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THE ROLE OF ION BALANCES IN EXAMINING THE RELIABILITY OF ANALYTICAL DATA: A CASE STUDY OF MENDIP STREAMS (SOMERSET)

by

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ABSTRACT

There is no formal scheme in the field of cave and Karst hydro-chemistry to allow workers to demonstrate the quality of their analytical work. Alternative procedures which allow them to do so (by calculating ion balances, or by comparing observed conductivity with the theoretical conductivity) both require the determination of ions which may be of no direct interest. Ion balance data from recent studies at sites on Mendip are presented. Ion balances provide a clear measure of analytical quality, and in many cases comparisons between imbalances and inter-ion regression data located analytical mistakes. Ion balance data are precise and easy to interpret, giving the method a significant advantage over conductivity-based procedures. Rigorous procedures in the use of primary standard solutions led to the detection and correction of an important source of systematic error.

INTRODUCTION

In cave and karst studies there is a long history of major research projects being carried out by researchers who are speleologists. Usually their training has been in geographical, biological or geological specialities but they have organised projects which have depended on the chemical analysis of water samples. Their contributions have been extremely important and will continue to be so. Such projects have been strongly influenced by the physical constraints imposed by caves: for example, the number and size of samples may be severely restricted by practical difficulties and there will be a tendency to bring samples to the surface rather than attempting on-site analyses. There is no need to elaborate on these considerations in the present publication. There is, however, a possibility that karst researchers may miss more subtle changes that develop in mainstream analytical chemistry. The present paper is designed to encourage workers to consider factors concerning the reliability of analytical data of speleological interest and the need to generate and publish data to support claims for analytical reliability and precision.

In cave and karst studies in the U.K., it is not usual for workers to provide evidence for the accuracy and reliability of their work. In the past even when a worker has, for his own satisfaction, undertaken such a step this part of his study has usually remained unpublished. As an example, when Stenner was establishing the reliability of his procedure for measuring the aggressiveness of water to calcium carbonate, he made determinations of total anion concentrations to provide evidence to demonstrate the absence of ions which were not determined directly (notably nitrate). He then proceeded to make ion balance calculations to provide evidence of the overall accuracy of his analytical work (Stenner, 1970 and 1971). Details of this aspect of his work were not published. Since that time, the importance of publishing evidence to quantify analytical reliability has become increasingly important in other fields of analytical chemistry. Stenner, for example, in his research into concentrations of a number of heavy metals in biological material, analysed standard orchard leaves purchased from the National Bureau of Standards in the U.S.A. to support evidence of analytical reliability (Martin *et al*, 1997).

Such measures have become standard practise in other areas of analytical chemistry, and in this respect a gulf has grown between procedures used in "mainstream" analytical chemistry and those used in cave and karst studies. To be specific, in attitudes towards the carrying out and reporting checks on analytical precision and reliability, there is a danger that standards in cave studies may become seriously weaker than elsewhere. The authors believe it is important to explain current practices in analytical chemistry, to encourage karst researchers to improve this aspect of the reporting of their work.

Establishing the reliability of analytical data goes far beyond establishing the standard error of individual procedures. There are two distinct stages in the process. The first stage is to generate data to be used to calculate the standard error of each individual analytical method used by the worker. The second stage is to devise a means of measuring the overall reliability of the battery of analyses used by the worker, and the way of doing this will depend on the nature and scope of the project. If the worker is lucky there may be standard samples available for him to use for this purpose. When data for the second stage have been establish, and the analytical chickens have come home to roost, the worker will have the extra job of relating the data to his standard error estimates. The authors here use their recent data to illustrate the general process.

Reference articles or manuals of laboratory procedures, such as those quoted in this paper, include standard errors obtained using those procedures. Workers should, in the course of their work, determine their own estimates of standard errors, and quote these figures in their reports, explaining how they were established. Specifically, they should say whether they were obtained by replicate analyses of standard solutions, or of real samples, or by the analysis of two separate duplicate samples. An example from within this paper is the evidence used to estimate the standard errors of the determination of sulphate and nitrate concentrations by ion chromatography (Table 1, below). However, this procedure cannot reveal the presence of a systematic source of error, which may have a serious effect. The balance used to weigh out the primary standard could be defective; there may be a fundamental flaw in the applicability of the method of analysis selected; there may be constraints or inter-species effects which have been ignored; a chemical may have been contaminated or a solution may have degraded. It may be appropriate to use more than one method to standardise the procedure. While falling short of providing data concerning overall reliability, this step will considerably enhance the reliability of the determination concerned. An example is quoted below where an atomic absorption spectrometer was calibrated using standard solutions prepared from a known weight of pure magnesium iodate crystals, and then used to analyse a separate standard solution prepared from a "spectrosol" magnesium solution sold by B.D.H. Ltd., using different volumetric glassware. However, establishing the basic reliability of each analytical method, and determining its standard error, must be a worker's first step in examining his own analytical reliability.

In the absence of "round robin" samples (or any other suitable evaluating procedures), the researcher's second stage will be to carry out a carefully designed all-inclusive test of reliability. In the context of the analysis of ionic solutions, there are two methods for doing so. One method is to analyse a sample for all ions present in significant concentrations, and, using a system of compatible units, to compare the total concentration of cations with that of anions. Another method (Wigley, 1972) is to measure the conductivity of a sample at a specified temperature (25°C, the temperature used in the reference data tables). As in the first method, the concentrations of all ions present in significant concentrations must be determined. The conductivity of each ion at the specified temperature is next looked up in standard tables, and the total theoretical conductivity is compared with the observed conductivity. It is essential to use a water bath with its temperature accurately and reliably controlled by a thermostat. This essential requirement is very difficult to achieve, and for this reason the authors chose ion balances to investigate analytical reliability.

In both of the methods, it is essential that analyses are made for all ions present in significant levels. Whereas in general, water samples will be analysed only for the ions of direct relevance to the study, the situation is different when it comes to establishing analytical reliability. There is a battery of analyses which must be carried out, some of which may be of no relevance whatever to the study in question. Because no system of supplying "round robin" samples suitable for karst studies has been set up in the U.K., this is the present less than ideal situation.

In the early stages of the preparation of this paper, it became clear that the fundamental need to establish analytical reliability has not been fully understood by respected figures in our field of study. In this paper, the authors have taken the opportunity to examine ion balance data from recent studies from the Mendip Hills. We show how we generated these data, and used them to evaluate the analytical reliability of the work. We are not producing a guide to analytical procedures, which have been described elsewhere. The choice of procedures is described, but not explained, this being outside the scope of this manuscript, except for instances where experience has shown this to be important. The choice of volumes used for analysis is one such instance.

Due to the physical constraints of sampling cave sites, obtaining the maximum information from the smallest possible sample is a rather obvious objective. For this reason, 25 ml aliquots were chosen for total hardness, alkaline hardness and chloride titrations, rather than a larger volume. Especially in the case of alkalinity, it is well known that using a "total container" analysis considerably increases the accuracy of the analysis (Rose, 1983). However, it was judged that adopting this procedure would be incompatible with the rest of the study, and a 25 ml aliquot was the volume chosen for the study. Although the collection of larger sample volumes (and hence the use of larger aliquots) was perfectly feasible at the surface sites, the authors chose to use the same sample collection regime at all sites, thus making all the sets of data directly comparable with one another. Included below are descriptions of sources and likely causes of errors. One of the errors described (in the standardisation of the acid used to determine alkalinity) is of particular relevance. Unusual values of a derived quantity, non-alkaline hardness, led to the discovery and description of an unusual phenomenon (a calcium sulphate surge in a stream following a severe drought, Heathwaite *et al*, 1999) and it was important to establish that the unusual data were, in fact, correct.

In the streams investigated by the authors, previous work has shown which ions must be analysed in order to assess analytical reliability. They are calcium, magnesium, sodium, potassium, bicarbonate/carbonate (as alkalinity), chloride, sulphate, nitrate and aggressiveness to calcite. Measurement of the last quantity will make it unnecessary to attempt to measure pH directly. It also makes it possible to calculate the concentrations of all species in the water/CO_2/calcite system, so important in many karst studies. If necessary, concentrations of additional ions from the equilibrium system can then be added to those already in the ion balance. In other karst areas, the list of ions that must be determined will not necessarily be the same.

In the earlier studies, many samples were also analysed for trace constituents (heavy metals, strontium, bromine, phosphate and fluoride) by X-Ray fluorescence spectrometry and colorimetric methods and all titrimetric determinations were carried out in duplicate. The earlier routines took so much time that the total number of samples analysed completely was relatively small (45 in Stenner, 1971). The results showed that the concentrations of these constituents were in every case too low to have any bearing on the ion balance investigations. In the present studies, Stenner carried out the titrimetric analyses (only total hardness and standardising titrations were in duplicate), atomic absorption analyses for magnesium, and flame emission analyses for sodium and potassium. Knights analysed samples for sulphate and nitrate using ion chromatography (at the same time confirming the absence of a number of anions). Helped by the advantages of the new choice of analytical techniques, the authors have, to date, completed the following analyses:

St. Cuthbert's Swallet feeder streams; 183.
St. Cuthbert's Swallet; 54.
Wookey Hole Cave; 22.
Other Mendip streams; 28.

Data from three sites on St. Cuthbert's Swallet feeder streams were examined in detail, providing a case study to show how the actual ion balance data were examined, revealing likely causes of significant individual imbalance. Whenever a significant imbalance was encountered, the question arose as to whether the imbalance was genuine (and an ion had been missed for some reason), or whether the imbalance had been caused by analytical mistake, or perhaps a transcribing mistake. Each imbalance was examined in relation to the full analysis of that sample, and to other factors in the sample (such as correlation with concentrations of other ions present at the particular site). The names of some sites are used below, but their only significance in this paper is to identify specific data. Their locations are therefore not described.

EXPERIMENTAL PROCEDURES

Samples were collected and stored in clean 250 ml or 130 ml plastic bottles. Duplicate samples for the determination of aggressiveness were saturated with AnalaR grade $CaCO_3$ on site, in 66 ml plastic bottles. On every occasion when the stream was seen to be carrying sediment, the sample was filtered through an 11 cm diameter Whatman 541 paper at the time of collection. This precaution has been shown to preserve samples (in clean sample bottles) from changes in salt concentrations for approximately 24 hours after collection. No preservatives were added. Samples were kept cool and in the dark until analysed. Because some solutes are more stable than others, the priority given to the various analyses varied from one quantity to another, as detailed below. Calculation of ion balances was the method chosen to examine the over-all analytical reliability.

Anion analyses

The choice of eluant made it impossible to use ion chromatography (IC) to determine bicarbonates, which were determined (as alkaline hardness) by titrating 25 ml aliquots with standard 0.02 M HCl (as described in Stenner, 1969). Except for the first five months of the study, B.D.H. 4.5 indicator was used to determine the end-point, and a wait of four minutes was used to ensure the end-point was stable. Silver nitrate titration (with potassium chromate indicator, using 25 ml aliquots) was chosen for chloride analyses. Determination of the "indicator blank" was essential in this procedure (Institute of Water Engineers, 1960). IC was used to determine sulphate and nitrate (full laboratory details are given in the appendix). IC would have shown the presence of many other anions, notably fluoride, phosphate, bromide or iodide, whose concentrations would then have been determined. In the event, not one of these ions was detected in any sample.

Cation analyses

Total hardness and aggressiveness to $CaCO_3$ were determined by titrating 25 ml aliquots with EDTA (standardised with magnesium iodate) and magnesium (determined by atomic absorption spectrophotometry (AAS) was subtracted to give the calcium concentrations (Stenner, 1971). This method, using Solochrome Black indicator in a buffered solution (Institute of Water Engineers, 1960), was preferred to the direct complexometric determination of calcium (with screened murexide indicator) because of its superior precision (due in part to the better colour change, to a vivid cornflower blue at the end-point). Careful use of "indicator blank" measurements, and fresh standardisation of the EDTA solution for each batch of samples (detailed below) were observed, and determinations were routinely carried out in duplicate. In many applications, AAS is appropriate to determine calcium concentrations (West and Herrin, 1965), and it might be thought that the procedure would be a good choice in karst studies because it is so much quicker than complexometric titration. But many ions, sulphate in particular, depress the calcium signal. Countering these effects (by, for example, swamping the sample with a lanthanum salt) destroys the advantages of AAS, and the use of AAS was therefore rejected as inappropriate for determining calcium in the present studies. Flame emission spectrophotometry (FES) was used to determine sodium and potassium concentrations. A Cambridge 917 instrument burning acetylene and compressed air was used for AAS and FES. The facility to use the instrument to measure concentrations with multiple aspiration (and to automatically calculate the standard deviation and relative standard error of each sample), was not used for the majority of samples, to conserve the sample.

The titration for total harness was the only determination carried out in duplicate (apart from samples analysed in duplicate to measure standard errors). Analyses for total hardness, alkaline hardness and aggressiveness were carried out as soon as possible after collection (usually within 6 hours, always within 24 hours), except for alkaline hardness in the first 5 months of the study. In those exceptions there were delays of up to a week before potentiometric titrations could be used. It was feared that the delay in the analyses for alkaline hardness, with the samples being held in a half-empty bottle, would lead to serious errors in the results for some of the samples involved. Some of the alkaline hardness results did in fact turn out to be at variance with those from earlier work, justifying these fears. The end-point of alkaline hardness titrations was subsequently found colorimetrically, using BDH 4.5 indicator, as soon as possible after collection, thereby ending delays in alkaline hardness determinations and consequent errors. Analyses for all other ions (which are known to be stable in suitably stored samples) were carried out as soon as was convenient.

The selection of simple titrimetric methods for determining total hardness, alkalinity and aggressiveness to calcite brought a very important feature to the present studies. Because these analyses are very suitable for use at home, there was no dependence on laboratory facilities and no impediment to completing the analyses within a very short time of sampling.

Units

In the IC work, concentrations were calculated as mg l^{-1} and then converted to Molar concentrations, the unit used throughout the remaining determinations. All concentrations were expressed as $10^5 \times$ Molarity. Total hardness, alkaline hardness, non-alkaline hardness, aggressiveness to $CaCO_3$ and calcium concentrations were therefore numerically equal to the concentrations expressed as ppm of $CaCO_3$, the unit most often used by limestone geomorphologists. The choice of units was appropriate for the investigation of ion balances. All calculations were carried out automatically by using a personal computer and appropriate spreadsheet programs.

ACCURACY AND PRECISION

The following standard errors were calculated from results of duplicate analysis of samples, as applicable for single analyses, except for total hardness (as $10^5 \times$Molar):
Total hardness, calcium and aggressiveness to $CaCO_3$, 0.8; alkaline hardness 3.0; Non-alkaline hardness, 3.8; magnesium, 0.17; sodium, 0.13; potassium, 0.05; chloride, 2.6; sulphate, 0.3; nitrate, 0.23.

Procedures for standardising solutions

The standardisation of the EDTA solution with magnesium iodate solution, and the standardisation of the HCl solution will be described in detail below, in the section discussing the accuracy of total hardness and alkalinity data. The same stock magnesium iodate solutions were used to make fresh working standard solutions for magnesium determinations. A serious error was discovered in the method first used to standardise the HCl solution. As a result, standardisation was changed to potentiometric titration with a standard sodium hydroxide solution. As the importance of this error was so great, the standardisation of the EDTA and HCl solutions are discussed in detail below. Prime stock solutions of KCl were prepared from a fresh batch of AnalaR grade KCl. They were used to prepare working standards for potassium determinations, for standardising silver nitrate solutions, and for preparing KCl solutions for measuring stream sizes by salt dilution. A stock solution of NaCl, made from dry AnalaR grade NaCl, was used to make up working solutions for sodium determinations. Stock solutions of sulphate and nitrate were prepared from AnalaR grade potassium nitrate and sodium sulphate (respectively) at 1000 mg l^{-1}, and freshly diluted to provide working standards for IC determinations.

Whenever a new stock solution was prepared, working standards were prepared from both the old and the new stock solution and the two sets of working standards were compared. Due to the rigorous use of this procedure, the serious error in borax solutions was discovered. If this systematic approach to standardising the working EDTA and HCl solutions had not been followed rigorously, the error might have gone undetected. This was the only occasion in the present study when a discrepancy between successive stock solutions was detected.

The 0.02 M hydrochloric acid solution was re-standardised every week, or for each batch of samples if the interval between batches was longer than one week. Changes in the concentration of the acid were insignificant for periods up to one month. Concentrations of silver nitrate solutions, used for chloride titrations, were also stable for the same time intervals. However, after only three days, changes in the concentration of the standard EDTA solution were significant and the precaution of re-standardising this solution for every batch of samples was a necessary measure. No problems were found with the standards used for other ions.

Accuracy of ion chromatography data

Three checks on the reliability of the determination of sulphate and nitrate by IC were carried out. They were:

A. Repeat analysis of a standard solution,

B. Repeat analysis of one sample (from Fair Lady Well, 25 October 1996)

C. Data from repeated analysis of samples (sometimes after a delay of more than a month, because a result appeared to be unusual, or because a result was close to the limit of the calibration range). The results are shown in Table 1.

Ion	No	Mean $10^5 \times$ M	Std Error $10^5 \times$ M	RSE %
A: Sulphate	8	10.4	0.11	1.15
A: Nitrate	8	12.8	0.23	1.80
B: Sulphate	10	8.9	0.05	0.55
B: Nitrate	10	12.3	0.13	0.99
C: Sulphate	21	8.4 - 50.1	0.30	1.91
C: Nitrate	16	0.9 - 40.0	0.23	2.55

Table 1. *The reliability of sulphate and nitrate determinations by ion chromatography, given by repeated analysis of A, a standard solution; B, a sample from Fair Lady Well; C, duplicated analysis of samples.*

The figures presented in Table 1 show that the use of IC greatly improved the precision of the sulphate determination, compared with the colorimetric method used previously by Stenner (1971), and at the same time gave data for nitrate concentrations of similar precision. The assessment of the total ion balance was therefore much more precise than in Stenner's previous attempts.

The accuracy of total hardness and alkalinity data and standardisation of EDTA and HCl solutions

Non-alkaline hardness measurements were unexpectedly important. Unusually high values of non-alkaline hardness in the Pool Outflow stream gave the first indication that an unusual event was in progress after the 1995 drought (Heathwaite. Knights and Stenner, 1999). Non-alkaline hardness is the difference between total hardness and alkaline hardness, so its accuracy depends on the accuracy of the standardisation of the EDTA and HCl solutions used in the primary determinations. Owing to the discovery of important problems, the procedures used to standardise the EDTA and HCl solutions must be described in detail.

Every time a new batch of samples was analysed, the EDTA solution was standardised by titrating it against aliquots of a standard 0.25 mM magnesium iodate solution, made from crystals of the salt of analytical grade, supplied by Hopkin and Williams Ltd. This reagent has been recommended for use in standard solutions because of its stability and its high molecular weight (Stenner, 1971). Whenever a new batch of magnesium iodate standard solution was prepared, its concentration was compared with the previous batch, by titrating both batches with the same EDTA solution. No discrepancy was ever detected between successive batches. One batch of magnesium iodate was used to provide a set of standards to calibrate the spectrometer for the determination of magnesium by AAS. A second 20.0×10^{-5} M Mg solution was prepared from a new bottle of BDH "Spectrosol" Mg standard solution. This solution was analysed by AAS, with the instrument calibrated with the magnesium iodate standards. The result was 20.0×10^{-5} M. This confirmed the reliability of the procedure in which magnesium iodate solutions were used to standardise the EDTA solution.

Standardising the acid for alkaline hardness gave the unexpected problem. Initially, the acid was standardised by the potentiometric titration of 0.01 M sodium tetraborate (borax), freshly diluted from a 0.05 M stock borax solution. When, after 18 months, a new stock borax solution was prepared, and the acid was titrated against both borax solutions, there was an unacceptable discrepancy. To resolve the difference, the acid was restandardised by potentiometric titration, using freshly made up 0.02 M NaOH, prepared from a "Volucon" capsule. It was discovered that both stock borax solutions were inaccurate. The bottle of AnalaR grade borax used to prepare both stock solutions had been contaminated. Each time a batch of samples had been analysed, the HCl solution had been standardised with a borax solution whose concentration had been incorrect. Using the measurements that had uncovered the discrepancy between the two stock borax solutions, together with the measurements obtained using 0.02 M NaOH, the true concentrations of the two stock borax solutions were calculated. Using the standardisation figures and the correct borax concentrations, the true concentration of the acid on each occasion was recalculated. Calculating the correct alkaline hardness and non-alkaline hardness figures obtained in the previous eighteen months was then possible.

After the alkaline hardness and non-alkaline hardness figures had been recalculated, some of the data from 1994 were still different from earlier and later data. Specifically, data for samples whose analysis had been delayed, and which had been suspected of being unreliable, were still anomalous. When the analysis of stored samples for nitrate and sulphate analyses was complete and anion/cation balances had been calculated, the unreliable data obtained in the early months of the study were reviewed. Samples in which the alkaline hardness differed greatly from the expected results did indeed have an unsatisfactory anion/cation balance. Furthermore, in samples from two sites at which sulphate and non-alkaline hardness are usually equal, sulphate values differed from non-alkaline hardness by a quantity equal to the ion imbalance. Finally, the imbalance was also seen in the difference between the non-alkaline hardness and the normal non-alkaline hardness. This was proof that delays in the analyses had indeed led to errors in the alkaline hardness results, and the ion imbalance measured the value of the error of the alkaline hardness titration. For these samples, and only for these samples, the value of the anion imbalance was used to calculate the correct value of the alkaline hardness. When this was done, in every case the corrected alkaline hardness and the recalculated non-alkaline hardness data became normal, and the discrepancy between the non-alkaline hardness and sulphate disappeared. In several samples from this period, IC data were not available. In these cases, alkaline hardness and non-alkaline hardness figures were unreliable, and because there was no rigorous procedure for correcting them, they were deleted from the database.

The reliability of atomic absorption and flame emission spectrometry data

It will be noticed that the examination of ion balance data indicated that in some samples, errors in sodium data far greater than the estimated standard error for sodium probably occurred. Because of the need to conserve samples, replicate analyses of samples were not routinely carried out. This prevented the routine assessment of the precision of every single analysis for magnesium, potassium and sodium being made. There was another consequence of this sample conserving measure. There are times when there is a temporary partial blockage of the instrument's aspirator system, caused by a tiny particle being drawn into the capillary tubing. When repeated analyses are carried out, the partial blockage leads to a high Relative Standard Error being audibly signalled for the sample, indicating the need to clear the aspirator system. Deciding to make only single analyses therefore also made it impossible to spot these temporary partial blockages. This would lead to an error considerably greater than the estimated standard error, and this is the likely cause of the errors in sodium concentrations which were located by examining ion balance data (Table 3, below).

Stability of samples

The stability of the chemical characteristics in water samples was investigated when methods for measuring aggressiveness were being compared (Stenner, 1969). It was shown that if water can be seen to be carrying sediment, pH, total, alkaline and calcium hardness of an untreated sample will, six hours after collection, be significantly different when compared with data from a sample which was filtered at the time of collection. If sediments on the streambed are stirred up when samples are being collected, similar changes will be likely unless the sample is immediately filtered. (Filtering the sample will cause pH and dissolved gas concentrations to change). This is the reason why, in the recent studies by the authors, water samples were filtered into the collecting bottles whenever sediments were visible in the streams being sampled.

Recent studies from St. Cuthbert's swallet have shown that when sediments are disturbed when samples are being collected, ions from the $CaCO_3/H_2O/CO_2$ system are not the only ions that will change. In the majority of such samples collected at stream junctions, K, Mg and SO_4 concentrations showed changes that were large enough to give impossible results for stream ratios calculated using these ions (Stenner and Stenner, 1999, in preparation). It is probable that complex aluminium silicates in the stirred-up mud, acting as natural ion-exchange media, caused these changes.

Repeated analysis for Mg, K, Na, Cl, SO_4 and NO_3 proved that careful storage of sediment-free samples (in cool dark conditions) did not result in detectable changes.

ANION/CATION BALANCES.

By adopting the present choice of units, it was straightforward to calculate the overall ion balances, providing evidence for analytical reliability. The raw data contained several instances when the balance was unexpectedly poor. However, the present data stand as examples of what can be achieved by analysing relatively small aliquots of small samples. Summaries of ion balance data are presented in Table 2.

In the sets of data from the Mineries Pool Outflow stream, Ladywell, and St. Cuthbert's Swallet, the standard deviation of the ion imbalance was close to 6.6×10^{-5} M, the total (univalent) standard error of all the determinations involved (Total S.E. = $(SE_1^2 + SE_2^2 + SE_3^2 + \ldots)^{1/2}$). In three of these four data-sets, the mean imbalance was close to zero. The general conclusion in these three data-sets, therefore, was that the over-all ion balance was that which was that which was predicted from the sum of the individual standard errors involved. It was also evident that the data-set from St. Cuthbert's Swallet was just as reliable as those from the surface sites

Study	No.	Mean Imbal.	%Mean Imbal.	Standard Deviation	R.S.E. %
Mineries Pool Outflow	52	-0.88×10^{-5} M	-0.18	6.95×10^{-5} M	1.58
St. Cuthbert's Stream Sink	89	-0.22×10^{-5} M	-0.05	9.08×10^{-5} M	1.56
Fair Lady Well	42	-3.44×10^{-5} M	-0.48	7.24×10^{-5} M	1.03
St. Cuthbert's Swallet (Cave)	53	-0.98×10^{-5} M	-0.34	5.12×10^{-5} M	1.55
Wookey Hole Cave	8	8.57×10^{-5} M	0.73	15.4×10^{-5} M	1.31
Other Stream Sites	10	2.32×10^{-5} M	0.17	13.4×10^{-5} M	1.01

Table 2. *Ion balance data from various studies of streams on or flowing from the Mendip Hills, Somerset. Molar concentrations, univalent.*

The data-set from Wookey Hole cave, which included samples brought to the surface from beyond Wookey 22, need more comment. Every sample collected in high-water conditions in November and December 1996 contained significant concentrations of suspended (or "colloidal") calcium carbonate. This made it difficult, or impossible to analyse the samples for total hardness (hence calcium) or alkalinity. The "best attempts" produced data known to be of dubious precision. In fact, ion balances confirmed the fact that these analyses were seriously inaccurate, even if the results obtained by analysing the samples for magnesium were still valid, and extremely interesting (Chapman *et al*, *in prep.*). Data from these collections were not included in Table 2.

A batch of river samples were collected and analysed in July 1996, and all samples showed evidence of the presence of "colloidal" calcium carbonate, and had very poor ion balances. These results were excluded from Table 2. When later samples from Wookey Hole Cave (from January 1997 and July 1997) were analysed, slight interference from "colloidal" calcium carbonate was detected and measured in three of the samples. However, the ion imbalance in these samples (which were excluded from the Table 2 data-set) were 41, 33 and 30 × 10^{-5} M, which proved that the "colloidal" calcium carbonate had an effect on the total chemistry of the water which was much greater than had been expected. Four of the river samples (from Cheddar Rising, Rodney Stoke Rising, the River Brue and the River Sheppey) were excluded from the final data-set in Table 2 because they contained detectable "colloidal" calcium carbonate and also yielded poor ion balances (46, 68, 22 and 28 respectively). Other examples from Wookey Hole Cave and other river sites also had rather poor ion balances, all probably due to the same cause (a slight colloidal calcium carbonate content), and these data had an adverse effect on the relevant statistical summaries in Table 2.

From the very first batch of ion-balance data, it was obvious that the procedure was much more than a means of quantifying analytical reliability. When used in conjunction with other data from a site (in which the extent and nature of variability at the site were known and where regression equations between species were known), ion balances could be used to show when an unexpected ion was present, or when an error had occurred. An example occurred at Fair Lady Well when there was no flow, on 24 August 1995 see below. Furthermore, it was often possible to show which individual analysis contained an error.

At the Pool Outflow, there were 5 samples for which the ion balance exceeded $\pm 10 \times 10^{-5}$ M (univalent); at the cave sink there were 14 such samples. At the Pool Outflow there is a high 1:1 correlation between non-alkaline hardness and sulphate concentrations, and 3 of these 5 suspect samples showed an abnormal difference between these two quantities. This was a very strong indication that there had been an equivalent error in the alkaline hardness determination in these three cases. At the Main Stream Sink where, as at the Pool Outflow, non-alkaline hardness was usually very close to the sulphate level, there were 4 such examples. There were also 3 more examples where an error in the alkaline hardness seemed to have been partly responsible for the ion imbalance (the ion balance being greater than a significant sulphate/non-alkaline hardness discrepancy).

For example, in the Mineries Pool Outflow stream on 14 May 1996, the ion imbalance (univalent) was 20×10^{-5} M. This value is large enough to suggest that there was an error in the data for this sample. The sulphate was 12.1×10^{-5} M, and the non-alkaline hardness was 3.0×10^{-5} M. The last two quantities had a linear 1:1 relationship at this site. Also, the non-alkaline hardness at the site on this occasion was much lower than usual. The cause of the ion balance was very likely to have been an error in the alkaline hardness titration, possibly the burette reading had been misread. The alkaline hardness was approximately 9×10^{-5} M too high, and the correct non-alkaline hardness should therefore have been 12×10^{-5} M, numerically the same as the sulphate, and which is close to the values at the site in previous and succeeding samples. The effect of the decrease in alkalinity of 9×10^{-5} M, which is 18×10^{-5} M mono-valent, was to reduce the imbalance to 1×10^{-5} M

Discrepancies in the sulphate/non-alkaline hardness association had therefore proved to be useful in pinpointing the reason for poor ion balance data at these two stream sites, because of the close relationship between sulphate and non-alkaline hardness at the two sites. Data from other samples at the two sites where there was a notable ion imbalance were re-examined. It was noticed that in some cases, the sodium ion concentrations differed significantly from those of chloride. At both sites, these two concentrations were usually numerically very close to one another, because of their common origin in wind-blown sea spray. For example, on 27 September 1995, the ion imbalance was 12×10^{-5} M. The chloride was 28.1×10^{-5} M, and the sodium was 40.9×10^{-5} M. The sodium was unusually high, and the chloride concentration was close to that in previous and succeeding samples. This suggested that the chloride/sodium imbalance indicates an error which resulted in the ion imbalance, and the error was a sodium concentration which was 13×10^{-5} M too high. There were other similar examples, where an imbalance between sodium and chloride was associated with an ion imbalance of the same (or similar) magnitude. This discovery thereby provided a second method for indicating the cause of a poor ion balance.

At the Main Stream Sink, there were 3 samples (23 July 1996, 15 January 1996 and 30 April 1996) in which the imbalance had been caused by an error in the alkaline hardness together with an error in sodium or chloride determinations. In one sample from the Pool Outflow (19 June 1996) and one from the Main Stream Sink (4 February 1996.) there was no obvious explanation for the poor ion balance.

The method of analysing ion balances, by referring to non-alkaline hardness/sulphate concentrations and to chloride/sodium balances, is not universally applicable. It could not be applied to the 12 samples from Fair Lady Well for which the imbalance exceeded $\pm 10 \times 10^{-5}$ M. This is because at this site, unlike the other two sites, there was no simple linear relationship between non-alkaline hardness and sulphate concentrations.

The results of the diagnostic examination of ion balances at the three surface sites are summarised in Table 3

Date	Imbalance (univalent)	Probable reason for imbalance (univalent)
Pool Outflow		
18/11/95	17×10^{-5}	chloride 14×10^{-5} M too high
20/11/95	11×10^{-5}	alkaline hardness 12×10^{-5} M too low
14/05/96	-20×10^{-5}	alkaline hardness 18×10^{-5} M too high
19/06/96	-17×10^{-5}	Unexplained
22/07/96	12×10^{-5}	alkaline hardness 9×10^{-5} M too low
Main Stream Sink		
23/07/95	16×10^{-5}	alkaline hardness 8×10^{-5} M too low, chloride 10×10^{-5} M too low
13/08/95	12×10^{-5}	alkaline hardness 9×10^{-5} M too low
24/08/95	11×10^{-5}	alkaline hardness 11×10^{-5} M too low
27/09/95	12×10^{-5}	sodium 12×10^{-5} M too high
15/01/96	12×10^{-5}	alkaline hardness 6×10^{-5} M too low, sodium 8×10^{-5} M too high
04/02/96	-11×10^{-5}	Unexplained
13/02/96	-10×10^{-5}	sodium 6×10^{-5} M too low
27/02/96	-12×10^{-5}	sodium 8×10^{-5} M too low
10/03/96	-13×10^{-5}	sodium 10×10^{-5} M too low
24/03/96	-12×10^{-5}	chloride 7×10^{-5} M too high
30.04.96	-11×10^{-5}	alkaline hardness 6×10^{-5} M too high, sodium 4×10^{-5} M too low
04/05/96	-10×10^{-5}	sodium 6×10^{-5} M too low
17/05/96	10×10^{-5}	alkaline hardness 10×10^{-5} M too low
29/07/96	-13×10^{-5}	alkaline hardness 9×10^{-5} M too high

Table 3. *Ion balance data from streams which enter St. Cuthbert's Swallet, listing the cases where a significant imbalance was found, and the probable mistakes responsible for the imbalance.*

There is one other source of possible errors, indicated very clearly by the results from Fair Lady Well on 24 August 1995. In this sample, collected when there was no flow from the spring, contamination of the well is known to have taken place. The imbalance was approximately 93×10^{-5} M (univalent). Concentrations of chloride, sulphate, nitrate and sodium had also become wildly different from normal. Chloride and nitrate concentrations both exceeded 70×10^{-5} M (univalent). The figures are consistent with contamination by urine and the authors suspect that an ammonium ion concentration of approximately 93×10^{-5} M was the likely cause of the imbalance, although this was not confirmed. Because this ion is not found in unpolluted natural waters, the determination of ammonium concentrations had not been considered in this study. Occasional instances of contamination of the Fair Lady Well, or of the Pool Outflow stream by this ion, cannot be ruled out as possible causes of ion imbalance.

CONCLUSIONS.

The reliability of the analytical data of ion concentrations in water samples has been examined. A large quantity of data was obtained following studies of streams which enter and pass through St. Cuthbert's Swallet, Mendip. The following conclusions have been made:

1. A rigorous approach to the preparation and comparison of prime standards is essential to minimise systematic errors.

2. If an alternative method of analysis is available for any determination, the worker should consider using it to support his data, especially if a fundamentally different principle is involved, and its use may confirm the absence of a systematic source of error.

3. Regression equations linking different quantities should be examined, because a data-point that does not fit a regression graph may be the result of an analytical mistake. However, if an apparent misfit is confirmed as being accurate, this may be a significant discovery.

4. Replicate analyses of samples, while providing data used to calculate the standard error of the measurements, do not guarantee the absence of a systematic source of error.

5. In the absence of the "blind" analysis of suitable solutions supplied by an accrediting organisation, calculating ion balances can provide evidence of a worker's quality of work. The imbalance should be explained by reference to the standard errors of the individual analytical methods.

6. To calculate ion balances, or to compare the actual conductivity with the theoretical conductivity, it will be necessary to determine concentrations of all ions present in significant concentrations, including those which may be of no direct interest to the worker.

7. If a stream can be seen to be carrying sediment, it is most important to filter the water into the sample bottle. Subsequent filtration is not a valid option.

8. Alkaline and non-alkaline hardness data from occasions when a delay in alkaline hardness titrations was unavoidable proved that considerable changes in the $CaCO_3/H_2O/CO_2$ system had taken place in half-empty sample bottles. This proves the need to analyse all samples for total and alkaline hardness, and aggressiveness to calcium carbonate as soon as possible.

9. Ion balances provide clear, precise measures of analytical reliability, and in many cases related data from the site can be used in conjunction with ion balance data to pin point individual sources of error. Because ion balance data are precise and easy to interpret, the method has a significant advantage over procedures which depend on conductivity measurements.

10. If a sample is carrying "colloidal" calcium carbonate (indicated by an unstable end-point in the total hardness titration), serious errors are likely, resulting in a grossly unsatisfactory ion balance.

11. The precision of an ion balance study may be improved by titrating larger aliquots in the volumetric analyses, and by replicate analyses in all the determinations.

12. It will be very difficult for the conductivity-based method to rival the precision of the ion balance method for assessing analytical reliability. Achieving a sufficiently stable control over the temperature of a water bath is a major problem in this alternative procedure.

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APPENDIX: TECHNICAL DETAILS OF THE DETERMINATION OF SELECTED ANIONS BY ION CHROMATOGRAPHY

Ion Chromatography is an extremely flexible analytical tool, widely used in water analysis (Welch *et al*, 1996, Tanner and Chan, 1996). By choosing a suitable ion exchange column and eluant, the same instrument can be set up to determine a wide range of anions or cations. The technique can analyse a small sample with good precision and accuracy. However, IC instruments are expensive, and the skills required to set up and maintain the facility are specific and demanding.

Nitrate and sulphate species were determined using a Dionex Model 2020i ion chromatograph fitted with an AG4 guard column and an AS4 separator column. The eluant system was 1.7 mM sodium carbonate/ 1.8 mM sodium hydrogen carbonate made up in double distilled water, at a flow rate of 1.5 to 1.7 ml min^{-1}. Suppression of cations in the column effluent prior to the conductivity cell was by a counter-flowing membrane suppressor using 0.0125M sulphuric acid as the regenerant. Individual standards were prepared from AnalaR grade potassium nitrate and sodium sulphate at the 1000 mg l^{-1} level and diluted to give mixed standards in the range 0 – 20 mg l^{-1} sulphate and 0 – 16 mg l^{-1} nitrate. Samples and standards were injected by an air operated slider valve fitted with a 50 µl sample loop. Signal output from the conductivity detector was fed to a Dionex 4270 integrator for determination of peak areas. Samples collected during the non-alkaline hardness/calcium sulphate event and some samples from Fair Lady Well when flow from the spring had ceased needed to be diluted prior to analysis for sulphate and nitrate respectively. Actual concentrations of sulphate and nitrate in the samples were then calculated using an EXCEL spreadsheet using either the in-built linear regression analysis or a modified standard addition programme to determine slope, intercept and correlation coefficients. Concentrations were calculated as mg l^{-1} and then converted to Molar concentrations.

As the natural range of chloride concentrations in the streams was relatively high, silver nitrate titration was preferred for chloride analyses. The choice of eluant made it impossible to use IC to determine bicarbonates. IC would have detected the presence of many other ions, notably fluoride, phosphate, bromide or iodide, and determined their concentrations.

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