

# **DETERMINATION OF METHOD UNCERTAINTY A PRACTICAL EXAMPLE FOR THE ANALYSIS OF TRIHALOMETHANES IN DRINKING WATER USING PURGE&TRAP GC-MS**

Speaker / Author: R. Avis  
Co-author: E. Mrozek  
Johannesburg Water  
PO Box 61542, Johannesburg 2107, South Africa  
e-mail: ravis@jwater.co.za  
Phone: 011 728 7373 Fax: 011 728 5444

## **Abstract**

Johannesburg water is responsible for the supply and monitoring of drinking water supplied to the greater Johannesburg area. Part of this monitoring programme involves the determination of Trihalomethane (THM) levels and the comparison thereof against the limits stipulated in SANS 241. This paper demonstrates the approach used to determine the uncertainty of the analytical method associated with the determination of THM's, at ug/L levels, in drinking water using Purge and Trap (P&T) GC-MS. It indicates the five steps used namely:

1. Specification of the method
2. Identifying the model
3. Identification of sources of uncertainty using the Ishikawa approach
4. Expanding and if needed combining the uncertainties of each of the chosen data sources
5. Combining the expanded uncertainties and expressing the final method uncertainty

The paper also attempts an evaluation of the data obtained and recommends possible improvements, identified from the data obtained, which could be implemented to improve the uncertainty of the analytical method.

## **1. Introduction**

A requirement of method validation in the ISO 17025 standard is the availability of an uncertainty budget. There are specific means of determining this and the laboratory decided on the GUM approach using the approximation method to calculate the sensitivity coefficients. It was decided to evaluate this approach initially on the THM method. If this was successful the rest of the methods used in the laboratory would follow a broadly similar approach.

## **2. Uncertainty determination:**

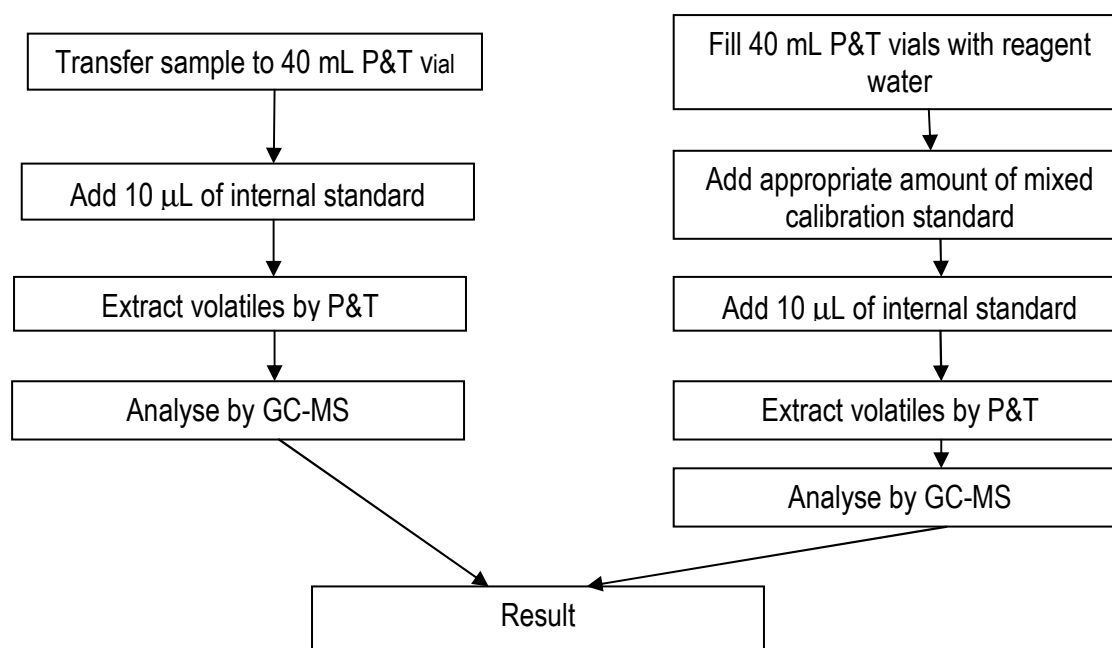
**Step 1.** Writing down the method specification.

The method specification must contain a full description of the method including measurand, parameters, matrix and equipment used. It must be of such a nature that it prompts one to consider any parameters that might influence the method uncertainty. A method statement and flow diagram as well as a description of the analytical procedure all assist in this step.

**The method statement:**

The determination of THM (measurand) in drinking water (matrix), using an internal standard method with purge and trap GC-MS (method). The method range is 1-100 ug/L.

As an example of obtaining information from the above statement, one can realise that the calibration equation will not be the usual straightforward linear equation, but rather the internal standard ratio calculation used in the instrument software. In addition all the factors of the purge and trap system would have to be evaluated.

**Method flow diagram:****The analytical procedure followed in this method:**

1. Drinking water samples, from various points, are taken in 500ml glass bottles and preserved with ascorbic acid. The bottles are filled completely to eliminate headspace and stored in a fridge until analyses.
2. A 40ml aliquot of the sample, QA or calibration standard is transferred to a glass vial.
3. A known amount of a certified internal standard is added with a syringe and the vial is then sealed.
4. Multipoint (five point) calibration curves are generated using standards prepared from certified stock solutions and transferred using a 10µL as well as a 50µL syringe.
5. Reagent water containing the internal standard is analyzed before calibration and again after the highest standard to check for carryover issues.
6. The QA sample is then analyzed using a purge and trap sampler coupled to a GC-MS system, followed by samples and a new QA every 10 samples thereafter. The analysis is completed with a final QA sample. The retention time and peak area of selected target ions are then determined.
7. A final result is calculated and reported

## Step 2. The model

MODEL :  $C_{\text{analyte}} = D \cdot I_{\text{analyte}} \cdot C_{\text{int.std.}} / A \cdot I_{\text{int.std}}$

Where:

- $C_{\text{analyte}}$  = the concentration of the analyte (an individual trihalomethane)
- D = the dilution factor
- $I_{\text{analyte}}$  = the peak area of the analyte
- $C_{\text{int.std.}}$  = the concentration of the internal standard
- A = the slope of the calibration curve
- $I_{\text{int.std}}$  = the peak area of the internal standard

## Step 3. Identifying the sources of uncertainty.

In this step the Ishikawa, popularly known as the fishbone, diagram is used.

The fishbone is a two stage process. In stage one all possible sources are added to the fishbone, as indicated in fig. 1 below

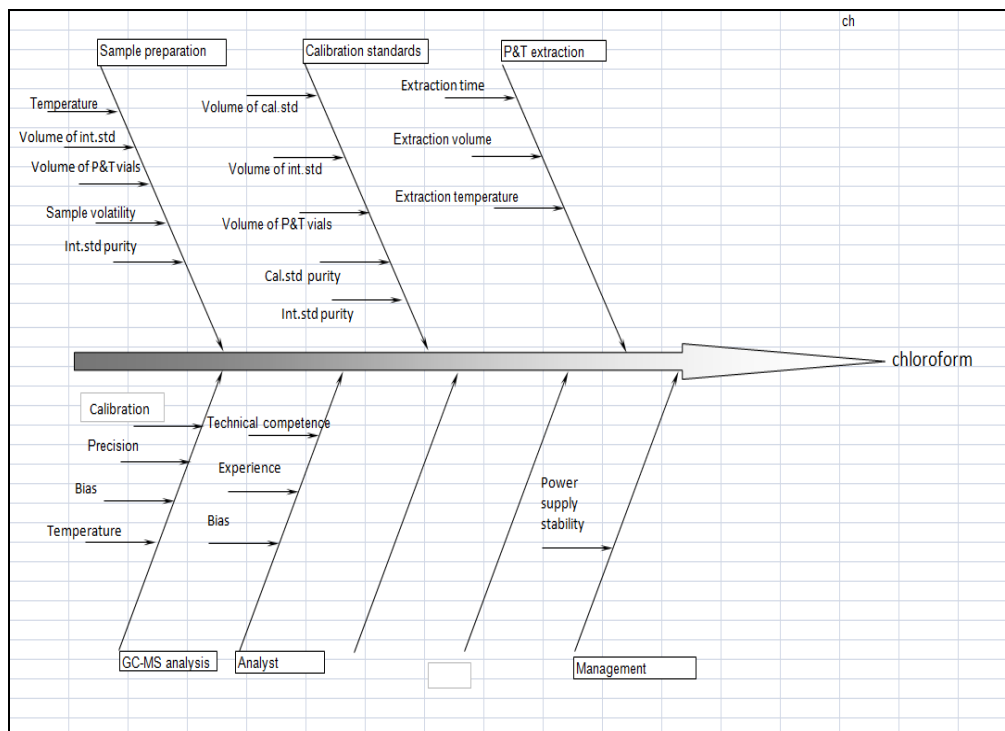


Fig. 1 possible uncertainty sources of chloroform. Similar diagrams exist for the other THM compounds

In stage 2 we simplify the fishbone and obtained fig. 2. The reasons for the simplifications are:

1. All instrument related parameters would be included in either the calibration or precision bone.
2. All glassware related parameters would be included in the precision bone except vial and syringe
3. The extraction is the same for standards, quality controls (QA) and samples and will be included in the precision data as is any analyst variation.

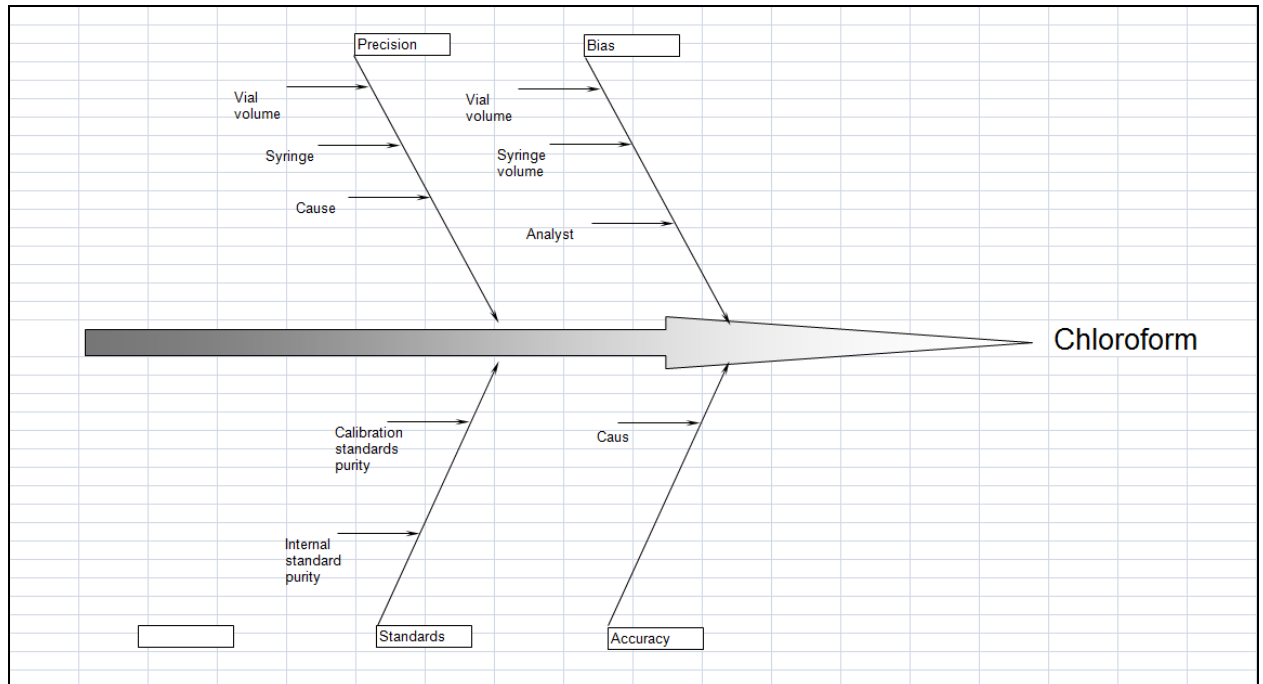


Fig. 2 simplified uncertainty sources of chloroform. Similar diagrams exist for the other THM compounds

The resultant fishbone indicated four major sources of uncertainty: (i) method precision including reproducibility and repeatability, (ii) method bias, (iii) accuracy and (iv) Calibration standards.

The possible sources of data for the four identified sources are method validation data, calibration data and QA data.

**Step 4.** Calculating the expanded uncertainties of the parameters identified in step 3.

*The model:*

The vial volumes were determined by statistical observation. Fifteen replicates were used and the standard uncertainty was calculated as the standard deviation divided by  $\sqrt{15}$ .

The standard uncertainty of the syringe was calculated from the manufactures data assuming a rectangular distribution and thus dividing the standard deviation by  $\sqrt{3}$

The standard uncertainty of the injection volume was calculated from the manufacturer supplied data assuming a rectangular distribution.

The detector response was obtained from historical data

*Method precision:*

Replicates of the method QA data were obtained from historical records. A best estimate and a standard deviation were calculated.

The standard uncertainty was the calculated as the standard deviation divided by  $\sqrt{n}$

The data obtained from the duplicate sample that was analysed during each run was utilised as a data source for method bias. A best estimate and a standard deviation were calculated. The data was normalised by means of the population standard deviation technique and relative standard deviation values were obtained which were used for final calculations.

The standard uncertainty was the calculated as the standard deviation divided by  $\sqrt{n}$

*Accuracy - Regression:*

Calibration data from the previous 36 calibrations was obtained. A best estimate and standard deviation was calculated for the slope of the calibration curve.

The standard uncertainty was calculated as the standard deviation divided by  $\sqrt{36}$

*Method bias:*

For the standards a best estimate and standard deviation was obtained from the certificates supplied. The best estimate was 100 with a standard deviation of 5. The standard uncertainty for each THM and the internal standard was calculated as the standard deviation divided by  $\sqrt{3}$ .

The data of 15 spikes, from the method validation document, was used to calculate the recovery standard uncertainty

The final stage of this process was to combine the standard uncertainty of each data source for each bone on the diagram. In order to do this the sensitivity coefficients had to be calculated using the method of approximation.

The model uncertainty for one component (chloroform) is shown below as fig. 3b and indicates the calculation of sensitivity coefficients fig. 3a.

MODEL	$C_{\text{analyte}} = D \cdot I_{\text{analyte}} \cdot C_{\text{int.std.}} / A \cdot I_{\text{int.std}}$			power	%change
				1	1
	$C_{\text{analyte}} =$	25.96		2	0.1
				-1	0.1
CHLOROFORM				-2	0.01

Parameter	best est.	%		$\Delta$	Y	Y'	$\Delta Y = Y' - Y$	$C_i$
$I_{\text{analyte}}$	10442485	1	0.01	104424.9	25.96126740	26.22088008	0.25961267	2.48612E-06
$C_{\text{int.std.}}$	25	1	0.01	0.25	25.96126740	26.22088008	0.25961267	1.038450696
A	2.834953	0.1	0.001	0.002835	25.96126740	25.93533207	-0.02593533	-9.148416947
$I_{\text{int.std}}$	3326815	0.1	0.001	3326.815	25.96126740	25.93533207	-0.02593533	-7.79584E-06
D	0.9379	1	0.01	0.009379	25.96126740	26.22088008	0.25961267	27.68020834

Fig. 3a sensitivity coefficients calculation of chloroform

Parameter	Units	Best estimate	Half-range	Probability distribution	Divisor	Standard uncertainty, $u_i$	Sensitivity coefficient, $c_i$	expanded uncertainty, $(c_i \cdot u_i)$	Uncertainty contribution, $(c_i \cdot u_i)^2$
CHLOROFORM $C_{\text{analyte}} = D \cdot I_{\text{analyte}} \cdot C_{\text{int.std.}} / A \cdot I_{\text{int.std}} = 25.96$									
$I_{\text{analyte}}$									
injection volume (P&T)	mL	25	0.25	rectangular	1.732050808	0.144337567	2.48612E-06	3.5884E-07	1.28766E-13
detector response	counts	10442485	3290470	normal	5.916079783	556190.9441	2.48612E-06	1.382757249	1.912017608
$C_{\text{int.std.}}$									
certificate, $\mu\text{g}/\mu\text{L}$	$\text{ng}/\mu\text{L}$	100	5	rectangular	1.732050808	2.886751346	1.038450696	2.997748945	8.986498734
syringe volume, $\mu\text{L}$	$\mu\text{L}$	10	0.1	rectangular	1.732050808	0.057735027	1.038450696	0.059954979	0.003594599
P&T vial volume, mL	mL	42.6	0.2347	normal	3.872983346	0.060599279	1.038450696	0.062929364	0.003960105
D	-	0.9379	0.0051585	normal	3.872983346	0.001331906	27.68020834	0.036867437	0.001359208
A	-	2.834953	0.412902	normal	6	0.068817	-9.148416947	-0.629566609	0.396354115
$I_{\text{int.std}}$									
injection volume, mL	mL	25	0.25	rectangular	1.732050808	0.144337567	-7.79584E-06	-1.12523E-06	1.26615E-12
detector response	counts	3326815	270022	normal	5.916079783	45642.04843	-7.79584E-06	-0.355818307	0.126606668
Root of sum of squares $u_c(y) = \sqrt{\sum (c_i \cdot u_i)^2}$								3.380886132	

Fig. 3b model uncertainty for chloroform

The calculation of uncertainty due to bias is shown in fig.4 below

Bias Uncertainty							
Parameter	Units	Best estimate	Half-range	Probability distribution	Divisor	Standard uncertainty, $u_i$	$U_{bias}$
<b>Analytes</b>							
Chloroform	µg/L	26.0233	1.004061	normal	3.872983	0.259247384	<b>1.53019</b>
Dichlorobromo	µg/L	25.2547	0.929161	normal	3.872983	0.239908448	<b>1.48212</b>
Dibromochloro	µg/L	24.6093	0.969178	normal	3.872983	0.250240773	<b>1.50753</b>
Bromoform	µg/L	24.0473	1.067463	normal	3.872983	0.275617863	<b>1.57252</b>

Fig. 4 calculation of bias uncertainty

The calculation of uncertainty due to precision repeatability is shown in fig.5 below

	CHCl3	CHCl2Br	CHClBr2	CHBr3
RSD Value	0.127217	0.115633	0.08393	0.083189
RSD Value	0.028024	0.016525	0.02746	0.124054
RSD Value	0.006107	0.000448	0.003424	0.064282
RSD Value	0.06834	0.054092	0.044249	0.186783
RSD Value	0.03151	0.018837	0.017493	0.094281
RSD Value	0.009684	0.009001	0.016381	0.208654
RSD Value	0.016675	0.020448	0.043676	0.132583
RSD Value	0.005287	0.006983	0.010216	0.022448
RSD Value	0.11647	0.126282	0.118751	0.202031
RSD Value	0.048843	0.067344	0.119444	0.749818
RSD Value	0.068669	0.053942	0.038166	0.058926
RSD Value	0.040946	0.044151	0.086994	0.471405
mean RSD	0.047314	0.044474	0.050849	0.199871
mean	33.4075	17.90333	5.0825	0.382083
ΣRSD	0.567773	0.533685	0.610185	2.398452
std.dev.	1.180592	1.136901	1.086183	0.82506
std.uncertainty	0.340807	0.328195	0.313554	0.238174

Fig. 5 calculation of precision repeatability uncertainty

The calculation of uncertainty due to precision reproducibility is shown in fig.6

Reproducibility from QA samples analysed in 2007				
	CHCl3	CHCl2Br	CHClBr2	CHBr3
QA Sample	27.85	24.87	25.25	24.54
QA Sample	26.48	23.95	24.8	24.02
QA Sample	27.5	24.5	24.55	23.77
QA Sample	27.54	24.47	24.49	22.69
QA Sample	28.01	23.93	24.37	23.9
QA Sample	29.96	25.72	26.81	26.33
QA Sample	28.21	25.12	26.44	26.05
QA Sample	30.14	25.72	27.96	27.34
QA Sample	20.07	18.06	19.53	19.82
QA Sample	20.26	19.07	19.15	20.13
QA Sample	20.76	19.53	20.14	20.13
QA Sample	21.94	20.75	20.61	21.19
QA Sample	28.51	25.17	25.88	25.4
QA Sample	25.9	22.56	23.06	21.88
QA Sample	26.42	23.26	23.56	22.13
QA Sample	21.55	19.18	19.91	19.36
QA Sample	22.06	19.41	20.02	19.26
QA Sample	21.61	19.2	19.21	18.25
QA Sample	29.14	26.58	26.47	25.36
QA Sample	27.25	25.32	25.22	23.97
QA Sample	25.51	23.67	23.45	22.85
QA Sample	27.34	24.58	24.26	23.77
QA Sample	31.9	29.56	28.65	27.3
QA Sample	31.74	29.28	28.62	27.41
QA Sample	26.54	24.89	24.82	24.36
QA Sample	23.49	22.13	21.79	21.4
QA Sample	26.72	25.06	24.68	24.07
QA Sample	28.05	26.4	26.47	26.02
QA Sample	27.22	25.41	24.96	24.06
QA Sample	30.74	28.47	28.19	27.73
QA Sample	27.21	24.94	24.62	23.89
QA Sample	28.12	26.14	26.01	25.86
QA Sample	29.85	27.13	26.32	25.39
QA Sample	26.67	24.88	24.79	24.24
QA Sample	26.27	25	24.66	23.51
QA Sample	28.82	27.3	26.78	25.91
std.dev.	3.190324	2.913648	2.716148	2.518447
mean	26.59306	24.20028	24.34722	23.7025
std.uncertainty	0.531721	0.485608	0.452691	0.419741

Fig. 6 calculation of precision reproducibility uncertainty

### Step 5

The expanded uncertainties are combined as per fig. 7 below using the root of the sum of squares.

Analyte	best estimate	Uncertainty Model	Uncertainty Bias	Uncertainty Precision Repeatability	Uncertainty Precision Reproducibility	combined Uncertainty	Method Uncertainty	% uncertainty
CHCl3	25.96	3.3808861	1.53019	0.340807	0.531721	3.764406	7.52881109	29.00158355
CHCl2Br	22.63	2.9181964	1.48212	0.328195	0.485608	3.325068	6.65013589	29.38637158
CHClBr2	21.74	2.8537865	1.50753	0.313554	0.452691	3.27414	6.54827901	30.12087859
CHBr3	20.94	2.6624146	1.57252	0.238174	0.419741	3.129565	6.25913098	29.89078786

Fig. 7 calculation of combined expanded uncertainties

The next step was to round and report the data as per the example below.

25.96 ug/L chloroform +/- 8 ug/L at a 95% LOC with a coverage factor of k=2

### 3. Discussion of results:

The data indicates an uncertainty for chloroform of 29% which is considered satisfactory for working with double digits in the ug/L range.

The data in fig. 7 above and fig. 8 below indicates that the largest source of uncertainty is the model followed by the bias.

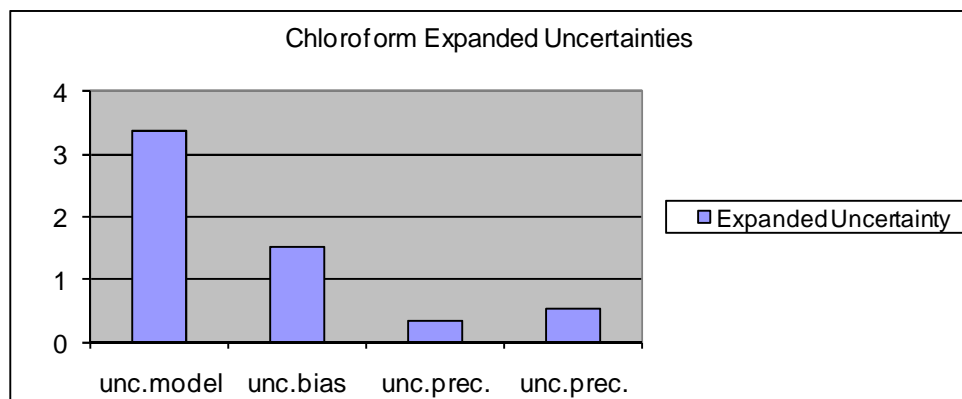


Fig. 8 graph of chloroform expanded uncertainties

Detailed examination of the model of chloroform indicates that the concentration of the internal standard is the largest contributor as per the graph in fig. 9.

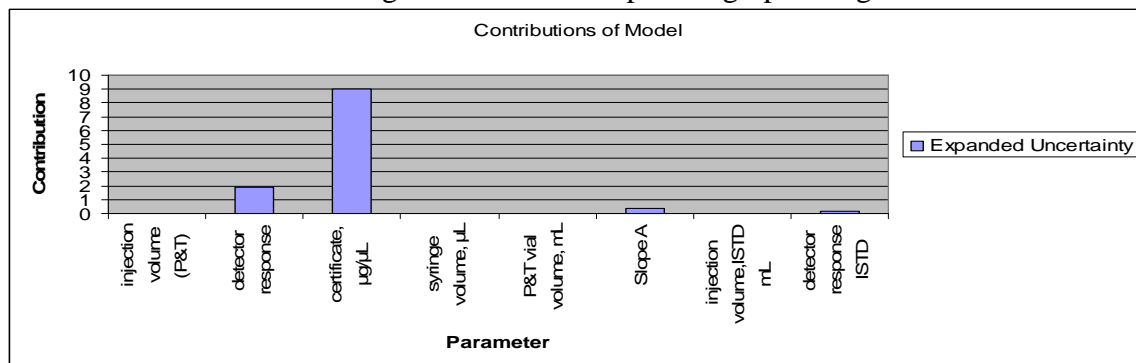


Fig. 9 Detailed graph of expanded uncertainties from the model

As the internal standard and the calibration standards have the same uncertainty contribution, an instant improvement to the method uncertainty can be made by using standard solutions with a tolerance of  $\pm 2$  instead of  $\pm 5$ . This will change the total uncertainty from 29% to 19.8%. Now as indicated in fig. 10 below the detector response of the GC-MS instrument has now become the largest contributor to the method uncertainty, and this is a non user changeable instrument parameter.

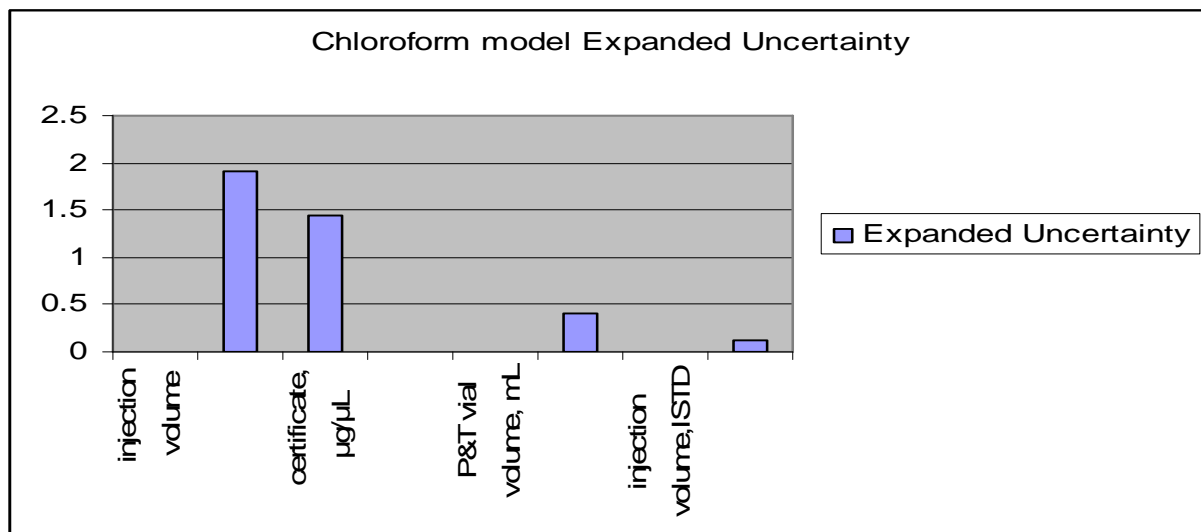


Fig. 10 Improved detailed graph of expanded uncertainties from the model

#### 4. Conclusion

This approach to uncertainty determination had an immediate benefit to the THM method in that we were able to improve method uncertainty by a simple change in the quality of standards purchased.

The laboratory has acquired an additional level of confidence in results generated.

This approach will be applied to the remainder of the methods used in the laboratory.

#### 5. References

Guide to the Uncertainty of Measurement (GUM) 1995

SANS 241

Chemstation reference manuals

NLA course notes for uncertainty of chemical measurement