Chapter Four - Materials and Methods

4.1. Introduction

The study focused on developing an appropriate headspace sampling technique as well as selecting optimal sensor array conditions and procedures to enable continuous monitoring in the laboratory whilst assessing the potential for an on-line field application at a drinking water abstraction facility.

4.2. Laboratory based development

4.2.1. Sensor array

Initial laboratory analysis was conducted using a modified eNOSE 5000 (Marconi Applied Technologies, UK). The commercial instrument contains an array of 12 conducting polymer (polypyrrole) sensors in a temperature controlled sensor chamber (35 °C). The 12 sensors used were types; 298, 401, 462, 463, 483, 501, 502, 503, 504, 505, 506, and 601. These sensors have a long lifetime over which they remain relatively stable. The sensors were grown electrochemically by Marconi Applied Technologies. Details on the exact fabrication and sensor components are confidential and were not available for reporting. However the actual sensor coatings were chosen as they each respond to a range of chemical standards hence assembling a versatile array of sensors that would provide a wide ranging non-specific analytical instrument (Marconi Applied Technologies, personal communication). Different sensors are responsive to different compound types.

4.2.1.1. Sample acquisition

The sampling cycle included pre-purge, sample acquisition, de-purge and idle. The times of which could be modified to suit different sampling methodologies. Filtered zero-grade nitrogen (N_2) (BOC, UK) was used to purge the sensors between each acquisition.

The pre-purge was utilised to flush the system of the previous gaseous sample and prepare the lines for the next. This also reduced the chances of potential cross-contamination between samples. During sample acquisition the sensors are exposed to the sample headspace. Their resulting change in resistance is recorded along with the values for the sampling parameters; gas flow, gas temperature, sample RH and sensor array temperature. After sampling valve states, within the array, switched to isolate the sensor chamber and allow a de-purge. A direct flow of N_2 gas is passed across the sensors for cleaning, allowing them to return to baseline ready for the cycle to repeat. The idle mode (reduced flow of N_2 across the sensors) was utilised so that the sampling frequency could be altered without changing the pre-purge, sample acquisition and de-purge sequence.

The sampling protocol for the sensor array analysis consisted of a 2-minute sensor pre-purge, a 1- minute sensor acquisition and a 4-minute de-purge (7-minute sampling cycle). Idle mode is not utilised in the standard operational mode but can be introduced if longer sampling intervals are required and gas supplies need to be preserved.

4.2.2. Headspace generation

The sampling system for experimentation consisted of a flow-cell chamber (Figure 4.2.2.1) for liquid headspace sampling, a sensor array module and a PC to allow remote system control and data processing and analysis. Controllable variables were incorporated into the design of the system to allow accurate control over a range of sampling parameters (Sample volume and temperature, sparger porosity and sparge gas flow rate). The flow cell temperature are controlled and regulated, by water jacket, to ±0.1 °C, using a Haake Dc50-K10 Heater-Cooler (Hakke, Germany). The sparge gas was delivered to the vessel using a sintered glass sparger (BDH, UK). The sparger was interchangeable to allow a range of sparge porosities. Sparging the partially filled flow-cell with zero-grade nitrogen generated the headspace samples. The resultant headspace was transferred to the sensor array module via a PTFE transfer line.

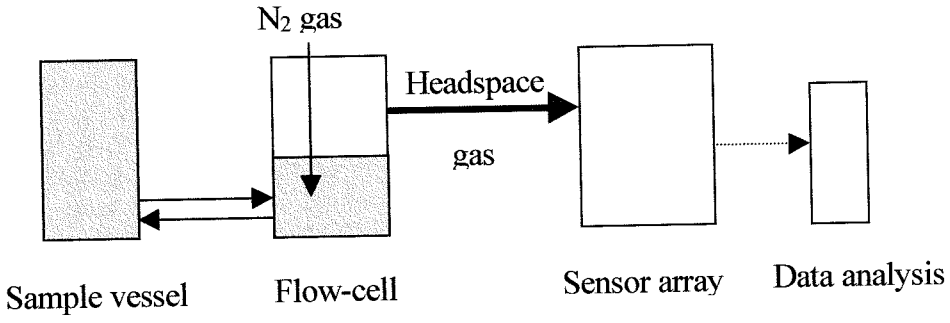

Figure 4.2.2.1 The sampling system for experimentation, consisting of a sample vessel, flow-cell chamber, sensor array module and a PC for data analysis/collection/control.

Preliminary laboratory assessments (Section 5.1.1) showed that this system had several instabilities in sensor response and RH generation (Figure 5.1.1.2). A series of modifications were made following the preliminary assessments; These were the installation of a temperature controlled regulator for the sensor array (and inlet and outlet fans to ensure temperature circulation and regulation), a temperature regulated sample transfer line (Astec, UK) and the application of an experimental design to assess optimal sampling conditions, this is introduced further in section 4.2.5.

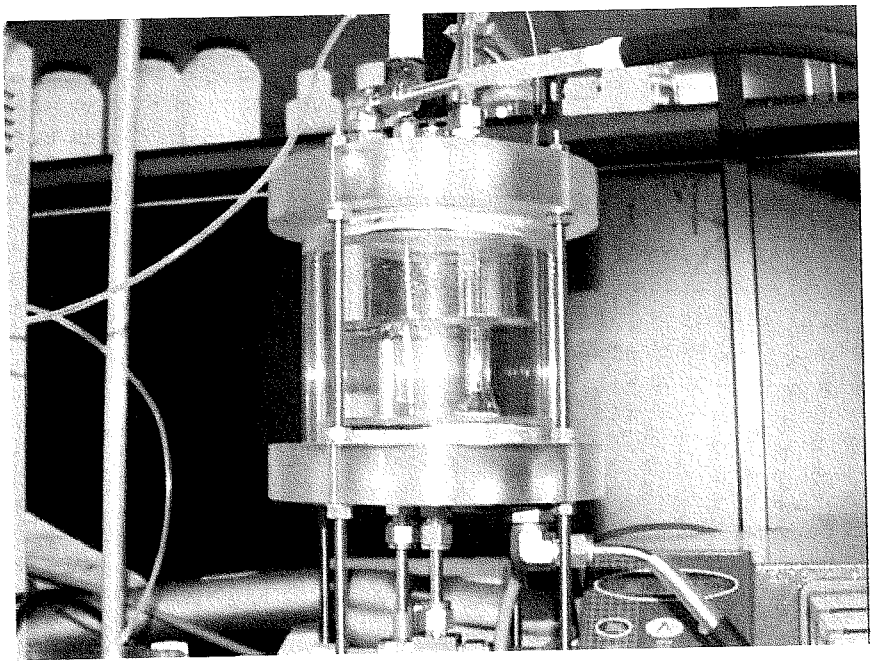

Figure 4.2.2.2 Photograph of the Flow-cell.

4.2.3. Sample preparation

RO water was obtained from an Elga Plus 15 RO unit (Elga, UK). Tainted water samples were prepared from stock solutions (stored at 5 °C) prior to experimental analysis by serial dilution for each of the pollutant concentrations.

Pollutant compounds (Table 5.2.1.1) were chosen from their use in previous trials and appearance in taste and odour related journal articles (Mallevialle and Suffet 1987, Young *et al.*, 1996, Drage *et al.*, 1998, Stuetz *et al.*, 1998, Suffet *et al.*, 1999). Detailed analysis using only two of the initial testing compounds, 2-Chlorophenol and Diesel, (Section 5.2.2) was conducted following their success in section 5.2.1.

4.2.4. Data mining

Each sampling cycle produces a raw data file. These files contain data values for each second of the acquisition and de-purge for each sensor in the array plus gas flow, gas temperature, sample RH and sensor array temperature. These files can be mined at a selected time point to determine the change in sensor resistance (% $\Delta R/R$) for each sensor and characterise the headspace sample (Figure 4.2.4.1). The mined data is received in an Excel document where further data analysis can take place (Section 4.5). Plots of the sensor responses (Figure 4.2.4.2) were constructed using data abstracted from large data sets selected using a macro at a one-minute time slice from all sampling runs. Each point on every graph represents the sixtieth second of the one minute sampling window when the profile headspace sample is generated. This method has been implemented before (Stuetz *et al.*, 1998). The time elapsed between each point is not shown and can depend on the time intervals built into the sampling cycle. The sensor response plots provided a simple visual indication of any rapid change in the quality of the water being assessed.

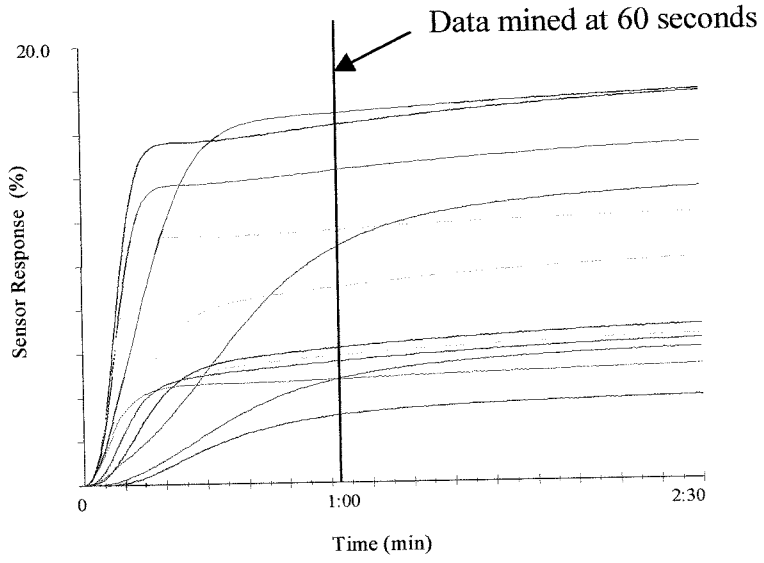

Figure 4.2.4.1 A response pattern generated by a chemical sensor array showing the sensor response (%) change verses time and the point at which data is mined to produce the representative pattern profile.

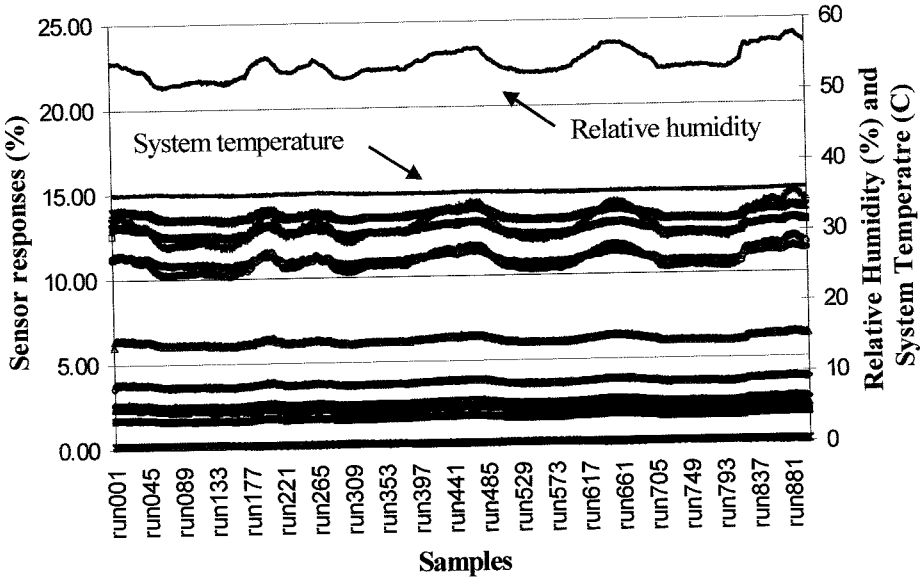

Figure 4.2.4.2 Graphical representation of the sensor responses, RH and system temperature over a period of 900 runs (105 hours continuous analysis).

4.2.5. Experimental design

A two level full factorial design was used to determine the optimal sampling parameters and to see what combination of temperature, flow rate and porosity, generated a more stable RH and consequently a more reproducible sensor response. The three variables were studied at two levels (high and low) which resulted in carrying out $2^3 = 8$ experiments.

Following modification and implementation of the experimental design results (Section 4.4.3) the system was evaluated for its potential to detect a pollutant introduced to the liquid phase. Sampling parameters were kept at constant values as were the headspace transfer line temperature (30 °C), and sensor module temperature (35 °C).

4.2.6. Pollutant introduction

Initial pollution introduction was achieved by direct injection into the flow-cell during pre-purge and just prior too sample acquisition. Injecting the flow-cell to soon would result in the pollutant being flushed away before the acquisition stage begun. Once pollutant detection has been established (Figure 5.1.4.1) the pollutant introduction procedure was modified to provide a more realistic pollution occurrence scenario. Rather than direct flow-cell spiking the supply waters for the flow-cell were spiked so that the polluted water would have to travel through the flow-cell rather then just being flushed out of the flow-cell. This produced sustained periods of sensor recognition.

4.3. Field based development

To test the technology under more realistic and variable conditions, field based studies at an operational water monitoring station were undertaken. Field based trials were conducted over several months, initial studies consisted of continuous monitoring of the abstraction water from the River Trent followed by spiking trials using 2-chlorophenol.

4.3.1. Sensor array

A ProSAT (Marconi Applied Technologies) was used for field operations. The system is very similar to the eNose 5000 used in laboratory analysis and was designed for a range of on-line applications. The instrument consists of 8 conducting polymer sensors in a temperature controlled sensor chamber (35 °C) and has an inbuilt PC for control and data acquisition/storage as well as a network connection for data transfer and remote operation. Unit operation and configuration is set up by menu-driven software. An inbuilt screen acts as PC monitor would enable visual monitoring of the sensor response profiles, sensor baselines, system and sampling parameters.

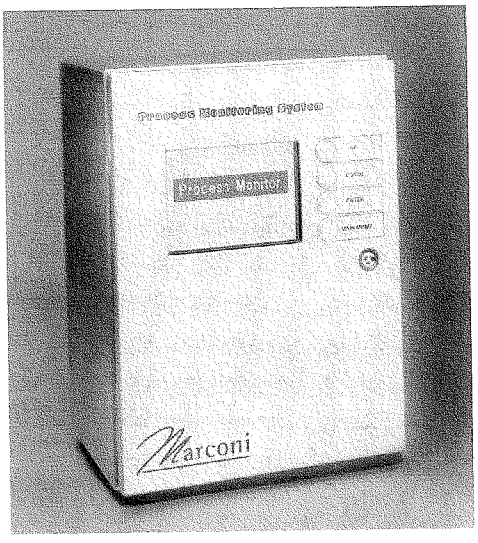

Figure 4.3.1.1 Photograph of ProSAT on-line process monitoring system (Courtesy of Marconi Applied Technologies, UK).

The choice of sensors for the ProSAT was based on the results of our initial laboratory using the eNose 5000. Sensors 501, 502 503 and 504 were selected as they were the only four sensors to respond to the range of chemicals tested. Sensors 298, 401, 462 and 506 were selected by Marconi Applied Technologies following discussions and recommendations based upon the stability and suitability of the sensors for this purpose.

4.3.2. System set up

The on-line system was constructed to mimic the laboratory apparatus and procedures (Figures 4.3.2.1 and 4.3.2.2). As the pilot system was remote the sampling protocol was amended so that sensor acquisitions occurred for sixty seconds in a 15-minute protocol; the sixtieth second being used to construct data plots as previously described (Section 4.2.4). Unlike previously described laboratory procedures the 'Idle period' option was utilised for 8-minutes in the 15-minute period to conserve gas consumption.

After analysis of background temperature variation on the River Trent it was decided that no temperature control would be incorporated. Although laboratory studies showed that sample temperatures played a significant role in promoting volatiles from solution into the headspace sample in a field application attempting to apply tight control over such a variable would be costly and difficult to implement in a changing environment. River Trent data showed that temperature changes in the river were never sudden and at most only changed by one degree in 24 hours or two degrees in 48 hours (Figures 5.3.1.1 and 5.3.1.2). Small windows of data were selected for viewing so only small changes would be expected during the course of simulated spiking events.

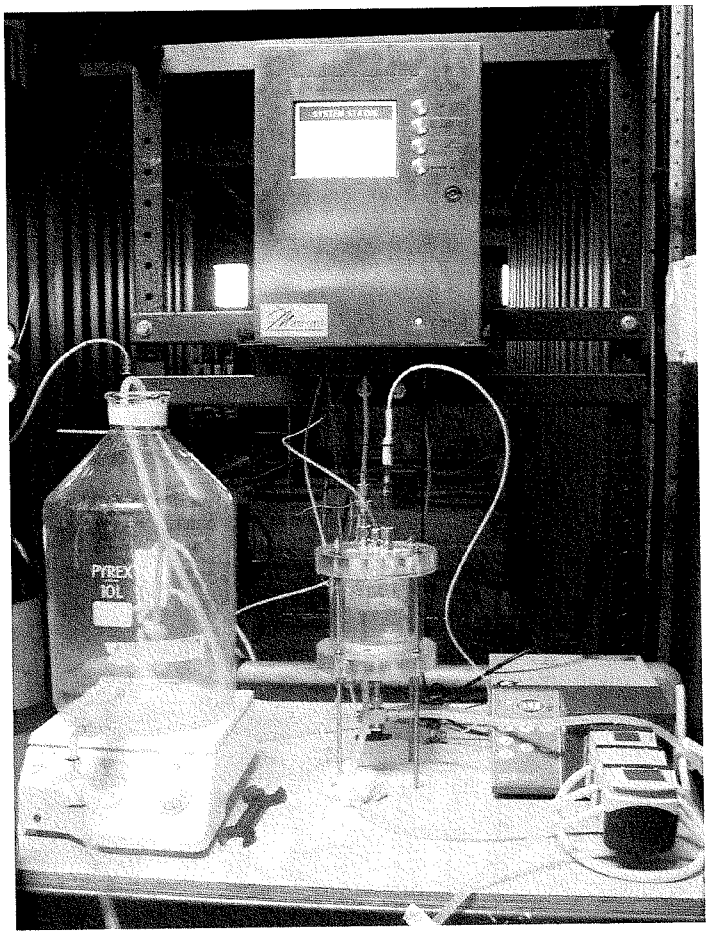

Figure 4.3.2.1 Photograph of the ProSAT and Flow-cell at the River Trent monitoring station

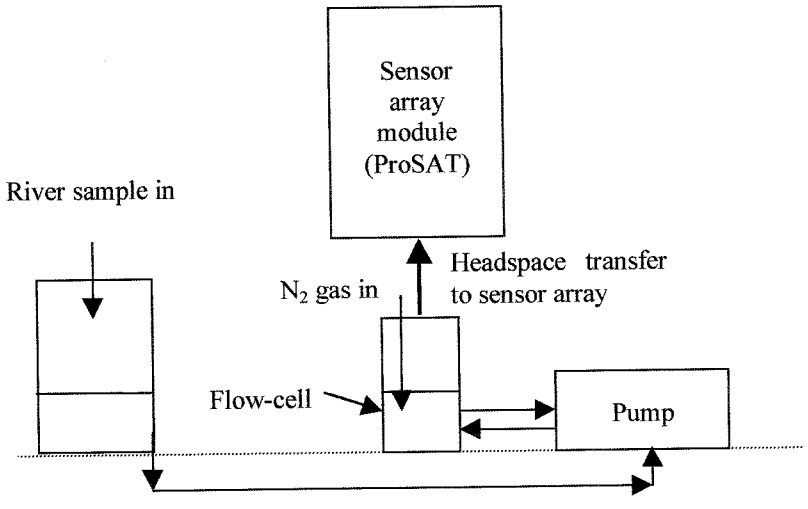

Figure 4.3.2.1 Schematic of the ProSAT and Flow-cell at the River Trent monitoring station

4.3.3. Pollutant introduction

Initial spiking events on-line produced false positives due to temperature differences (between the abstracted water and the spiked sample) whilst introducing test pollution events. In order to minimise these instabilities a system modification was proposed (described in section 5.3.3) although unfortunately the adapted system remained untested before the conclusion of the project.

4.4. Data analysis

4.4.1. Graphical representation

Plots of the sensor responses were used to monitor for changes in sensor resistance (Section 4.2.4). The plots were used as a simple visual indication of any rapid change in the water quality due to the injection or introduction of a tainting compound into the flow-cell or sample vessel, respectively.

4.4.2. Statistical analysis

Pattern recognition techniques were also used to reduce the dimensionality of the sensor array data, so that relationships between the observations could be explored using one or two dimensions. The statistical package UNISTAT was used to compare and correlate the sensor array data and produce principal component analysis (PCA) plots. PCA, a non-supervised linear pattern recognition technique, was utilised to reduce the dimensionality of the sensor array data so that relationships between observations could be explored. Plots of these data should exhibit grouping of odour types and concentration if trends exist in the data being assessed.

4.4.3. Experimental design

A two level full factorial design was used to determine the optimal sampling parameters and to see what combination of temperature, flow rate and porosity, generated a more stable RH and consequently a more reproducible sensor response. The three variables were studied at two levels (high and low). For a process with three variables being studied and two levels, the required number of experiments is $2^3 = 8$ experiments (Table 4.2.5.1)

Table 4.2.5.1 Experimental design matrix

Run	Variable 1	Variable 2	Variable 3	Response
1	-	-	-	?
2	+	-	-	?
3	-	+	-	?
4	+	+	-	?
5	-	-	+	?
6	+	-	+	?
7	-	+	+	?
8	+	+	+	?

The whole procedure was repeated on three different days, using reverse osmosis (RO) water, to ensure that the results were significant and that the choice of a method for sensor array analysis was based on a model that was considered to be as robust as possible.

4.4.3.1. Main effects

The results from this design can be manipulated to give a ranked average effect for each variable giving us a numerical representation of which is the most significant and least significant variable. The average contribution of each variable is found by considering each variable separately. The average result for all the runs where each variable is at the low level is found by summing and dividing by four. This is then repeated using runs where the variable is at the high level. The average effect of each variable is then found

by calculating the difference between the high and low contributions. It is the magnitude of the effect that determines its contribution.

4.4.3.2. Variable interactions

The main point of a factorial experiment is to measure interactions between the variables and use the data to predict measurements at other variable levels. This enables observations to be made as to whether one variable or factor is effecting another and to what degree.

The design results are used to construct a mathematical model called the 'response equation'. This relates to the variable levels (in coded units) to the response result. This can be used to predict the result for any value of the variables subject to them being continuous and within the experimental area.

The response equation contains coefficients that are calculated from the results. The coefficients are either main coefficients or interaction coefficients. All coefficients are labeled A with a subscript to define them.

e.g. A_1 is the main effect coefficient of V1
A_2 is the main effect coefficient of V2
A_{12} is the main effect coefficient of V1V2 etc.

The coefficients are calculated by the method of 'contrast patterns' which are generated by the design matrix. The contrast pattern matrix is shown in Table 4.4.3.2.1. Where V1, V2 and V3 are main effect coefficients and V1V2, V1V3, V2V3 and V1V2V3 are the interaction coefficients. The pattern of + and – for the interaction columns are generated by simple multiplication of the relevant variables sign.

e.g. V1V2 = -1 x –1 = +1
V1V3 = -1 x –1 = +1
V1V2V3 = -1 x –1 x –1 = -1

This is repeated for each line in the contrast pattern matrix

Table 4.4.3.2.1 Contrast pattern matrix

Run	V1	V2	V3	V1V2	V1V3	V2V3	V1V2V3	Result
1	-1	-1	-1	+1	+1	+1	-1	?
2	+1	-1	-1	-1	-1	+1	+1	?
3	-1	+1	-1	-1	+1	-1	+1	?
4	+1	+1	-1	+1	-1	-1	-1	?
5	-1	-1	+1	+1	-1	-1	+1	?
6	+1	-1	+1	-1	+1	-1	-1	?
7	-1	+1	+1	-1	-1	+1	-1	?
8	+1	+1	+1	+1	+1	+1	+1	?

V1 = Gas flow rate, V2 = Sample temperature and V3 = Sample concentration.

The coefficients are found by summing the results, column by column, using the columns contrast pattern and then dividing by eight (the number of runs). The final coefficient is A_0, the average of all the results. The response equation for a D8/3 design is $RH = A_0 + A_1.V1 + A_2.V2 + A_3.V3 + A_{12}.V1.V2 + A_{13}.V1.V3 + A_{23}.V2.V3 + A_{123}.\ V1.V2.V3$. The response equation was derived using coded variables and therefore, coded valuables are used in its application. Where A1, A2 etc are the interaction coefficients and V1, V2 and V3 are the coded values for their respective variables. These are determined, assuming a linear relationship, in the form of a $y = mx + c$ equation where y is the coefficient required. The response equation can be used along with coded variables for unknown values to predict responses.

Data from the expanded studies using 2-chloropehnol and diesel were reincorporated into the experimental design to test its ability to predict the change in sensor resistance under known parametric changes including sample concentration, sparge gas flow rate and sample temperature. The results of which are presented in Results section 5.4.2.

4.4.3.2.1. Significance

It is assumed that the three-way interaction is extremely improbable (V1V2V3) and therefore represents the standard error of the system hence the use of A_{123} as the standard error term to test the significance of the others. The significance of each interaction coefficient is calculated by dividing each separate coefficient by the value represented by coefficient A123 the value calculated for the interactions between all three components. The resultant value is considered to be statistically significant if greater than 2.