at 50 degrees week 2, the significant factor was amphoteric surfactant, with a positive effect of 0.22. The significant interactions were IA12, IA13, IA23, IA123, and they had a positive effect of 0.30, 0.34, 0.44, 0.24 respectively, when these interactions moved from level-1 to level+1.

4.3.2.2 Interactions

A detailed interaction analysis was performed in “effect of factor” section

4.3.2.2.1 Week 0 4°C

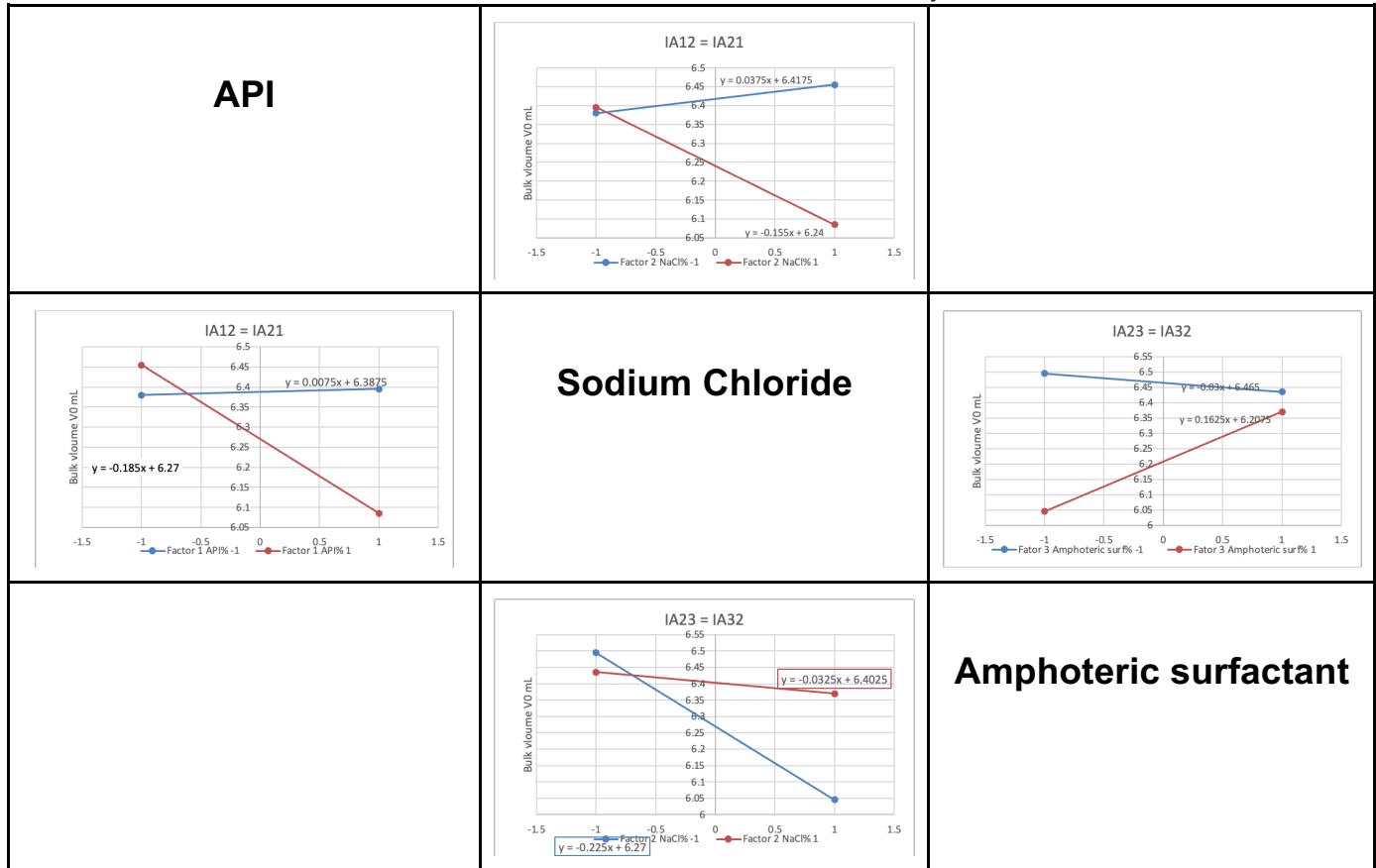
Table 13. The interactions between three factors in viscosity at week 2 at 50°C



The intersection points in the graphs of IA12, IA21, IA23, IA32, IA13 indicated that there might be significant interaction between Isethionate% and NaCl%, NaCl% and Amphoteric Surf%, Isethionate% and Amphoteric Surf%, which might have significant effects on pH .

4.3.2.2.2 Week 2 4°C

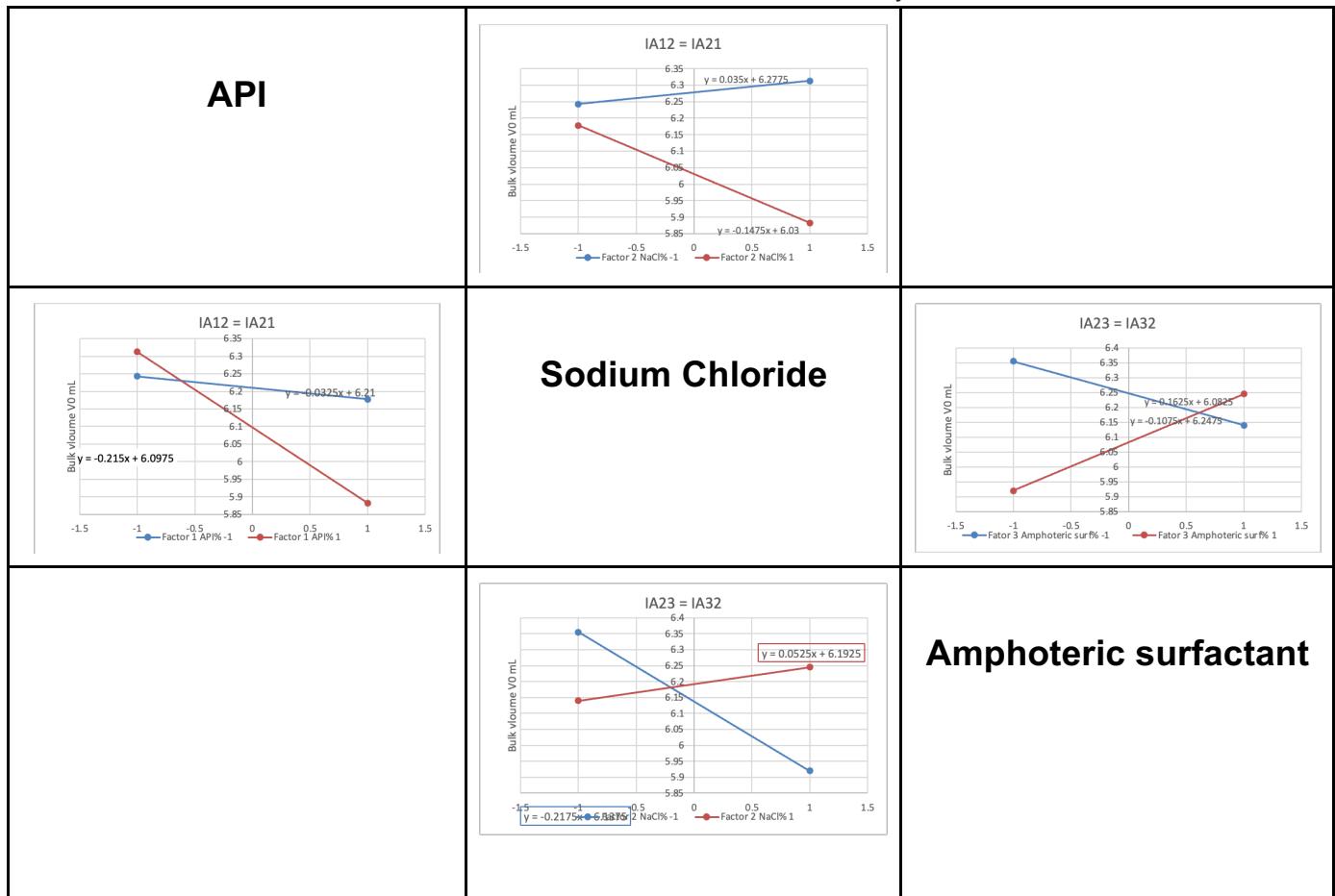
Table 14. The interactions between three factors in viscosity at week 2 at 4°C



There might be significant interactions between Isethionate% and NaCl%, NaCl% and Amphoteric Surf% as shown in the graph of IA12, IA21 and IA23, IA32 which could impact on pH.

4.3.2.2.3 Week 0 25°C

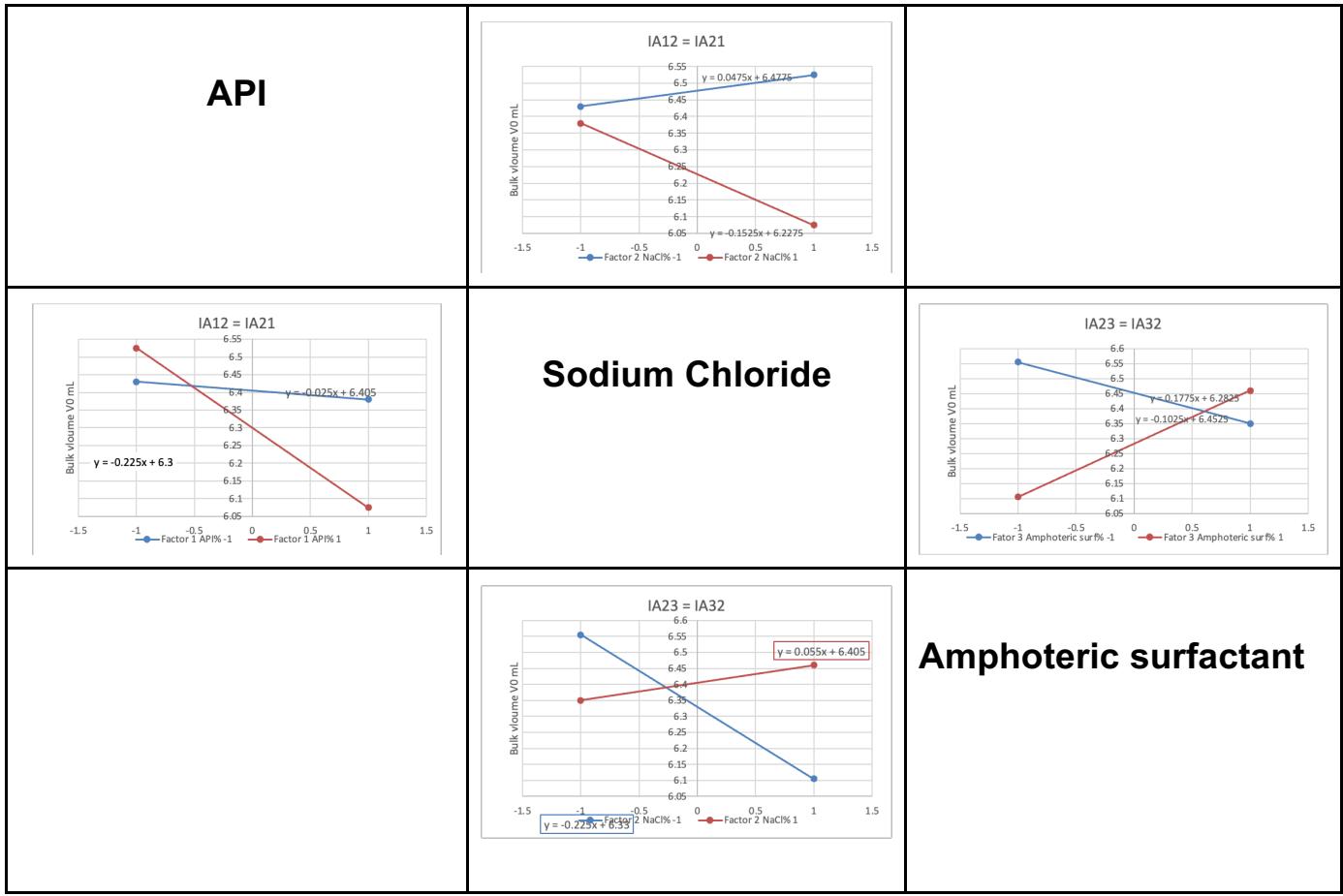
Table 15. The interactions between three factors in viscosity at week 0 at 25°C



There might be significant interactions between Isethionate% and NaCl%, NaCl% and Amphoteric Surf% as shown in the graph of IA12, IA21 and IA23, IA32 to affect the pH.

4.3.2.2.4 Week 2 25°C

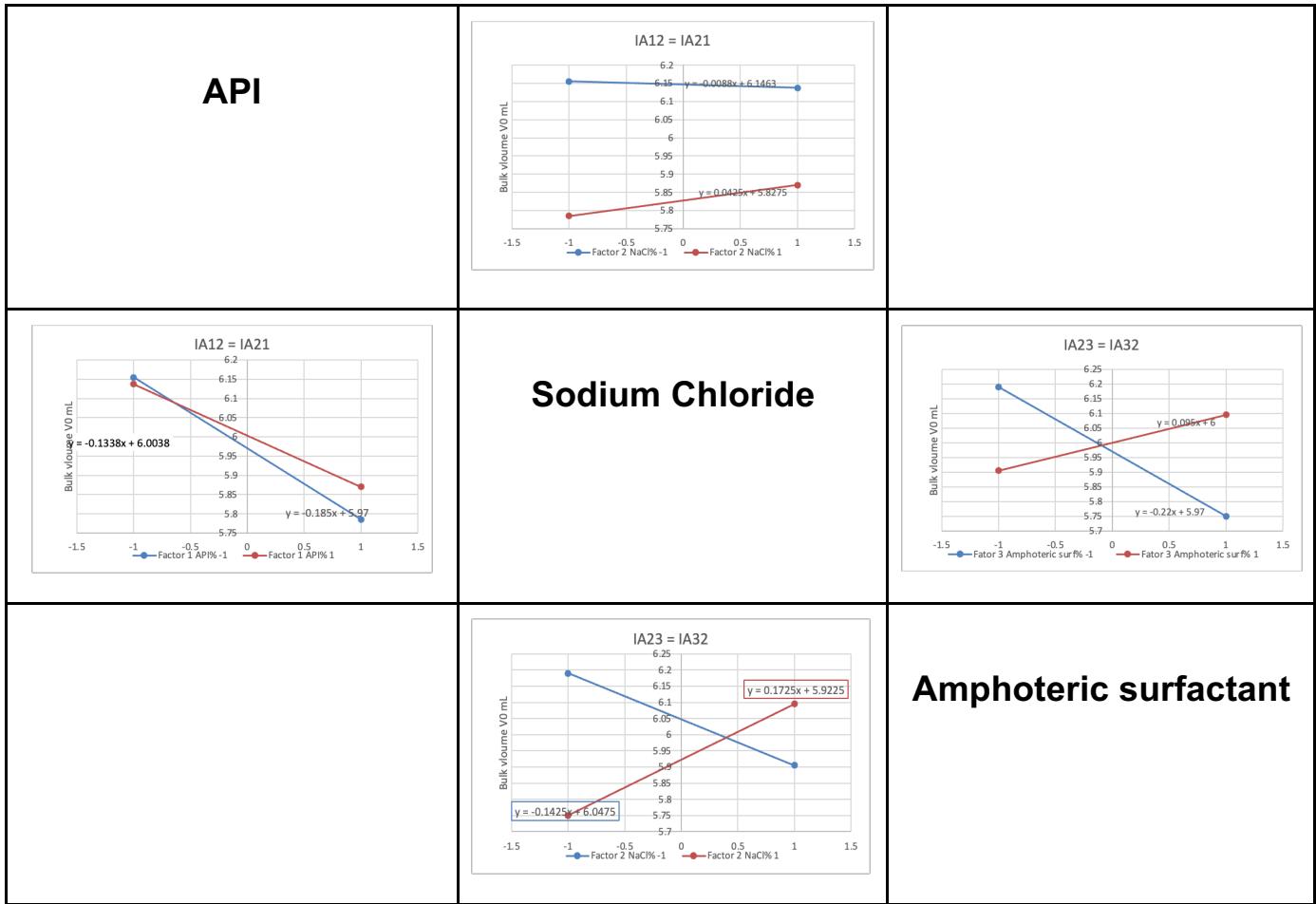
Table 16. The interactions between three factors in viscosity at week 2 at 25°C



There might be significant interactions between Isethionate% and NaCl%, NaCl% and Amphoteric Surf% as shown in the graph of IA12, IA21 and IA23, IA32 to effect pH.

4.3.2.2.5 Week 0 50°C

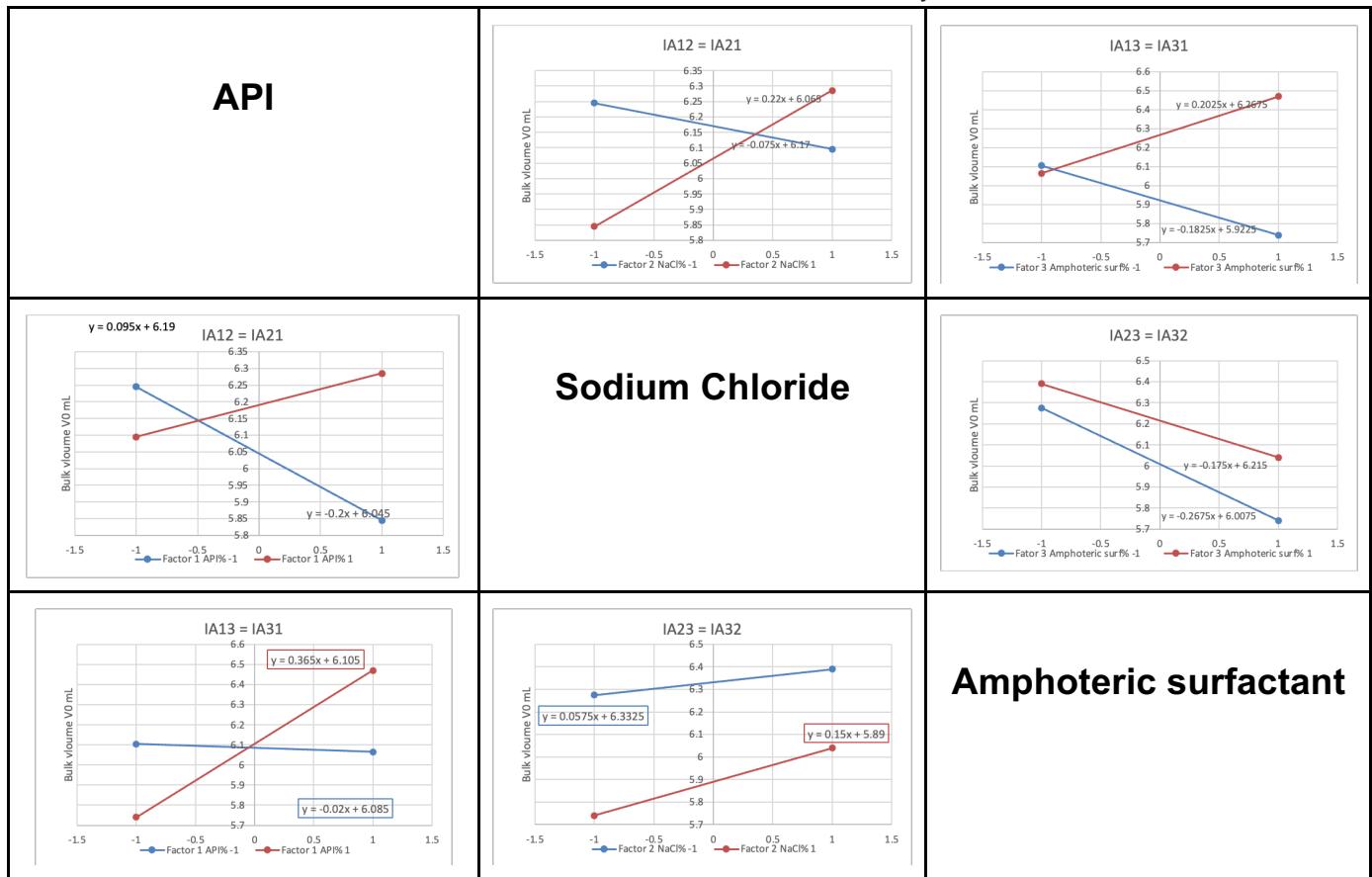
Table 17. The interactions between three factors in viscosity at week 0 at 50°C



There might be significant interactions between Isethionate% and NaCl%, NaCl% and Amphoteric Surf% as shown in the graph of IA12 21 and IA23 32 which might have significant effects on pH.

4.3.2.2.6 Week 2 50°C

Table 18. The interactions between three factors in viscosity at week 2 at 50°C



There might be significant interactions between Isethionate% and NaCl%, Isethionate% and Amphoteric Surf% as shown in the graph of IA12, IA21 and IA13, IA31.

4.3.2.3 ANOVA

4.3.2.3.1 Reduced model for average results of pH at 4°C week 0 and week 2

Table 19. Reduced model ANOVA table for average results of pH at 4°C week 0 and week 2

Response 7: pH WK0 4							Response 10: pH WK2 4						
Source	Sum of Squares	df	Mean Square	F-value	p-value		Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	0.1525	3	0.0508	15.36	0.0032	significant	Model	0.3133	4	0.0783	7.72	0.0228	significant
A-API	0.0237	1	0.0237	7.15	0.0368		A-API	0.0276	1	0.0276	2.72	0.1598	
B-NaCl	0.0488	1	0.0488	14.76	0.0085		B-NaCl	0.063	1	0.063	6.21	0.0550	
AB	0.08	1	0.08	24.18	0.0027		C-Amphoteric Surfactant	0.1485	1	0.1485	14.64	0.0123	
Curvature	0.0013	1	0.0013	0.39	0.5577		AB	0.0741	1	0.0741	7.31	0.0426	
Residual	0.0198	6	0.0033				Curvature	0.0343	1	0.0343	3.38	0.1252	
Lack of Fit	0.0182	4	0.0045	5.45	0.1609	not significant	Residual	0.0507	5	0.0101			
Pure Error	0.0017	2	0.0008				Lack of Fit	0.0474	3	0.0158	9.68	0.0951	not significant
Cor Total	0.1736	10					Pure Error	0.0033	2	0.0016			
							Cor Total	0.3983	10				

From the reduced ANOVA table for week0 and week2, both models were significant by comparing their p-values (0.0032, 0.0228) with 0.05, Lack of fit were non-significant demonstrated that the model can be used, the p-value of curvature (0.55, 0.13) illustrated the predictability of both models as values were larger than 0.05.

4.3.2.3.2 Reduced model for average results of pH at 25°C week 0 and week 2

Table 20. Reduced model ANOVA table for average results of pH at 25°C week 0 and week 2

Response 8: pH WK0 25						Response 11: pH WK2 25							
Source	Sum of Squares	df	Mean Square	F-value	p-value	Source	Sum of Squares	df	Mean Square	F-value	p-value		
Model	0.2847	4	0.0712	10.47	0.0120	significant	Model	0.307	4	0.0768	12.31	0.0084	significant
A-API	0.0253	1	0.0253	3.72	0.1116		A-API	0.022	1	0.022	3.54	0.1188	
B-NaCl	0.1225	1	0.1225	18.01	0.0081		B-NaCl	0.125	1	0.125	20.05	0.0065	
C-Amphoteric Surfactant	0.0703	1	0.0703	10.34	0.0236		C-Amphoteric Surfactant	0.08	1	0.08	12.83	0.0158	
AB	0.0666	1	0.0666	9.79	0.0260		AB	0.08	1	0.08	12.83	0.0158	
Curvature	0.025	1	0.025	3.68	0.1132		Curvature	0.0019	1	0.0019	0.2978	0.6087	
Residual	0.034	5	0.0068				Residual	0.0312	5	0.0062			
Lack of Fit	0.0297	3	0.0099	4.65	0.1822	not significant	Lack of Fit	0.0287	3	0.0096	7.76	0.1163	not significant
Pure Error	0.0043	2	0.0021				Pure Error	0.0025	2	0.0012			
Cor Total	0.3438	10					Cor Total	0.3401	10				

From the reduced ANOVA table for week0 and week2, both models were significant as the p-values were less than 0.05, Lack of fit were not significant demonstrated that the model can be used, the p-value of curvature (0.1132, 0.6087>0.05) illustrated that these were linear models, resulting in predictability of models as values were larger than 0.05.

4.3.2.3.3 Full and reduced model for average results of pH at 50°C week 0 and week 2

Table 21. Full and reduced ANOVA table of pH at 50°C week 0 and full ANOVA table at 50°C week 2

Response 9: pH WK0 50							Response 12: pH WK2 50						
Source	Sum of Squares	df	Mean Square	F-value	p-value		Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	0.2349	3	0.0783	49.13	0.0001	significant	Model	1.05	7	0.1502	25.74	0.0379	significant
B-NaCl	0.2032	1	0.2032	127.50	<0.0001		A-API	0.0421	1	0.0421	7.21	0.1152	
C-Amphoteric Surfactant	0.0023	1	0.0023	1.43	0.2770		B-NaCl	0.0221	1	0.0221	3.78	0.1913	
BC	0.0294	1	0.0294	18.45	0.0051		C-Amphoteric Surfactant	0.0968	1	0.0968	16.59	0.0553	
Curvature	0.0048	1	0.0048	3.01	0.1335		AB	0.1741	1	0.1741	29.84	0.0319	
Residual	0.0096	6	0.0016				AC	0.2312	1	0.2312	39.63	0.0243	
Lack of Fit	0.0078	4	0.0019	2.16	0.3410	not significant	BC	0.3698	1	0.3698	63.39	0.0154	
Pure Error	0.0018	2	0.0009				ABC	0.1152	1	0.1152	19.75	0.0471	
Cor Total	0.2492	10					Curvature	0.0025	1	0.0025	0.4366	0.5767	
							Pure Error	0.0117	2	0.0058			
Cor Total							Cor Total	1.07	10				

The reduced ANOVA for week 0 50 degrees was shown in the table above, the significant model (0.0001) and insignificant lack of fit (0.341) illustrated that this model can be used, the curvature again nonsignificant (0.1335>0.05). Hence, the model of pH at 50 degrees week0 was predictable. For the week2 model, as IA123 was significant in ANOVA full table, this model cannot be reduced due to the hierarchical mode in Design Expert. Week 2 full model was significant ($p<0.05$). IA12, IA13, IA23, IA123 were significant interactions in this model (0.0319, 0.243, 0.0154, 0.0471<0.05). The high p-value for curvature demonstrated the linear characteristic of this model so that it is predictable.

4.3.3 Interpretation

In the study, the pH test was done in week 0 and week 2, followed by 3 different temperature environment 4°C, 25°C, and 50°C from the ANOVA tables, factor 1 - API, factor 2 - NaCl, factor 3 - amphoteric surfactant, interaction 12 and 23 showed significant impacts on the pH results. In factor 1 - Isethionate API, it showed only a significant effect on pH in week 0 at 4°C. Based on the research, the amount of API had a little effect on the response, therefore, this significance might be due to the experimental error that occurred in the study. For factor 2 sodium chloride, the original pH adjuster NaOH and citric acid was not added into body wash to control pH. Accordingly, sodium chloride was used as both viscosity and pH adjuster in the study. Since a pH adjuster can be used to alter pH or potential hydrogen level (Claire Gillespie, 2022), the result of pH will drive downward when it was added into the solution. Therefore, NaCl showed a negative effect on pH value where the increased amount of sodium chloride will cause a low pH result. For factor 3, amphoteric surfactant contains an ionic charge which causes the surfactant change between anionic and cationic properties (negative and positive charge). As a result, amphoteric surfactant showed both acidic and basic properties depending on whether the solution is alkaline or acidic (Fan, Q, 2008). According to the flow chart, amphoteric surfactant was added after the presence of isethionate. While Isethionate has a pH value from the range of 6-8 which is an alkaline environment, amphoteric surfactant will show acidic properties in the body wash to lower the pH result. Therefore, the addition of amphoteric surfactant will cause a decrease in pH value. The hypothesis was then supported by the experimental data from the study which was discussed in the analysis part.

4.3.4 Conclusion

In conclusion, factor 2 - NaCl and factor 3 - Amphoteric surfactant showed a significant effect on the final pH results regarding their physicochemical properties. Despite factor 1 - Isethionate API showed an impact on pH in week 0 at 4°C, the overall hypothesis was supported by the analysis of experimental data where NaCl acts as a pH adjuster to lower pH value and amphoteric surfactant shows acidic property in the body wash. Therefore, when we wanted to minimise the pH value, the level of sodium chloride and amphoteric surfactant should be set up at +1 which is 5% and 20%.

4.4 Foam ability / stability

4.4.1 Evaluation

From the song graph of foam stability of body wash in both week 0 and 2 from 4°C to 50°C, all experimental data were correctly recorded on the batch record, and no transferring error was made in the result table. The reduction of foam stability data was retrieved from the percentage of division of bubbles reduced in 1 minute and initial bubble volume. Since all error bars were attached to the data, the results of centre points were consistent, and no significant outlier occurred in the graph. In the curve, the significant level gradient lines of both factors and interactions were presented from experiment 1 to 8. Therefore, the results were sufficient to move on to the analysis part.

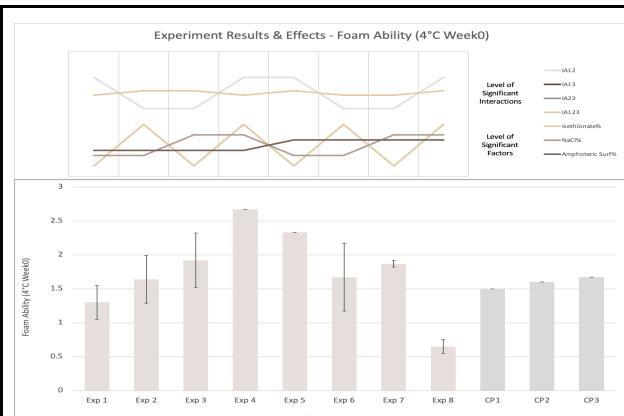


Figure 40. Average results for Foam Ability at 4°C week 0, with SD (error bar), compared to CPs, significant factors and interactions (Isethionate%, NaCl%, Amphoteric Surf%, IA12, IA23)

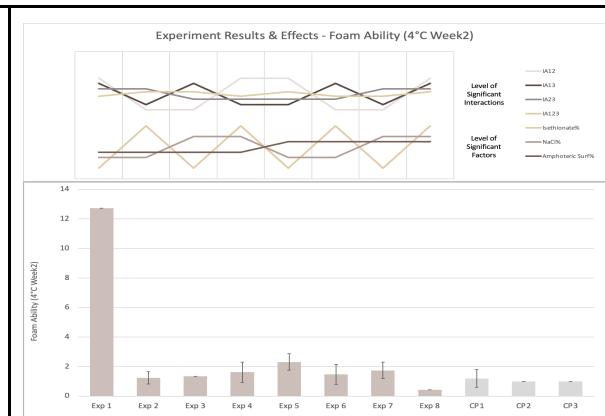
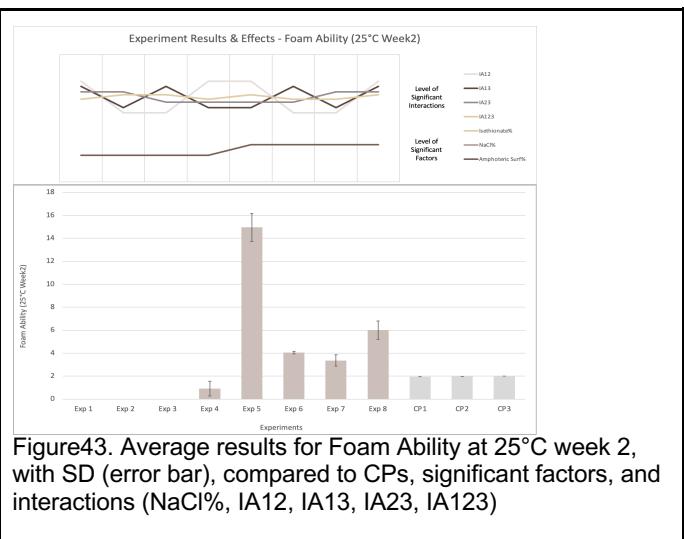
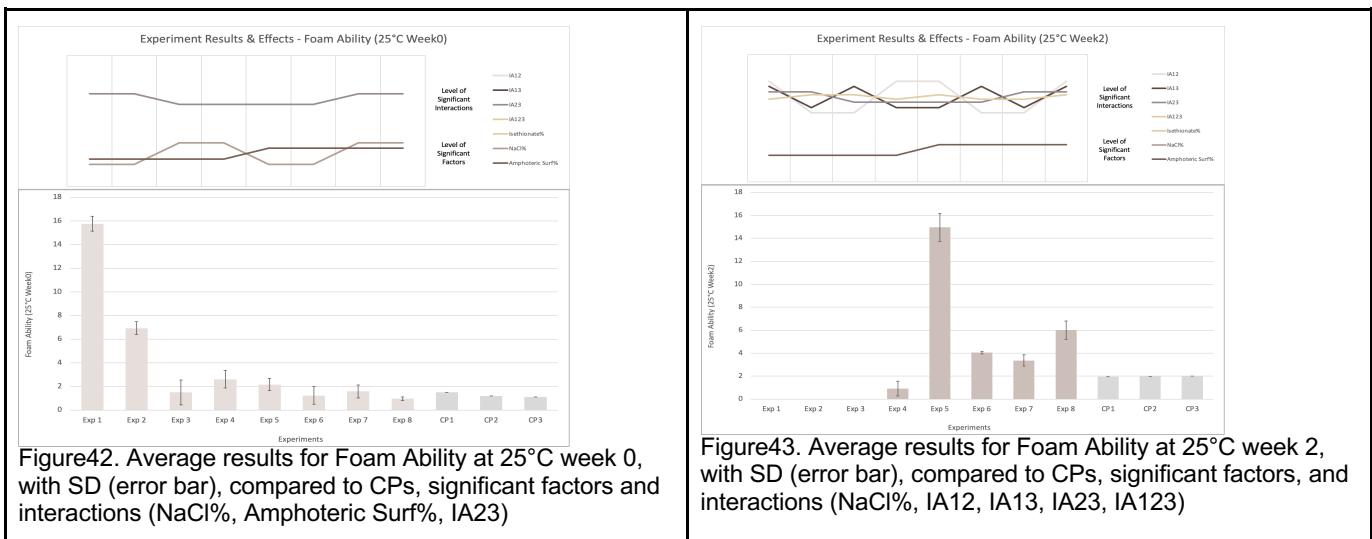
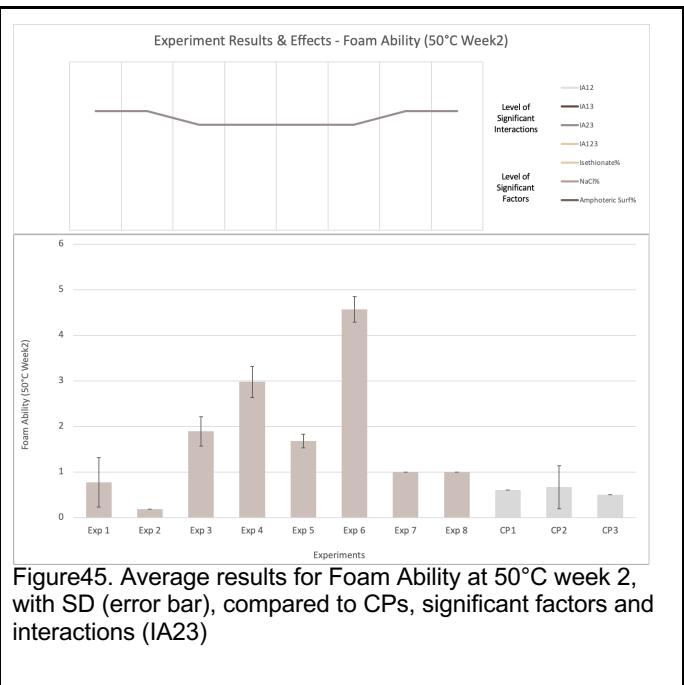
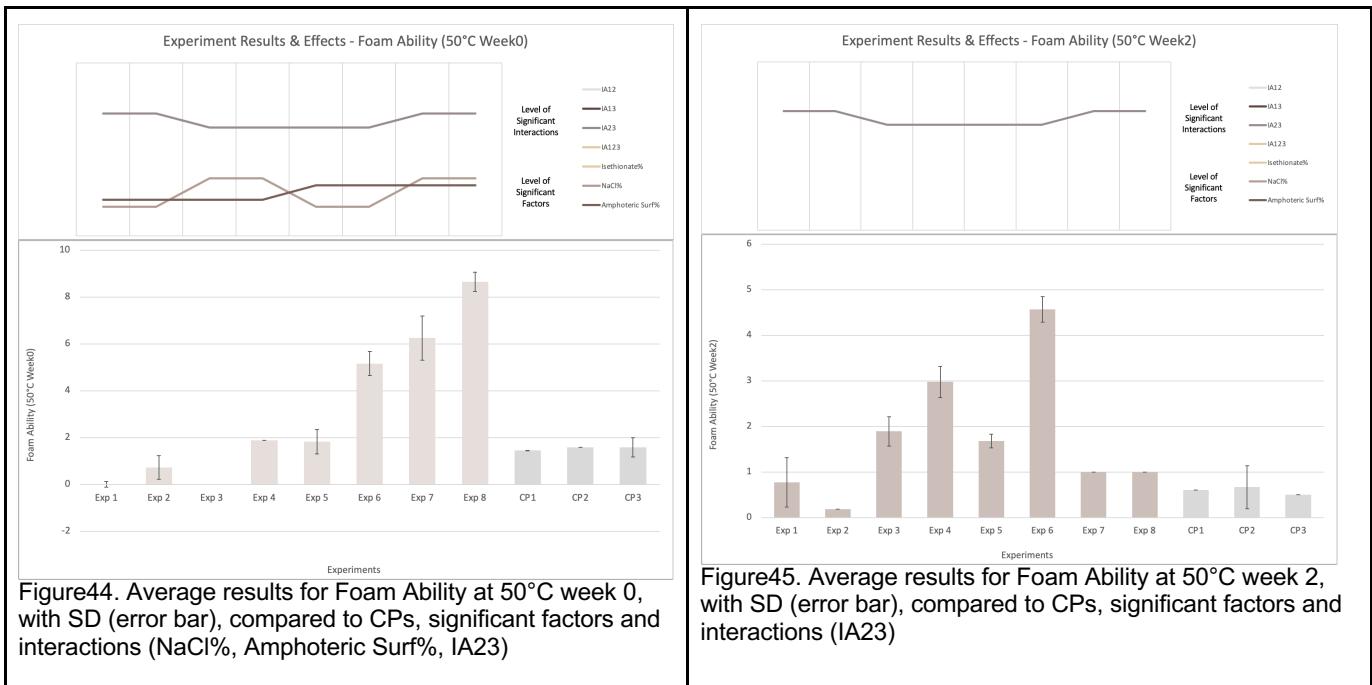


Figure 41. Average results for Foam Ability at 4°C week 2, with SD (error bar), compared to CPs, significant factors and interactions (Isethionate%, NaCl%, Amphoteric Surf%, IA12, IA13, IA23, IA123)

For foam stability at 4°C at both week 0 and week 2, based on the level gradient line and experimental data, no significant factors can be determined. For interaction, IA13 and 23 show a significant effect on the response.



For foam stability at 25°C at both week 0 and week 2, based on the level gradient line and experimental data, all factors showed a significant effect on the response. For interaction, IA23 in week 0 and all interactions in week 2 also had significant impacts on the response.



For foam stability at 50°C at both week 0 and week 2, based on the level gradient line and experimental data, factor 2 - NaCl and factor 3 - amphoteric surfactant in week0 and no factors in week 2 showed a significant effect on the response. For interaction, for both week 0 and 2, only IA23 had significant impacts on the response.

However, the data obtained were dramatically different. However, this difference may be due to systematic error instead of the instability of samples.

Variance factors and interactions were determined having significant effects on foam ability/stability. However, the data obtained were dramatically different. However, this difference may be due to systematic error instead of the instability of samples.

4.4.2 Analysis

4.4.2.1 Effect of factor

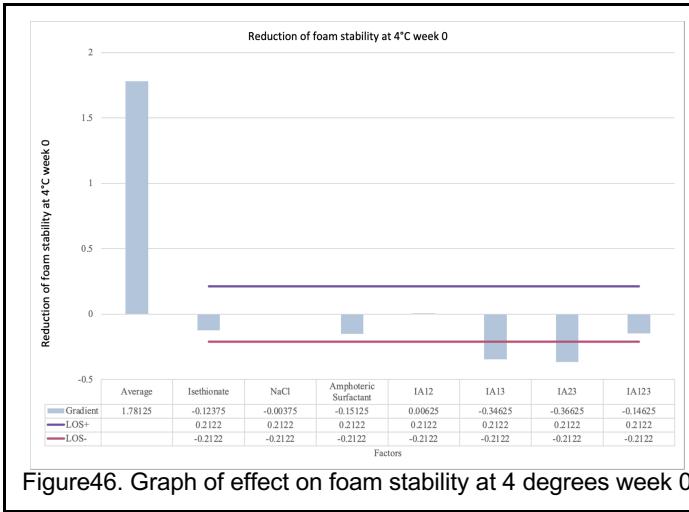


Figure46. Graph of effect on foam stability at 4 degrees week 0

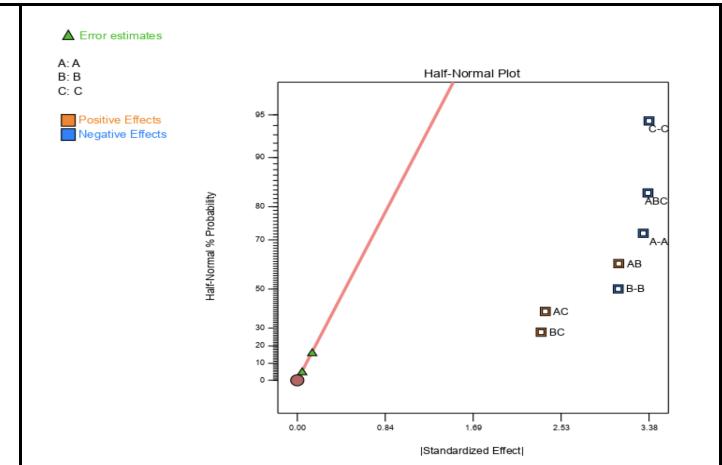


Figure47. Half normal plot of foam stability at 4 degrees week 0

In week 0 at 4 degree, since no factors passed the LOS line at both positive and negative values, there was no significant factor that will have an impact on reduction of foam ability result. However, interaction 13 and 23 showed a negative impact on the reduction of foam ability. Hence, increase the foam stability.

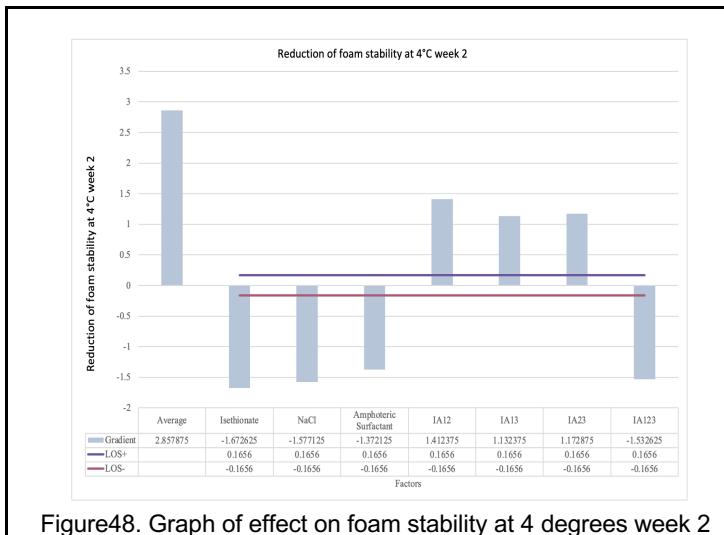


Figure48. Graph of effect on foam stability at 4 degrees week 2

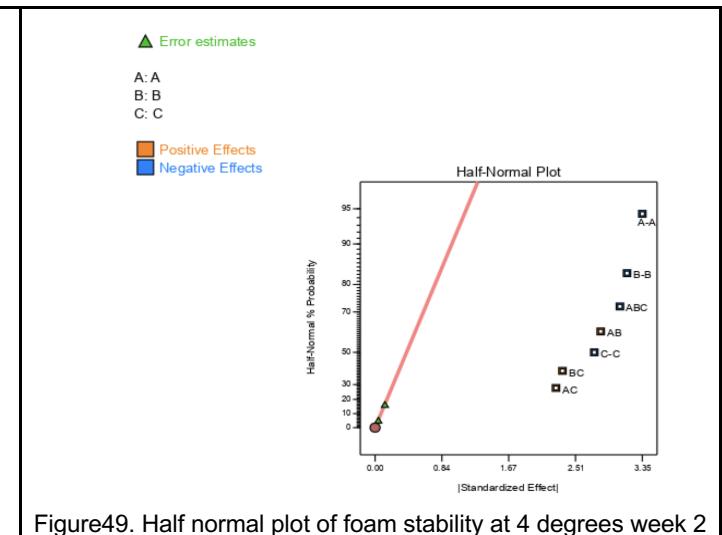


Figure49. Half normal plot of foam stability at 4 degrees week 2

In week 2 at 4°C, all factors and interactions showed a significant effect on the reduction of foam ability. Based on the gradient effect result, factor 1, 2 and 3 had a negative impact on reduction of foam ability, therefore, changing the level of API, NaCl and amphoteric surfactant from -1 to +1 will decrease 3.35, 3.16 and 2.74 respectively in reduction of foam ability. Hence, increase the foam stability. Besides, interaction 12,13 and 23 showed a positive effect on the response, while interaction 123 showed a negative impact.

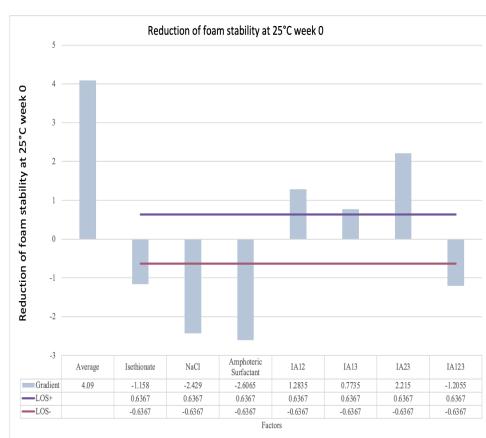


Figure50. Graph of effect on foam stability at 25 degrees week 0

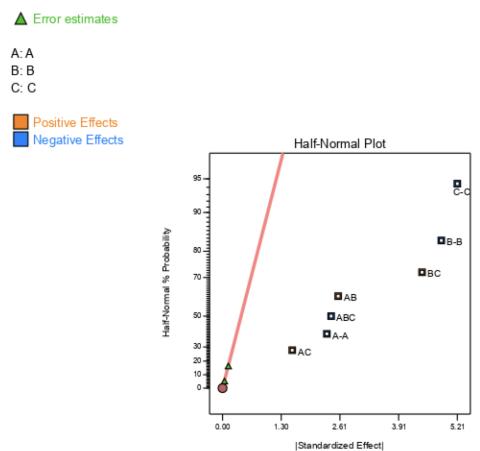


Figure51. Half normal plot of foam stability at 25 degrees week 0

In week 0 at 25°C, like the effect in week 2 4°C changing the level of API, NaCl and amphoteric surfactant from -1 to +1 will decrease 2.32, 4.86 and 5.214 respectively. in reduction of foam ability. Hence, increase the foam stability. And interaction 12,13 and 23 showed a positive effect on the response, while interaction 123 showed a negative impact.

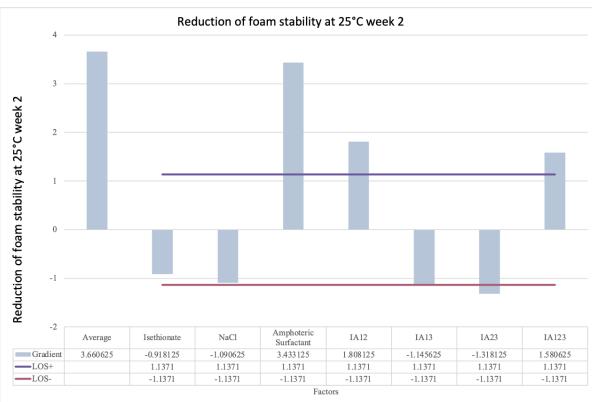


Figure52. Graph of effect on foam stability at 25 degrees week 2

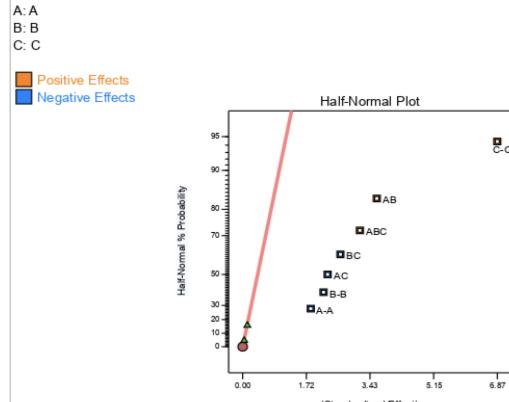
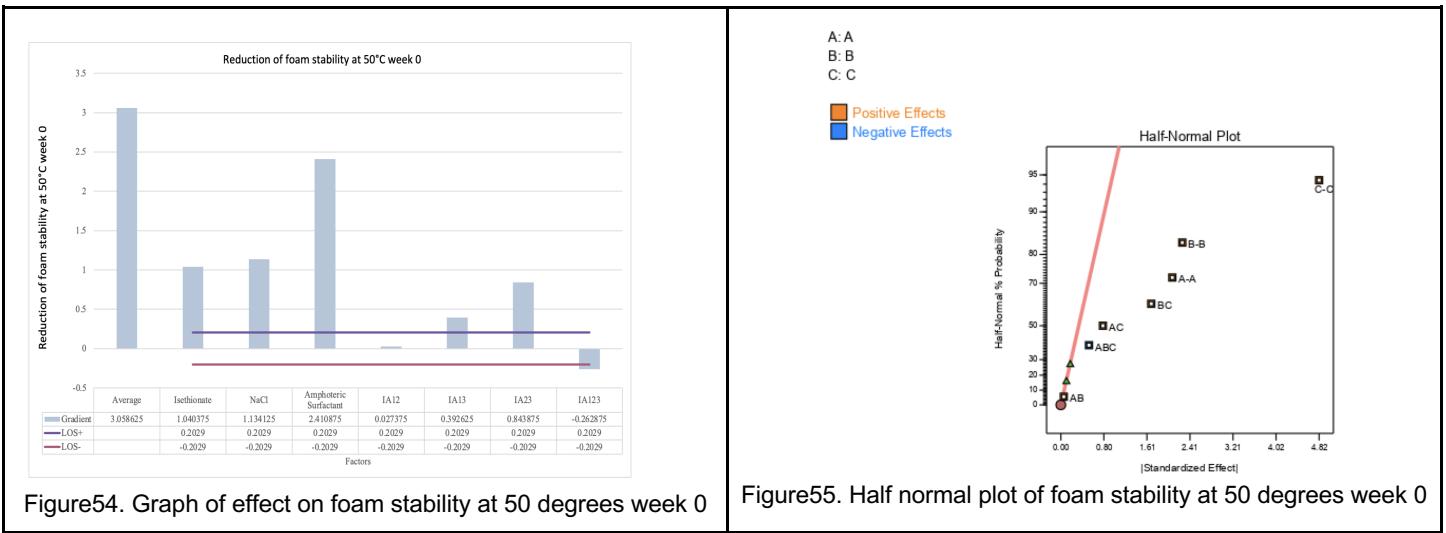
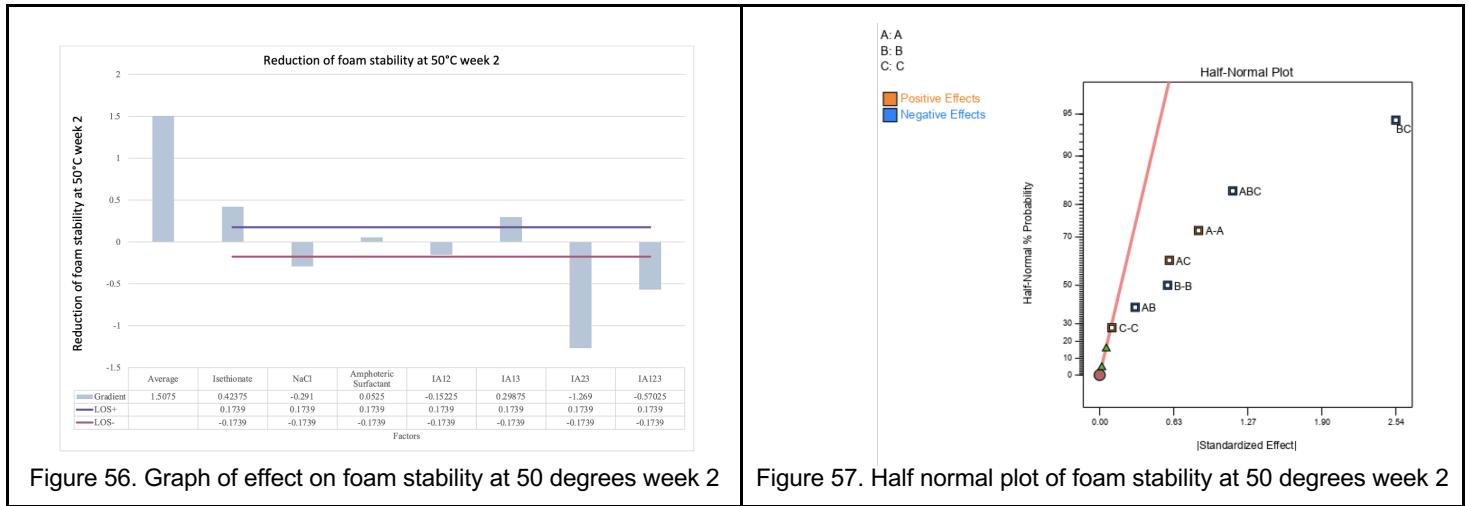


Figure53. Half normal plot of foam stability at 25 degrees week 2

In week 2 at 25°C, only factor 3 - amphoteric surfactant showed a significantly positive effect that changing the level of amphoteric surfactant from -1 to +1 will increase 6.87 respectively in reduction of foam ability. Hence, decreased the foam stability. Also, interaction 13 and 23 showed a negative effect on the response, while interaction 123 showed a positive impact.



In week 0 at 50°C, all factors showed a positive effect on the reduction of foam ability which means changing their level from -1 to +1 will lead to an increase of 2.08, 2.27, 4.82 respectively in the reduction of foam ability. Hence, decreased the foam stability. Also, the interaction 13 and 23 would cause a positive impact on the response whereas interaction 123 can decrease the reduction of foam ability.



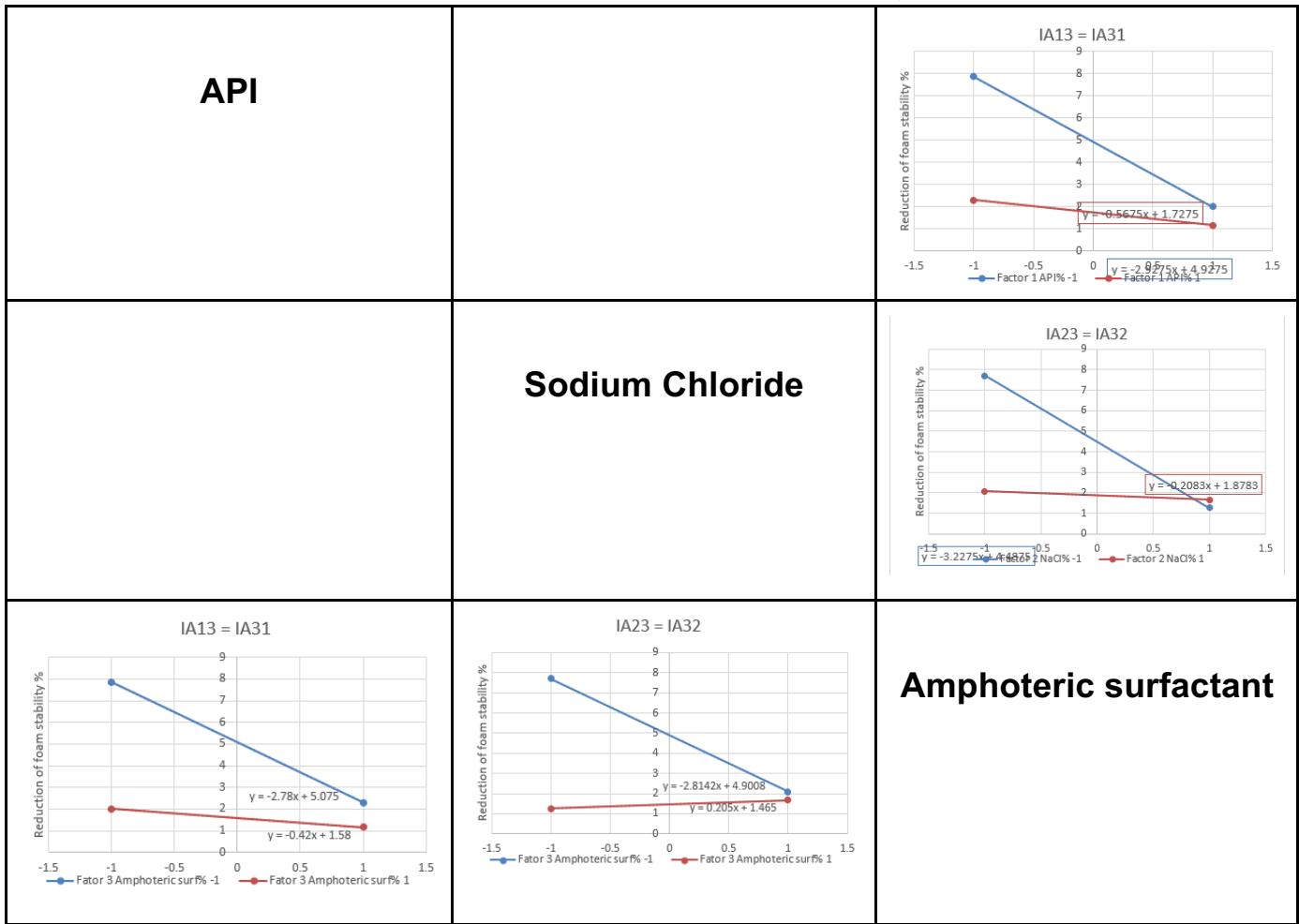
In week 2 50°C, factor 1 -isethionate showed a positive effect on the response when changing from level -1 to +1, the foam ability will increase by 0.85, and factor 2 NaCl showed a negative effect on the reduction of foam ability, changing its level from -1 to +1 will lead to a decrease of 0.58 in the reduction of foam ability. Hence, increase the foam stability. Also, the interaction 23 and 23 will cause a negative impact on the response whereas interaction 13 can increase the reduction of foam ability.

4.4.2.2 Interactions

A detailed interaction analysis was performed in “effect of factor” section

4.4.2.2.1 Week 0 4°C

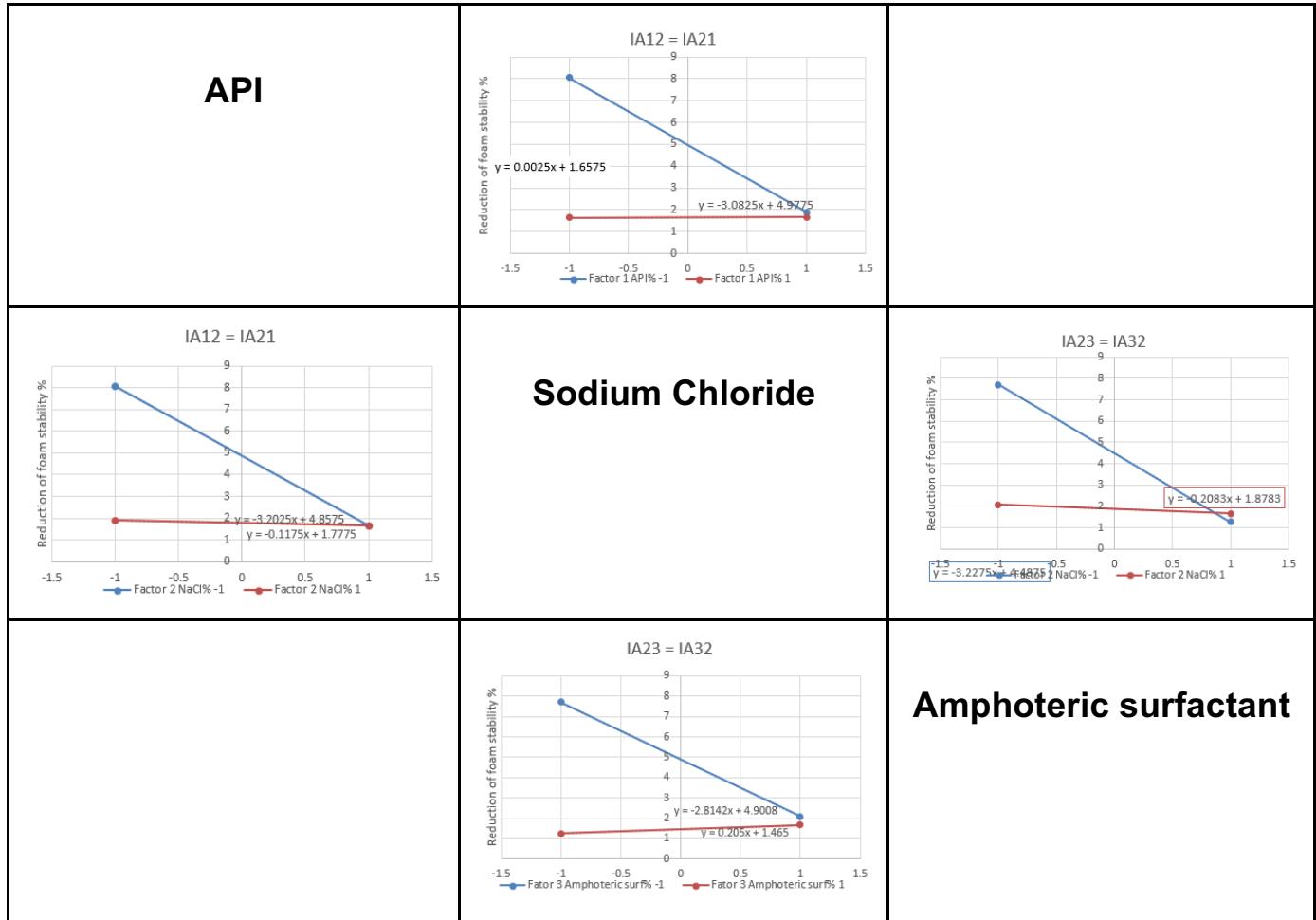
Table 22. The interactions between three factors in foam stability at week 0 at 4°C



The intersection points indicated that there might be significant interactions between Isethionate% and Amphoteric Surf%, NaCl% and Amphoteric Surf% as shown in the graph of IA13 31 and IA23 32 which might have significant effects on reduction of foam stability.

4.4.2.2.2 Week 0 25°C

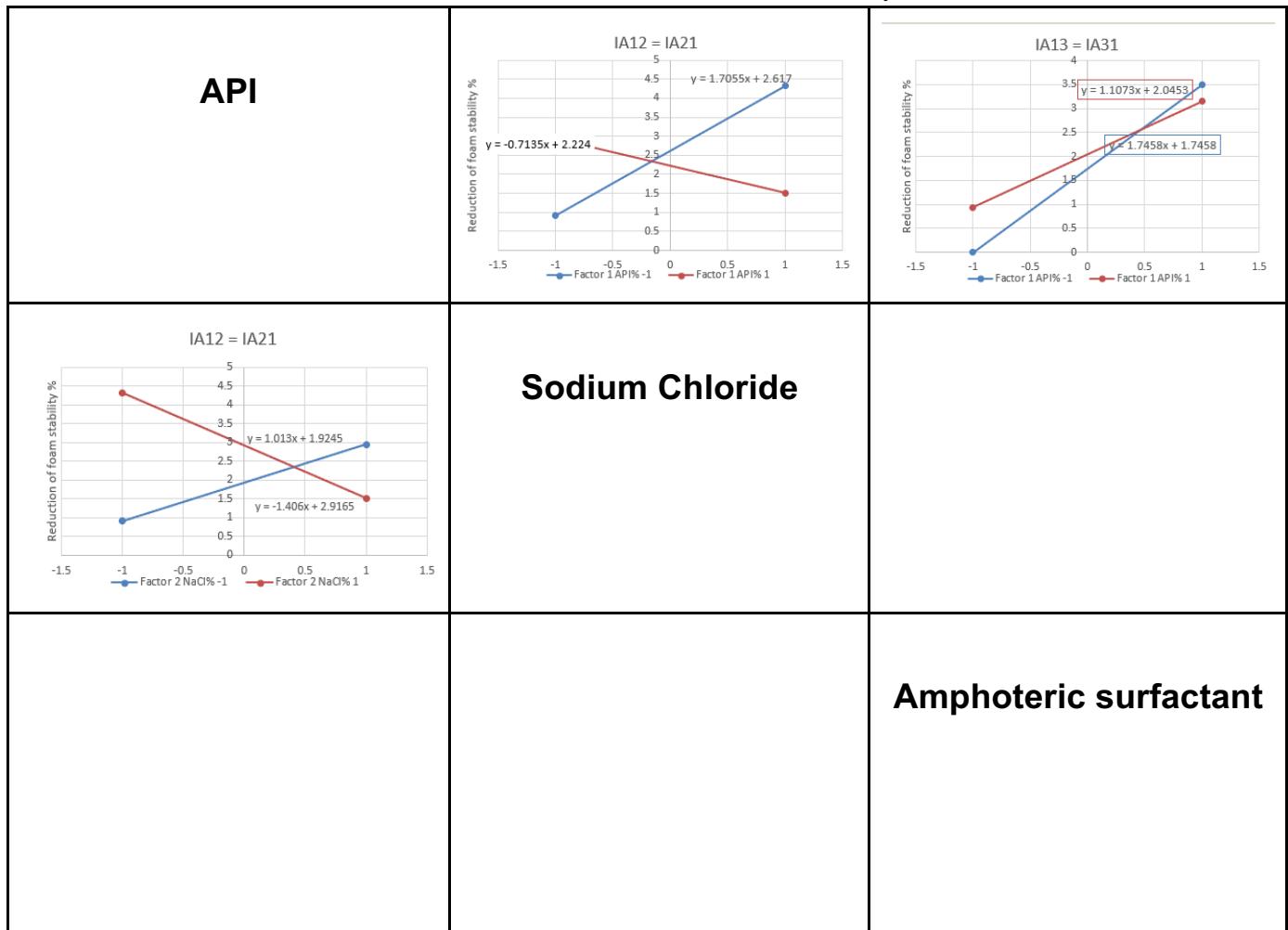
Table 23. The interactions between three factors in foam stability at week 0 at 25°C



There might be significant interactions between Isethionate% and NaCl%, NaCl% and Amphoteric Surf% as shown in the graph of IA12 21 and IA23 32 which might have significant effects on reduction of foam stability.

4.4.2.2.3 Week 0 50°C

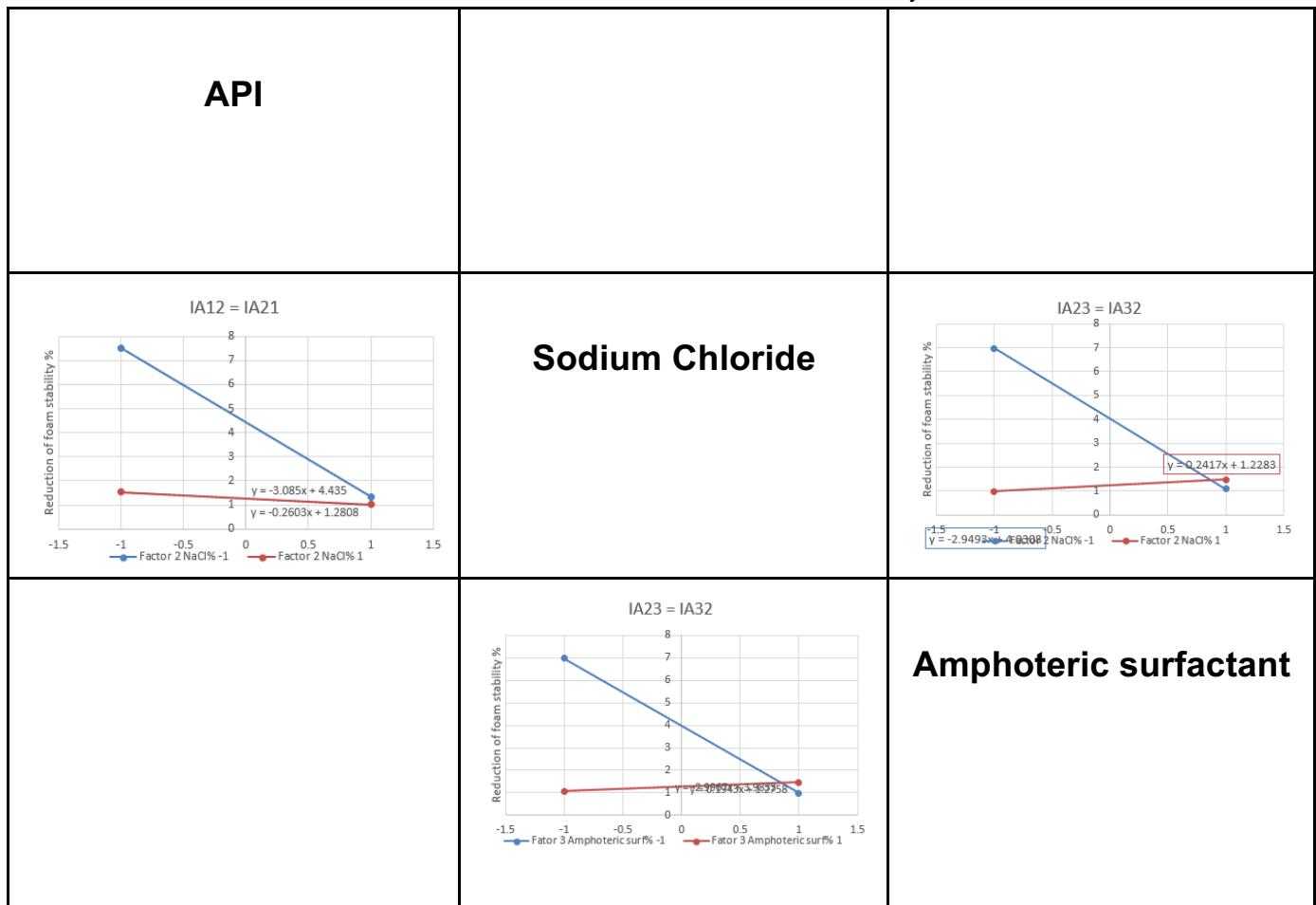
Table 24. The interactions between three factors in foam stability at week 0 at 50°C



There might be significant interactions between Isethionate% and NaCl%, NaCl% and Amphoteric Surf% as shown in the graph of IA12 21 and the interaction of IA13 might reveal some significant effects on reduction of foam ability

4.4.2.2.4 Week 2 4°C

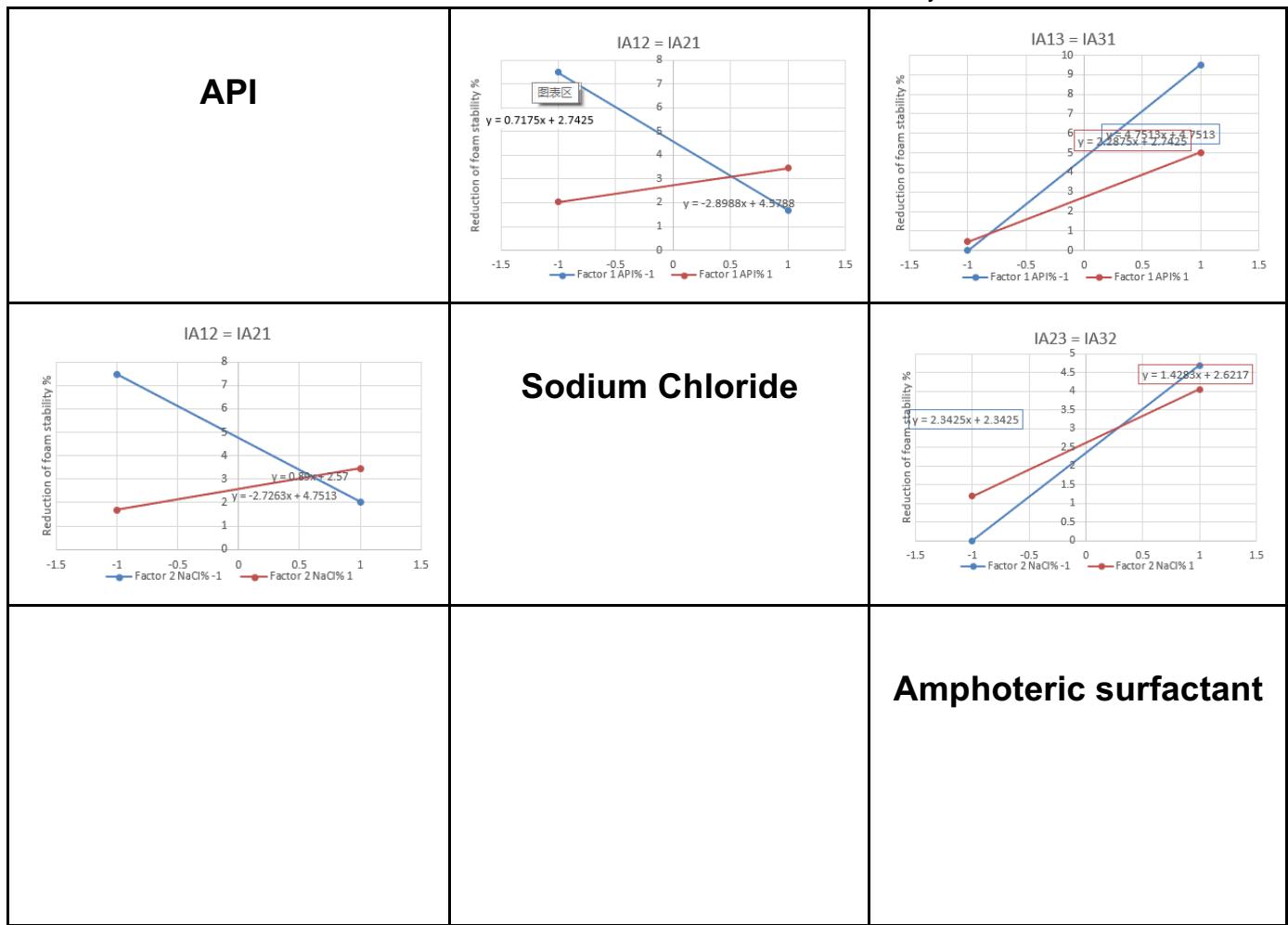
Table 25. The interactions between three factors in foam stability at week 2 at 4°C



There might be significant interactions between Isethionate% and NaCl% which is IA21, followed by NaCl% and Amphoteric Surf%, Amphoteric Surfactant% and NaCl% as shown in the graph of IA23 32 which might have significant effects on reduction of foam ability.

4.4.2.2.5 Week 2 25°C

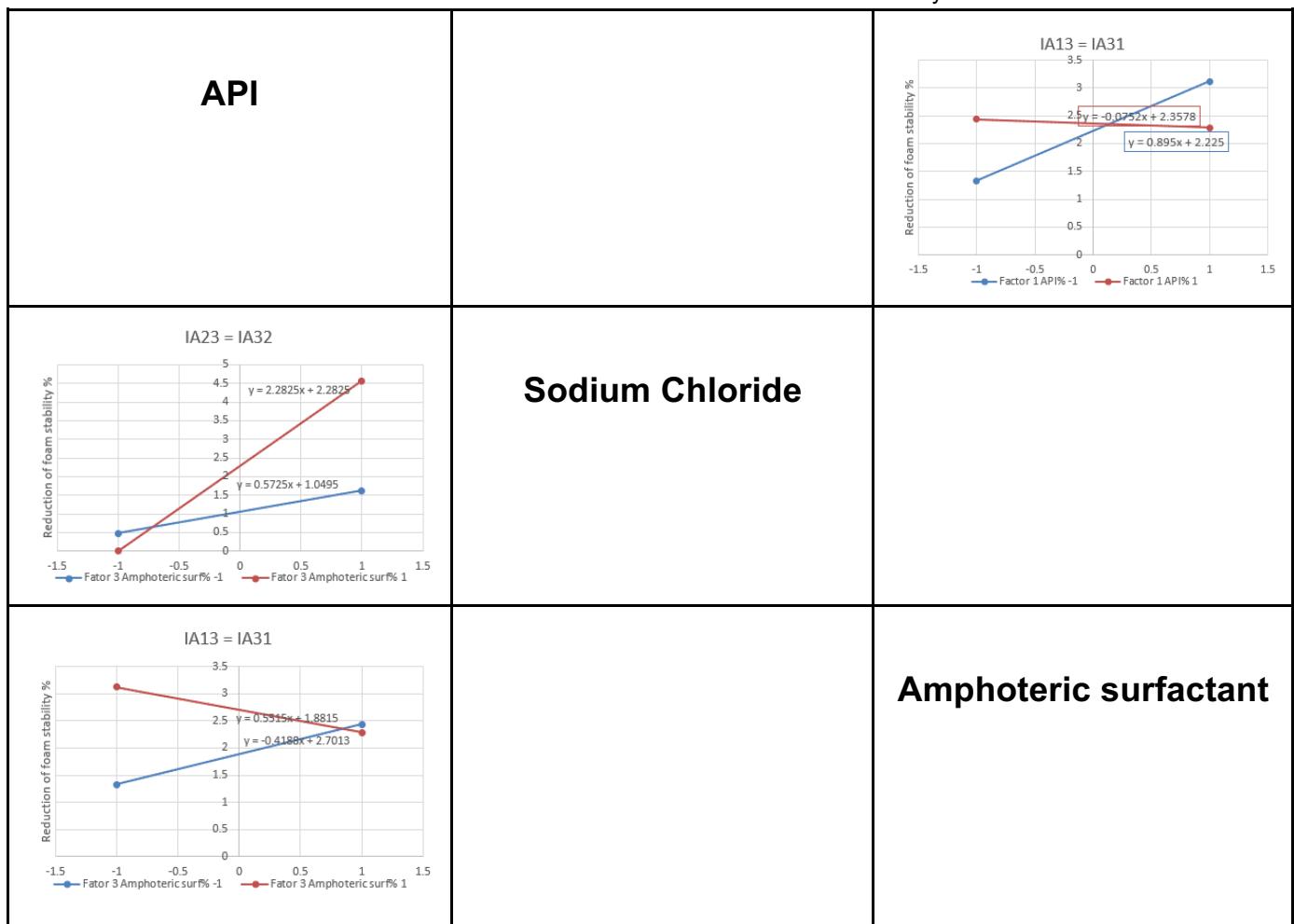
Table 26. The interactions between three factors in foam stability at week 2 at 25°C



There might be significant interactions between Isethionate% and NaCl%, NaCl% and Isethionate%, NaCl% and Amphoteric Surf%, as well as Amphoteric Surfactant% and NaCl% as shown in the graph of IA12 21 and IA13 32 which might have significant effects on reduction of foam stability.

4.4.2.2.6 Week 2 50°C

Table 27. The interactions between three factors in foam stability at week 2 at 50°C



There might be significant interactions between NaCl% and Amphoteric Surfactant%, Amphoteric Surfactant% and NaCl% as shown in the graph of IA23 32 which might have significant effects on the reduction of foam stability. Moreover, interaction between Isethionate% and Amphoteric Surfactant% may also have some impact on the response.

4.4.2.3 ANOVA

4.4.2.3.1 Full model for average results of foam ability/stability at 4°C week 0 and week 2

Table 28. Full model ANOVA table for foam stability at 4°C week 0 and week 2

Response 13: Foam Stability WK0 4							Response 16: Foam Stability WK2 4						
Source	Sum of Squares	df	Mean Square	F-value	p-value		Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	127.76	7	18.25	19.63	0.0493	significant	Model	113.36	7	16.19	23.69	0.0411	significant
A-API	22.04	1	22.04	23.71	0.0397		A-API	22.38	1	22.38	32.74	0.0292	
B-NaCl	18.97	1	18.97	20.41	0.0457		B-NaCl	19.9	1	19.9	29.11	0.0327	
C-Amphoteric Surfactant	22.78	1	22.78	24.51	0.0385		C-Amphoteric Surfactant	15.06	1	15.06	22.03	0.0425	
AB	19.03	1	19.03	20.48	0.0455		AB	15.96	1	15.96	23.35	0.0403	
AC	11.33	1	11.33	12.19	0.0732		AC	10.26	1	10.26	15.01	0.0606	
BC	10.95	1	10.95	11.78	0.0754		BC	11.01	1	11.01	16.1	0.0569	
ABC	22.65	1	22.65	24.36	0.0387		ABC	18.79	1	18.79	27.49	0.0345	
Curvature	16.63	1	16.63	17.89	0.0516		Curvature	12.36	1	12.36	18.09	0.0511	
Pure Error	1.86	2	0.9296				Pure Error	1.37	2	0.6835			
Cor Total	146.25	10					Cor Total	127.09	10				

From the full model ANOVA table for week0 and week2 at 4°C, both models were significant while their p-values are less than 0.05, also, the curvature is greater than 0.05 which showed it was not significant and the model is linear. Therefore, the model was reliable and can be used to predict the response.

4.4.2.3.2 Full and reduced model for average results of foam ability/stability at 25°C week 0 and week 2

Table 29. Full and reduced model ANOVA table for foam stability at 25°C week 0 and week 2

Response 14: Foam Stability WK0 25							Response 17: Foam Stability WK2 25						
Source	Sum of Squares	df	Mean Square	F-value	p-value		Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	101.55	2	50.78	4.38	0.0583	not significant	Model	181.09	7	25.87	35.97	0.0273	significant
B-NaCl	47.2	1	47.2	4.08	0.0833		A-API	6.74	1	6.74	9.38	0.0922	
C-Amphoteric Surfactant	54.35	1	54.35	4.69	0.067		B-NaCl	9.52	1	9.52	13.23	0.068	
Curvature	24.31	1	24.31	2.1	0.1907		C-Amphoteric Surfactant	94.29	1	94.29	131.09	0.0075	
Residual	81.06	7	11.58				AB	26.15	1	26.15	36.36	0.0264	
Lack of Fit	79.57	5	15.91	21.32	0.0454	significant	AC	10.5	1	10.5	14.6	0.0622	
Pure Error	1.49	2	0.7466				BC	13.9	1	13.9	19.32	0.0481	
Cor Total	206.92	10					ABC	19.99	1	19.99	27.79	0.0342	

From the full model ANOVA table for week 0 and week 2 at 25°C, both models were significant while their p-values are less than 0.05, however, for the curvature in week 0, it was less than 0.05 which showed the model was not linear, thus the model cannot be used for prediction and needs further reduced model analysis. For week 2, the curvature was not significant since the p-value is greater than 0.05, therefore, the week 2 model was reliable and can be used to predict the response.

4.4.2.3.3 Reduced model for average results of foam ability/stability at 50°C week 0 and week 2

Table 30. Reduced model ANOVA table for foam stability at 50°C week 0 and week 2

Response 15: Foam Stability WK0 50							Response 18: Foam Stability WK2 50						
Source	Sum of Squares	df	Mean Square	F-value	p-value		Source	Sum of Squares	df	Mean Square	F-value	p-value	
Model	56.79	2	28.39	11.6	0.0060	significant	Model	13.58	3	4.53	5.19	0.0418	significant
C-Amphoteric Surfactant	46.5	1	46.50	19	0.0033		B-NaCl	0.6774	1	0.68	0.7769	0.4120	
Curvature	7.19	1	7.19	2.94	0.1301		C-Amphoteric Surfactant	0.022	1	0.02	0.0253	0.8789	
Residual	17.13	7	2.45				BC	12.88	1	12.88	14.77	0.0085	
Lack of Fit	16.15	5	3.23	6.58	0.1372		Curvature	3.61	1	3.61	4.14	0.0882	
Pure Error	0.9817	2	0.49			not significant	Residual	5.23	6	0.87			
Cor Total	81.11	10					Lack of Fit	4.94	4	1.23	8.37	0.1095	not significant
							Pure Error	0.2948	2	0.1474			
							Cor Total	22.42	10				

The reduced ANOVA for both week 0 and week 2 at 50°C were shown in the table above, the p-values of both models were less than 0.05 which shows the models are significant enough. Moving on for lack of fit and curvature, all p-values were greater than 0.05 which indicate that both curvature and lack of fit are not significant, the models were linear and sufficient to give the prediction of responses. Therefore, both week 0 and week 2 models were predictive.

4.4.3 Interpretation

Referring to the ANOVA tables above, factor 1, which is the amount of API isethionate, was only significant at 4°C. This was not logical because isethionate is an excellent foaming agent, as its amount in a body wash increases, the foaming stability should increase as well. Also, factor 3, amphoteric surfactant should be the most significant factor, because the concentration of surfactant significantly impacts the body wash's foaming ability and stability. However, the factor 3's p value at week 2 50°C is as high as 0.7362, which is also not logical. Not only that, factor 2, the amount of NaCl should be significant too, addition of a salt to a foaming solution will cause a significant reduction in foam volume over time (Behera, Varade, Ghosh, Paul & Negi, 2014). However, the rate of surfactant molecules adsorption at the air–water interface can be increased by salt, which then leads to a higher stability of foaming (Behera, Varade, Ghosh, Paul & Negi, 2014). Anyways, factor 2 was insignificant at week 2 50°C and week 2 25°C. The variation of factors' significance from expectation was caused by the low precision of foam height observation. In this study, the foam stability was calculated by the percentage of foam reduction in 1 minute. Therefore, scientists were required to observe and record the foam height twice, this process caused many deviations of foam stability from its actual value.

4.4.4 Conclusion

In conclusion, the experimental data in both week 0 and week 2 from temperature in 4°C, 25°C and 50°C showed that all factors and interactions were significant in the foam stability. However, since the methodology required human reading of the bubble volume instead of exact measurement. Therefore, the data of foam height was not accurate enough to calculate the reduction of foam stability, hence the actual stability of foaming. According to the limitation, the ANOVA model and significance of all factors and interactions were not reliable and thus cannot support the hypothesis.

4.5 Appearance

4.5.1 Evaluation

		Appearance		
	4°C	25°C	50°C	
EXP 1	Turbid	Transparent	Transparent	
EXP 2	Turbid	Transparent	Transparent	
EXP 3	Turbid	Transparent	Transparent	
EXP 4	Turbid	Turbid	Transparent	
EXP 5	Turbid	Transparent	Transparent	
EXP 6	Turbid	Transparent	Transparent	
EXP 7	Turbid	Transparent	Transparent	
EXP 8	Turbid	Turbid	Transparent	
cp1	Turbid	Transparent	Transparent	
cp2	Turbid	Transparent	Transparent	
cp3	Turbid	Transparent	Transparent	

Figure 58. Non-numerical results for appearance at 4°C, 25°C and 50°C

The results of appearance are formatted in a non-numerical manner, the appearance was transcribed in “turbid” or “transparent” according to the transparency of body wash formulation in each formulation.

4.5.2 Interpretation

Referring to the non-numerical graph above, there is a trend that can be found - body washes in 4°C are all turbid, in 25°C and 50°C are mainly transparent. This can be explained by the relationship between dissolution rate and kinetic energy. This body wash formulation is a viscous solution, where some solutes are dissolved in water, such as isethionate and aloe vera extract. These solutes' solubility changes over temperature; at 4°C, the lower kinetic energy causes some solutes to precipitate and lead to the turbidity of body wash. Oppositely, at 25°C or 50°C, the kinetic energy is high enough to allow complete dissolution of solute, and this makes the formulation transparent. The difference of transparency between 3 temperatures, are the outcome of kinetic energy impacts

4.5.3 Conclusion

To conclude, the non-numerical results of appearance exhibit the ANOVA test. The transparency of body wash varies at different temperatures, due to the impact of kinetic energy. The body wash formulations were transparent at room temperature and above, when the temperature decreased, formulations tended to become turbid.

4.6 Odour

4.6.1 Evaluation

The song graphs of odour vs. Experiments with error bars were attached below, all the raw data were accurately transcribed from experiment batch records. To rationalise the odours of each formulation, the results of odour were formatted in a score out of 10, which were given by scientists after sniffing the formulation. 1 stands for very unpleasant odour and 10 stands

for pleasant odour of body wash. The curved lined in song graphs stand for the significant factors and interactions' level at experiment 1 - 8. Refer to the graph of results for odour at 50°C, as there were 0 significant factors and interactions, no song graph was attached with the graph of result.

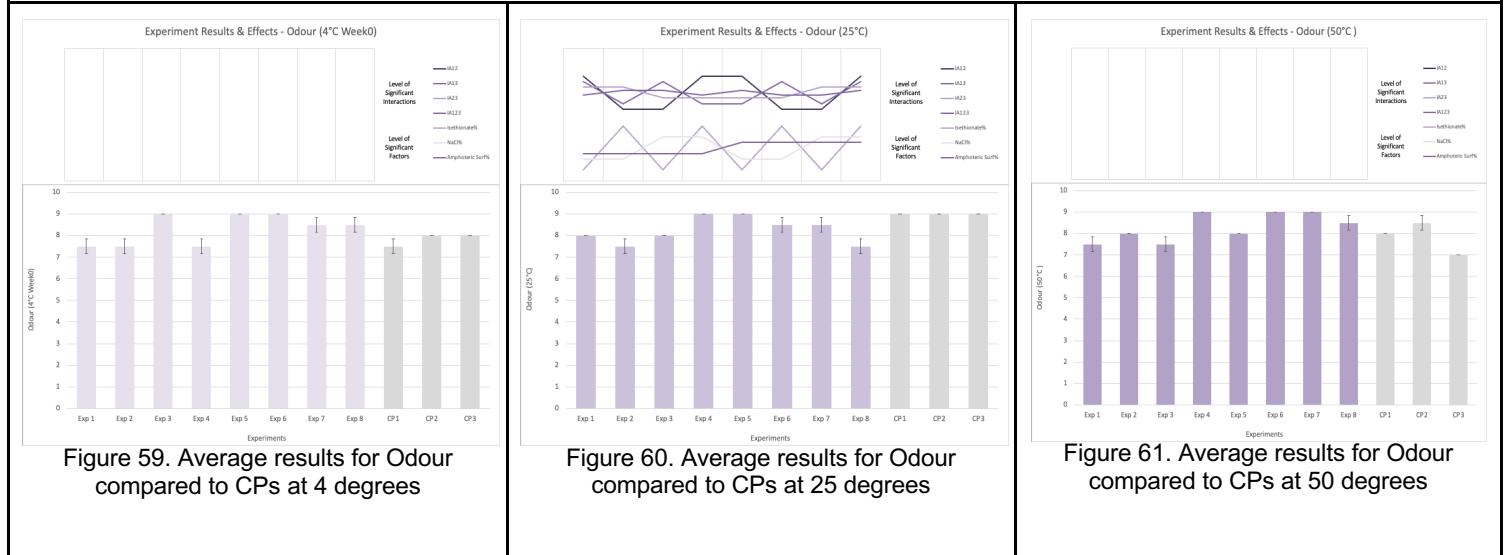


Figure 59. Average results for Odour compared to CPs at 4 degrees

Figure 60. Average results for Odour compared to CPs at 25 degrees

Figure 61. Average results for Odour compared to CPs at 50 degrees

4.6.2 Analysis

4.6.2.1 Effect of factor & interaction

The graphs of effects are attached below, the average results of experiments were attached at the left, the gradient effect of each factor/interaction was attached at the right. The two lines stand for the values of LOS- and LOS+, which were used to examine the significance of factors/interactions. However, as all the centre points' values were 9 at 25°C, therefore the two LOS lined in 25°C have a value of 0, overlapping at the x-axis.

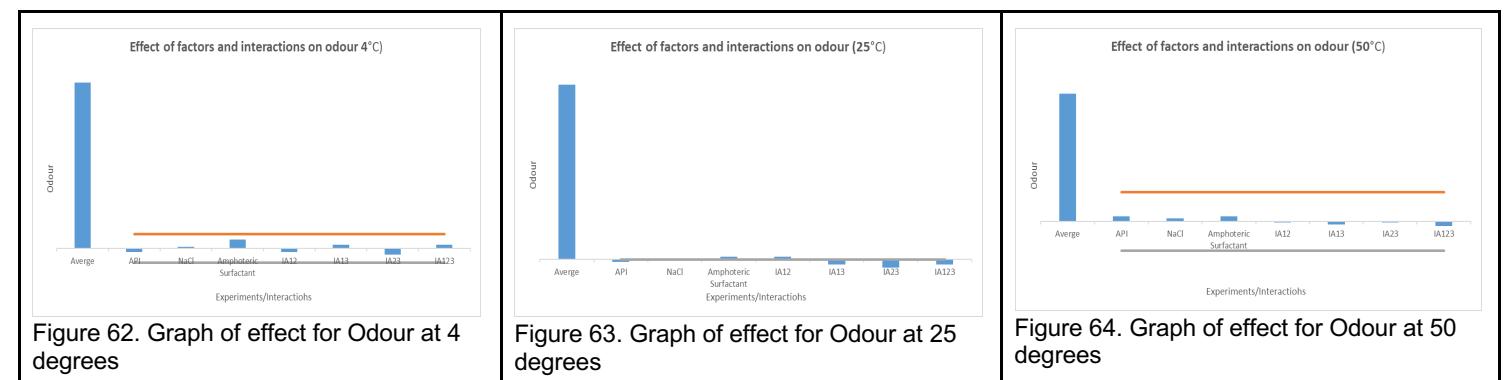


Figure 62. Graph of effect for Odour at 4 degrees

Figure 63. Graph of effect for Odour at 25 degrees

Figure 64. Graph of effect for Odour at 50 degrees

4.6.3 Interpretation

Referring to the graphs of gradient effects and song graphs above, the odour results in each temperature are varied, the significance of each factor and interaction were also different in 3 temperature levels. This can be proven by the LOS values in each temperature, at 4°C, the LOS+ value is 0.72, and it was 0 at 25°C and 1.90 at 50°C. Therefore, there were 0 significant factors or interactions at 4°C and 50°C, but all the factors and interactions were significant at 25°C. This can be explained by the fact that each individual experimenter has a varied sense of smell from others, for example, an experimenter may especially like the odour of a centre point formulation, but all the other experimenters considered the odour as unpleasant. Not only that, as the analysis of body wash odour was also conducted at different temperatures and times, it is more easily that the senses of smell of experimenters become impacted by other random factors.

Therefore, there was no need to conduct ANOVA tests. Using enough fragrance will give the body wash a pleasant odour.

4.6.4 Conclusion

To conclude, due to the uncertainty of senses of smell and impact of other random factors, the results of odour and the significance of factors/interactions in this study were distributed in a disordered manner. Therefore, there was no need to conduct ANOVA tests to verify the significance of the model.

4.7 Correlation analysis

In the Table 31 below, the correlations between different responses were shown in terms of correlation coefficient (r value). In total, there were 11 trials in the experiments and the t-value is 2.26. Then the R-value was calculated as 0.60, which means when the R-value between two responses was higher than 0.60 or lower than -0.60, the correlation between them can be considered as significant.

Therefore, the critical R-value was 0.60, and then a LOS value of 0.60 was applied as an indication of significant correlation in the table below, R-values that were greater than 0.55 are highlighted in red and less than -0.60 was in green.

Table 31. The significant correlations between the responses are highlighted in red and green below, with a selected R value of 0.60

	API	NaCl	Amphoteric Surfactant	Viscosity(4, Week 0)	Viscosity(4, Week 2)	Viscosity(25, Week 0)	Viscosity(25, Week 2)	Viscosity(50, Week 0)	Viscosity(50, Week 2)	pH(4, Week0)	pH(4, Week 2)	pH(25, Week 0)	pH(25, Week 2)	pH(50, Week 0)	pH(50, Week 2)	Foam Stability(4, Week 0)	Foam Stability(4, Week 2)	Foam Stability(25, Week 0)	Foam Stability(25, Week 2)	Foam Stability(50, Week 0)	Foam Stability(50, Week 2)	Odour(4)	Odour(25)	Odour(50)	
API	1.00																								
NaCl	0.00	1.00																							
Amphoteric Surfactant	0.00	0.00	1.00																						
Viscosity(4, Week 0)	0.01	0.40	0.54	1.00																					
Viscosity(4, Week 2)	0.00	0.38	0.54	0.99	1.00																				
Viscosity(25, Week 0)	-0.41	0.10	0.75	0.77	0.77	1.00																			
Viscosity(25, Week 2)	-0.18	0.15	0.57	0.83	0.84	0.83	1.00																		
Viscosity(50, Week 0)	0.08	0.03	0.45	0.80	0.82	0.60	0.72	1.00																	
Viscosity(50, Week 2)	0.06	0.03	0.42	0.80	0.83	0.60	0.72	1.00	1.00																
pH(4, Week0)	-0.37	-0.53	-0.16	-0.43	-0.40	-0.12	0.05	-0.01	-0.02	1.00															
pH(4, Week 2)	-0.26	-0.40	-0.61	-0.79	-0.77	-0.56	-0.48	-0.42	-0.41	0.80	1.00														
pH(25, Week 0)	-0.27	-0.60	-0.45	-0.74	-0.72	-0.46	-0.36	-0.33	-0.34	0.90	0.94	1.00													
pH(25, Week 2)	-0.25	-0.61	-0.49	-0.62	-0.61	-0.40	-0.26	-0.17	-0.17	0.90	0.92	0.97	1.00												
pH(50, Week 0)	0.10	-0.90	0.10	-0.35	-0.34	-0.11	-0.13	-0.15	-0.16	0.32	0.16	0.42	0.37	1.00											
pH(50, Week 2)	0.20	-0.14	0.30	0.14	0.10	-0.01	0.04	-0.02	-0.04	-0.18	-0.37	-0.15	-0.21	0.44	1.00										
Foam Stability(4, Week 0)	-0.39	-0.36	-0.39	-0.54	-0.53	-0.40	-0.54	-0.46	-0.45	0.16	0.33	0.40	0.32	0.44	0.17	1.00									
Foam Stability(4, Week 2)	-0.42	-0.40	-0.34	-0.50	-0.50	-0.34	-0.49	-0.42	-0.43	0.18	0.32	0.42	0.33	0.49	0.22	0.98	1.00								
Foam Stability(25, Week 0)	-0.23	-0.48	-0.51	-0.63	-0.62	-0.53	-0.53	-0.55	-0.54	0.25	0.42	0.51	0.44	0.59	0.23	0.93	0.91	1.00							
Foam Stability(25, Week 2)	-0.19	-0.22	0.71	0.11	0.12	0.55	0.14	-0.04	-0.04	-0.13	-0.30	-0.22	-0.30	0.23	0.20	-0.19	-0.15	-0.28	1.00						
Foam Stability(50, Week 0)	0.33	0.36	0.76	0.38	0.37	0.31	0.30	0.16	0.13	-0.40	-0.67	-0.58	-0.67	-0.09	0.44	-0.30	-0.30	-0.37	0.33	1.00					
Foam Stability(50, Week 2)	0.25	-0.17	0.03	-0.44	-0.44	-0.31	-0.54	-0.09	-0.12	0.07	0.30	0.20	0.18	-0.03	-0.41	0.02	0.00	-0.10	0.10	0.00	1.00				
Odour(4)	-0.30	0.00	0.60	0.11	0.10	0.35	0.10	0.21	0.18	0.21	-0.03	0.06	0.01	-0.07	0.33	-0.10	-0.06	-0.29	0.54	0.37	0.24	1.00			
Odour(25)	-0.33	0.00	0.00	0.37	0.40	0.52	0.38	0.34	0.37	-0.11	-0.13	-0.23	-0.11	-0.19	-0.69	-0.19	-0.18	-0.26	0.10	-0.40	-0.05	-0.37	1.00		
Odour(50)	0.43	0.43	0.43	0.27	0.21	0.17	0.15	0.12	0.05	-0.44	-0.45	-0.49	-0.48	-0.31	-0.05	-0.54	-0.50	-0.56	0.09	0.57	0.34	0.08	-0.07	1.00	

4.7.1 pH

From the full correlation table above, it was found that all the correlations between pH of body wash at 4°C and other responses were not significant at week 0. But 2 weeks later, it was highly correlated to the viscosity of week 0 and week 2 at 4°C with the correlation coefficient of -0.79, -0.77 respectively, Between the pH and viscosity under the same temperature, a highly significant correlation was presented with a negative correlation coefficient, indicating the pH at 4°C will be lower with the increasing viscosity.

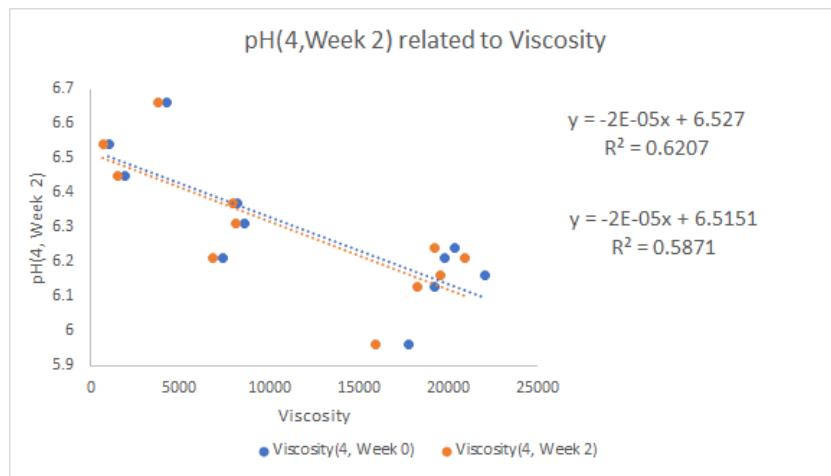


Figure 65: The significant correlation between pH of week 2 at 4°C and viscosity

For both pH of week 0 and week 2 at 25°C, they showed a highly significant correlation to the viscosity of week 0 and week 2 at 4 °C. That means the viscosity under that situation will also have a great and negative impact on the pH of week 0 and week 2 at 25°C.

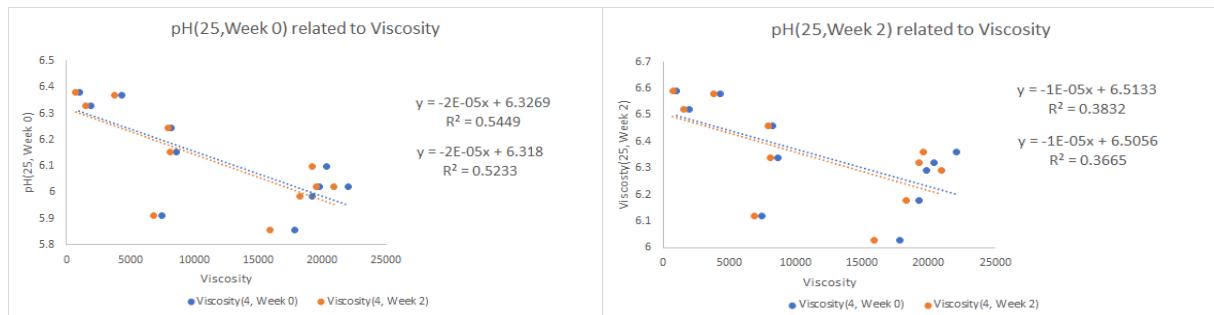


Figure 66: The significant correlation between pH of week 0 and 2 at 25 °C and viscosity.

4.7.2 Foam Stability

The foam stability of week 0 at 25°C is significantly correlated to the viscosity of week 0 and week 2 at 4 °C with the correlation coefficient of -0.63 and -0.62. The R-value showed the relation between these two responses was negative, which meant the foam stability of week 0 at 25 °C will decrease with the increasing viscosity.

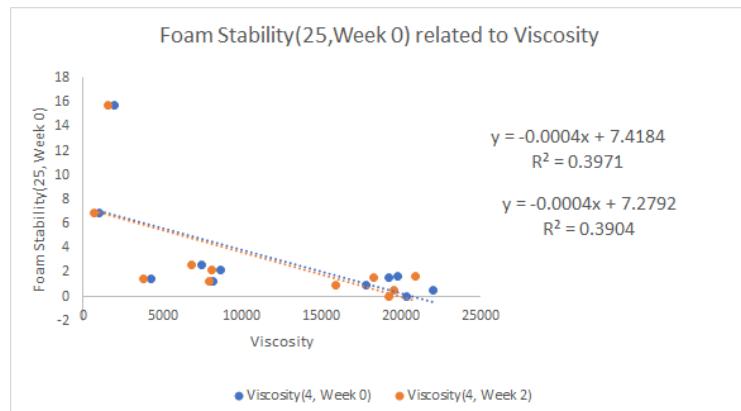


Figure 67: The significant correlation between foam stability of week 0 at 25°C and viscosity

When the temperature increases to 50 °C, the foam stability of week 0 was significantly correlated with pH of week 2 at 4°C and 25°C with R-value of -0.67 for both. The relationship between them was negative, indicating that the increasing pH can bring a negative effect on foam stability of week 0 at 50°C.

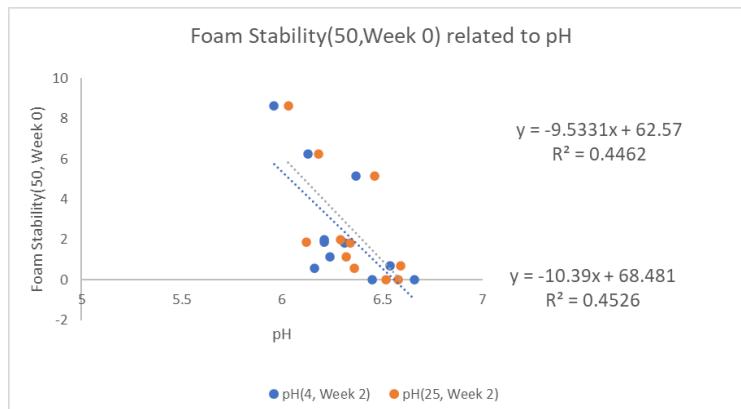


Figure 68: The significant correlation between foam stability of week 0 at 50°C and pH

4.7.3 Odour

The correlation between odour at 25°C and pH of week 2 at 50°C also showed a great significance with the correlation coefficient of -0.69. The relationship between them was negative, indicating that the increasing pH of week 2 at 50°C can decrease the odour level of the body wash.

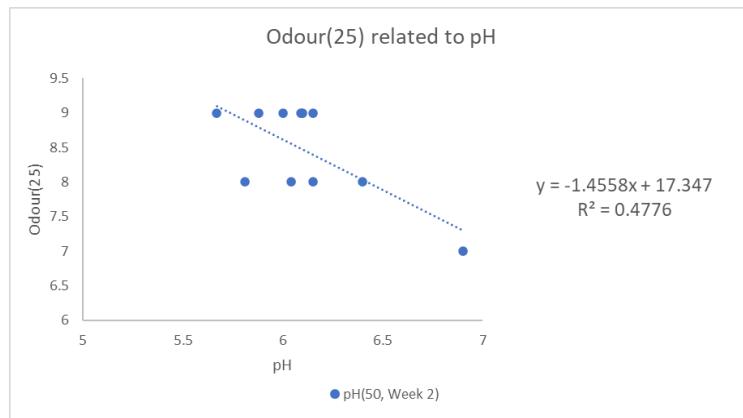


Figure 69: The significant correlation between odour at 50°C and pH

4.7.4 Interpretation

Among the significant correlations between the pH and viscosity, all of them showed an inversely proportional relationship, which meant that a high viscosity indicates a relatively low pH value. In a previous study, it was found that the quantity of sodium chloride shows a significant and negative effect on pH value of bodywash. At a higher pH environment, less sodium chloride is dissociated, and the surface charge is low. Therefore, the interaction between molecules was reduced, causing a lower viscosity.

The correlation between the foam stability (% reduction rate) and viscosity also showed a negative relationship. That meant the high viscosity of the liquid film that forms the bubble walls contributes to film durability and strength because the resistance to deformation was increased (A.K. Roy Choudhury, 2013). When the temperature rises to 50°C, the more sodium chloride has been dissociated, the increasing number of ions in the solution and the stability of foam can decrease because of Brownian motion. As mentioned before, increasing dissociation can reduce the pH value, thus the lower pH value of the body wash at 50°C, the foam produced by it will be less stable.

For the significant correlation between odour at 50°C and pH, currently, it is hard to find a theory to support the negative relationship. According to the full correlation table, it is obvious that only one significant correlation was presented, and others can be considered as not correlated due to the very low correlation coefficient. Therefore, the correlation between odour at 50°C and pH was recognized as a coincidence.

4.8 DoE full ANOVA table recap

In this project, we aimed to formulate a body wash with high viscosity (> 5000cP), a suitable pH (4.5-7.5), and a high foam ability stability over time. Therefore, the formulation needs to give us a high viscosity, suitable pH and high foam stability, i.e., maximise viscosity and foam stability. Overall, Isethionate Amount, NaCl Amount and Amphoteric Surfactant Amount all led to significant changes in most models.

Among three temperatures, 25°C (room temperature) is the most important model to be analysed as it can mimic the real-life situation. Isethionate content, NaCl content, and amphoteric surfactant content were a significant factor in most models, but NaCl content and Isethionate content cause minor effects to viscosity at 25°C, which may lead to a small but acceptable difference in pH. Meanwhile, the interactions between factors were commonly detected but mostly were interactions between two factors.

In terms of viscosity of the body wash, when increasing the Isethionate amount from 4% to 6%, the viscosity at 25°C decreases by an average of 558cP, amphoteric surfactant increases from 15% to 20%, the viscosity increases by an average of 1295cP, and no significant effect on changing the NaCl amount. Therefore, it is recommended that to maximise the viscosity, 4% Isethionate, high level of amphoteric surfactant, and most convenient level of NaCl (0%) were used in the formulation.

To the pH aspect, when NaCl increases from 0% to 5%, the pH decreases by an average of 0.3 hence the higher amount of NaCl required to achieve a suitable pH and decreases the potential harm caused by inappropriate pH. Likewise, Isethionate and amphoteric surfactant cause small changes in pH hence the levels of these two factors can be kept at 4% Isethionate and high levels of amphoteric surfactant.

As for the foam ability/stability, when NaCl% amount increases from 0% to 5%, the foam ability/stability decreases by an average of 2.87% reduction. Together with the negative correlation between foam ability/stability and viscosity, the higher the viscosity, the lower the percentage of foam reduction, which means the higher stability. Hence to maximise the foam ability/stability, the highest level of NaCl and amphoteric surfactant should be used (5% and 20%, respectively).

Since the model of viscosity is not predictable, the exact amount of amphoteric surfactant cannot be determined. The highest level (20%) amphoteric surfactant results in a semi-solid instead of a solution; the lowest level (15%) amphoteric surfactant results in a viscosity < 5000 cP. Additionally, pH and viscosity have a moderate negative correlation hence increasing viscosity significantly will be highly decreased. Therefore, 16.8% amphoteric surfactant is decided to be used in the final formulation after experimental practices.

In summary, 4% Isethionate, 2.5% NaCl and 16.8% amphoteric surfactant were decided to be used to form the final body wash, which yield with a viscosity of 5678 cP, 6.3 pH, transparent and pleasant smell body wash.

Table 32. The significant factor(s) and/or interaction(s), ANOVA p-values of factors and interactions were included.

	Intercept	A-API %	B-NaCl %	C-Amphoteric Surfactant %	AB	AC	BC	ABC
Viscosity WK 0 4	8568.75		3630.50	4898.75				
p-values			0.0010	0.0002				
Viscosity WK 0 25	2247.38	-366.875		676.875		-237.375		
p-values		0.0002		< 0.0001		0.0019		
Viscosity WK 0 50	474.875	48.625	16.375	294.625	-165.875	132.375	-55.875	-142.125
p-values		0.0912	0.4084	0.0029	0.0089	0.0139	0.0713	0.0121
Viscosity WK 2 4	7891		3320	4670.5				
p-values			0.001	0.0001				
Viscosity WK 2 25	1752.88	-191.125	166.125	618.125	-395.875	-237.875	330.875	
p-values		0.0159	0.0233	0.0005	0.002	0.0087	0.0034	
Viscosity WK 2 50	411.996	38.6712	18.6788	260.004	-141.346	113.329	-53.3288	-120.004
p-values		0.0362	0.1321	0.0008	0.0028	0.0044	0.0195	0.0039
pH WK 0 4	6.2075	-0.054375	-0.078125		-0.1			
p-values		0.0368	0.0085		0.0027			
pH WK 0 25	6.15375	-0.05625	-0.12375	-0.09375	-0.09125			
p-values		0.1116	0.0081	0.0236	0.026			
pH WK 0 50	5.98688		-0.159375	0.016875			0.060625	
p-values			< 0.0001	0.277			0.0051	
pH WK 2 4	6.32875	-0.05875	-0.08875	-0.13625	-0.09625			
p-values		0.1598	0.055	0.0123	0.0426			
pH WK 2 25	6.3525	-0.0525	-0.125	-0.1	-0.1			
p-values		0.1188	0.0065	0.0158	0.0158			
pH WK 2 50	6.1175	0.0725	-0.0525	0.11	0.1475	0.17	0.215	0.12
p-values		0.1152	0.1913	0.0553	0.0319	0.0243	0.0154	0.0471
Foam Stability WK 0 4	3.3175	-1.66	-1.54	-1.6875	1.5425	1.19	1.17	-1.6825
p-values		0.0397	0.0457	0.0385	0.0455	0.0732	0.0754	0.0387
Foam Stability WK 0 25	4.09		-2.429	-2.6065			2.215	
p-values			0.0405	0.0315			0.0553	
Foam Stability WK 0 50	3.05863		1.13413	2.41088				
p-values			0.0795	0.0033				
Foam Stability WK 2 4	2.85788	-1.67263	-1.57713	-1.37213	1.41238	1.13238	1.17288	-1.53263
p-values		0.0292	0.0327	0.0425	0.0403	0.0606	0.0569	0.0345
Foam Stability WK 2 25	3.66063	-0.918125	-1.09063	3.43312	1.80813	-1.14563	-1.31813	1.58062
p-values		0.0922	0.068	0.0075	0.0264	0.0622	0.0481	0.0342
Foam Stability WK 2 50	1.5075		-0.291	0.0525			-1.269	
p-values			0.412	0.8789			0.0085	

Table 33. The model significance, significant factor(s) and/or interaction(s), ANOVA p-values of models, curvatures, and lack of fits, predictivity and equations for predictable models were included.

Responses	Model significance	Significant Factor(s)	Significant IA	Curvature p-value	Lack of Fit p-value	Predictivity	Realistic Importance	Equation
Viscosity WK 0 4	0.0001	2,3		0.0001	0.2424	No	Medium	
Viscosity WK 0 25	< 0.0001	1,3	IA13	0.0001	0.4702	No	High	
Viscosity WK 0 50	0.011	1,2,3	IA12,IA13,IA23,IA123	0.0009		No	Medium	
Viscosity WK 2 4	0.0001	2,3		0.0001	0.179	No	Medium	
Viscosity WK 2 25	0.0019	1,2,3	IA12,IA13,IA23	0.0005	0.0919	No	High	
Viscosity WK 2 50	0.0033	1,2,3	IA12,IA13,IA23,IA123	0.0002		No	Medium	
pH WK 0 4	0.0032	1,2	IA12	0.5577	0.1609	Yes	Medium	pH (WK0,4) = 6.21-0.0544F1-0.0781F2-0.1IA12
pH WK 0 25	0.012	2,3	IA12	0.1132	0.1822	Yes	High	pH (WK0,25) = 6.15-0.0563F1-0.1238F2-0.0938F3-0.0912IA12
pH WK 0 50	0.0001	2	IA23	0.1335	0.341	Yes	Medium	pH (WK0,50) = 5.99-0.1594F1+0.0169F2+0.0606IA23
pH WK 2 4	0.0228	3	IA12	0.1252	0.0951	Yes	Medium	pH (WK2,4) = 6.33-0.0587F1-0.0888F2-0.1363F3-0.0963IA12
pH WK 2 25	0.0084	2,3	IA12	0.6087	0.1163	Yes	High	pH (WK2,25) = 6.35-0.0525F1-0.1250F2-0.1F3 -0.1IA12
pH WK 2 50	0.0379		IA12,IA13,IA23,IA123	0.5767		Yes	Medium	pH (WK2,50) = 6.12+0.0725F1-0.0525F2+0.11F3+0.1475IA12+0.17IA13+0.215IA23+0.12IA123
Foam Stability WK 0 4	0.0493	1,2,3	IA12,IA123	0.0516		Yes	Low	F.S (WK0,4) = 3.32-1.66F1-1.54F2-1.69F3+1.54IA12+1.19IA13+1.17IA23-1.68IA123
Foam Stability WK 0 25	0.0239	2,3		0.1907	0.0454	Yes	High	F.S (WK0,25) = 4.09-2.43F2-1.54F2-2.61F3
Foam Stability WK 0 50	0.006	3		0.1854	0.1036	Yes	Low	F.S (WK0,50) = 3.06+2.41F3
Foam Stability WK 2 4	0.0411	1,2,3	IA12,IA123	0.0511		Yes	Low	F.S (WK2,4) = 2.86-1.67F1-1.58F2-1.37F3+1.41IA12+1.13IA13+1.17IA23-1.53IA123
Foam Stability WK 2 25	0.0273	3	IA12,IA23,IA123	0.0893		Yes	High	F.S (WK2,25) = 3.66-0.9181F1-1.09F2+3.43F3+1.81IA12-1.15IA13-1.32IA23+1.58IA123
Foam Stability WK 2 50	0.0418		IA23	0.0882	0.1095	Yes	Low	F.S (WK2,50) = 1.51-0.2910F2+0.0525F3

4.9 Conclusion / Implication

Refer to the 3 hypotheses of this study:

- It can be hypothesised that by increasing the API (**Isethionate**) amount from 10% to 15%, the cleaning capability of body wash would increase as the **Isethionate** can help water to cling to the dirt and oil on your skin and allow it to be washed away; hence facilitate the cleaning capability.

As the cleaning capability of the body wash formulations were not tested, the veracity of this hypothesis was unknown.

- It can be hypothesised that by changing amphoteric surfactant amount from 15% to 20%, the viscosity and foaming stability of body wash will increase as amphoteric surfactant helps to decrease the surface tension; hence increase the viscosity and foaming stability.

According to section 4.1.1.1, the percentage amount of amphoteric surfactant had a positive effect on the viscosity of body wash at all 4°C, 25°C and 50°C conditions. However, due to the deviation in foam height deviation, the amount of amphoteric surfactant did not have a significant effect on foam height/stability at week 0 4°C and week 2 50°C. It was also illogical that this factor had a significantly negative effect at week 0 25°C and week 2 4°C. Therefore, this hypothesis was rejected.

- It can be hypothesised that if the amount of pH adjuster (NaCl) were to increase from 0 to 5%, the viscosity of body wash will increase as the NaCl increases the electrostatic attraction between the layers of water; hence increasing the viscosity.

At 4°C, the percentage amount of sodium chloride showed a significantly positive effect on viscosity of body wash. However, at 25°C and 50°C, this factor became less significant as the temperature increased. Thus, this hypothesis was rejected.

Table 34. The updated Relative Risk Ranking System

Formulation Factors	Product CQA				
	Physical appearance	Odour	Foaming stability	pH	Viscosity
API amount (%)			Medium	Low	Medium
pH adjuster amount (%)			Medium	High	Medium
Amphoteric surfactant amount (%)			High	Low	High

The risk levels of 3 factors API amount, NaCl amount and Amphoteric Surfactants amount were unknown, this is because ANOVA tests were not performed on the results of these 2 CQAs.

The risk levels of factors on foaming stability/ability remained consistent as the risk assessment system in protocol. When API amount and NaCl amount had medium - risk, and Amphoteric Surfactants amount have high - risk. This was because amphoteric surfactants decreased the surface tension between phase A and phase B, thus increasing the foaming properties of body wash formulation.

The risk levels of factors on pH also remain consistent, the amount of sodium chloride had a high risk of affecting pH of body wash. Refer to section 4.1.3, NaCl was used as a viscosity adjuster and a pH adjuster, therefore it showed a negative effect on pH value where the increased amount of sodium chloride will cause a low pH result.

For the risk levels on viscosity, the risk levels of 3 factors again remained consistent, as the risk assessment system in protocol.

At the end of this study, as the risk - levels of factors on formulation appearance and odour were unknown, the models of these 2 CQAs were not predictive as well. Therefore, the recommended formula was found by experimental practices. The composition of the recommended formula and its specifications were shown below.

Table 35. The final formulation of the Sukin body wash

Water	Q.S.
Isethionate	4%
Glycerine	2%
Aloe	0.2%
Amphoteric Surfactants	6.8%
Phenoxyethanol	0.9%
Benzyl Alcohol	0.3%
APG	4%
Fragrance	0.3%
NaCl	2.5%

Table 36. Comparison of Recommended formulation to Requirements

CQAs	Requirements	Recommended formulation
Viscosity	>5000 cP	5687 cP
pH	4.5 - 7.5	6.3
Skin feels	Moderate	Moderate
Appearance	Little colour - transparent	Transparent

5. Reference

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