

## **KARL FISCHER TITRATION, A SELECTIVE METHOD FOR DETERMINING WATER IN FOOD**

**H.-D. Isengard**

University of Hohenheim, Institute of Food Technology,  
Garbenstr. 25, D-70593 Stuttgart, Germany

### **Abstract**

*Water determination is therefore the most frequent analysis performed on foodstuffs. Various methods are applied. Very often the sample is dried and the mass loss is regarded as water. The Karl Fischer titration is based on a selective chemical reaction and yields, therefore, really the water content of the sample as result. Because of its selectivity for water the Karl Fischer titration is the ideal reference method for water determination.*

**Keywords:** *water, food, Karl-Fischer titration, selectivity*

### **Introduction**

Water is present in practically all foodstuffs and water content is important for many reasons, for instance in terms of

- technological processes,
- physical properties,
- rheological behaviour,
- specifications based on dry matter,
- microbiological stability,
- shelf life,
- logistic aspects,
- economic issues,
- legal regulations.

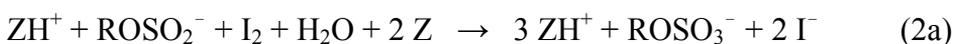
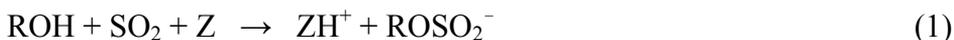
Water determination is therefore the most frequent analysis performed on foodstuffs. Various methods are applied. Very often the sample is dried and the mass loss is regarded as water. This is in many cases not correct. The expression “moisture” is therefore more and more applied for this mass loss. It comprises all the matter volatile under the conditions applied. The Karl Fischer titration is based on a

selective chemical reaction and yields, therefore, really the water content of the sample as result.

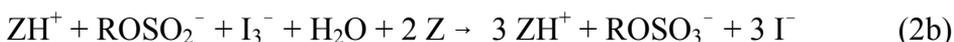
## 1. Chemical background

### 1.1. Karl Fischer reaction

Water is determined by a selective chemical reaction. In protic solvents like alcohols the reaction follows a two-step scheme (Scholz 1984, Grünke and Wünsch 2000):



or



ROH is usually methanol, but recently reagents on the basis of ethanol have been developed (Schöffski 1998). Other alcohols can be utilised as well under certain conditions. Z is a base, in modern reagents very often imidazole and no longer the 'historical' pyridine that was used by Karl Fischer when he introduced the method (Fischer 1935).

From the reaction scheme it is obvious that one mole iodine reacts with one mole water. From the consumption of iodine the amount of water can therefore be concluded.

### 1.2. Chemical interferences

Two principal possibilities exist:

The sample may contain a substance that interferes with the Karl Fischer reaction. Iodine may be reduced in another way and thus pretend a higher water content. Or, on the contrary, iodide may be oxidised. When these reactions are much more rapid than the Karl Fischer reaction, substantial interferences are possible. If the content of these substances and the stoichiometry of the interfering reaction are known, the erroneous result can be corrected.

It may also happen that compounds are released by the dissolving of the sample in the working medium which can react with each other by forming or by consuming water. These substances may be contained in the original sample like carbonates and solid acids. But

this may also concern a compound of the sample and a component of the Karl Fischer reagent. Such a case would be the reaction of a carbonyl compound and methanol to form a hemiacetal or hemiketal and water.

It is often possible to deal with these interfering reactions in an appropriate way. In particular, special reagents exist for the titration of samples that contain carbonyl compounds. They contain other alcohols than methanol which have a much less expressed tendency to react with carbonyl compounds.

### *1.3. Physical interferences*

As water must react chemically with the reagents, it must come in physical contact with them. Samples that are insoluble or only partially soluble in the working medium must be treated in a way that the water migrates into the working medium. In extreme cases a determination is not possible.

It may, however, be the aim of the analysis to determine only the surface water. In these cases, dissolution of the sample should be avoided.

## **2. End-point determination**

### *2.1. General procedure*

Different techniques exist. In any of these cases the titration cell which contains the working medium is first titrated to dryness, a procedure called conditioning. Then the sample is introduced, the water is titrated and the amount of water is calculated on the basis of the amount of iodine consumed. After the analysis the cell is kept dry (conditioned) and a further sample may be introduced and analysed.

These steps are carried out automatically by computer-controlled titrators.

### *2.2. Detection of the end point*

At the end of the determination iodine is no longer consumed. The first excess of iodine indicates the end point. It reacts then with iodide formed during the titration to yield brown triiodide. Although a visual or photometric end-point determination is therefore principally possible, electrochemical techniques are usually applied.

A pair of electrodes is submerged in the working medium in the titration cell. They are polarised by a constant current and the voltage necessary to maintain this current is monitored. This is the voltametric technique. At the end point the voltage drops abruptly, because at both electrodes a simple reaction is now possible, the anodic oxidation of iodide to give iodine and triiodide and the cathodic reduction of iodine and triiodide to give iodide. An analogous procedure is the amperimetric technique. The electrodes are polarised by a constant voltage and the current is monitored. Here, a sudden rise of the current is observed at the end point. In both cases, a certain value is chosen as stop criterion. When the voltage falls and remains (see 2.3) below it or the current rises and remains (see 2.3) above it, the analysis is stopped. These values are called stop voltage and stop current respectively.

### *2.3. Stop delay time*

An “ideal” sample is soluble in methanol. The water contained in the sample is then completely free and can immediately react with the reagent. The end point is reached very quickly and “definitely”. Many samples, however, do not fulfil this condition. The water contained in the more or less insoluble sample reaches the working medium only by and by. If in such situations the analysis would really be definitely stopped immediately after the first reaching of the stop voltage or the stop current respectively, delayed water would not be detected. To avoid this, a stop delay time can be programmed. Only when the voltage or the current remains below (voltage) or above (current) the chosen end-point value during this time is the titration really ended and the water content calculated from the amount of iodine consumed.

### *2.4. Drift as end-point criterion*

During conditioning (keeping dry) the titration cell, small amounts of iodine must be added to account for the water that intrudes in the cell from the environment. This consumption of reagent in the course of time is called drift. It can be measured by carrying out a titration without addition of a sample. The reagent consumption to keep the cell dry for a certain time is measured.

This drift can be used as end-point criterion. Assuming that the conditions immediately before and after a determination are the same, the drift before and after the analysis is the same. During titration the reagent consumption is higher because of the water from the sample.

When, therefore, the reagent consumption falls back to the value of the drift, all the water of the sample has been detected. Usually, however, a stop drift slightly higher than the drift measured before the start of the titration is set. The reason is that conditions may change and, if the drift becomes higher during titration, the analysis would never be stopped.

### *2.5. Titration curves*

A good means to check the correctness of determinations are so-called titration curves. Various entities can be recorded versus time. In the case of voltametric end-point determination the voltage or the difference between voltage and stop voltage can be recorded ( $U/t$  or  $\Delta U/t$  curves). The most common curves are those of titration volume versus time ( $V/t$  curves). The  $V/t$  curve rises quickly at first (free water is determined) and then bends to become more and more flat (more or less bound water is determined). At the end the curve should run practically parallel to the time axis (when the drift is near 0) or have a constant very low slope (when a drift exists).

### *2.6. Water equivalent or titre*

The water equivalent or titre of a reagent is the water mass that is indicated by 1 ml of reagent. It exists only for the volumetric techniques (see 3.1). It is determined by titrating a known amount of water. For this purpose either pure water or a substance with known water content is used. Water itself is the cheapest substance, but errors in dosing this "sample" have a 100% consequence. A frequently used compound is sodium tartrate 2-hydrate. It contains 15.66% water by mass and is very stable. Liquid standards with defined and certified water content are also on the market. The water equivalent of reagents must be checked from time to time.

## **3. Titration methods**

Two principally different techniques exist, the volumetric method (3.1) and the coulometric method (3.2). In both cases, a so-called working medium is placed in the titration cell. This working medium is titrated to dryness before the start of the analysis. The sample is then introduced into the cell. Solid samples are introduced by opening a stopper in the lid of the cell. Such an opening is only possible in the

volumetric technique. Liquid samples can be introduced by a syringe through a septum in the stopper. Gaseous samples are led into the cell through a needle. A drying tube filled with molecular sieve is used for pressure equilibration.

### *3.1. Volumetric method:*

In the volumetric method, a solution of iodine is dosed into the titration cell and the volume necessary is recorded. From this volume, the water equivalent of the reagent and the sample size the water content is calculated. Other factors can also be accounted for, for instance the water content of a solvent that was used to dissolve the sample or the consumption for the drift taking the titration time into account.

Two different volumetric techniques can be distinguished.

In the one-component technique, all the necessary reaction components, iodine, sulphur dioxide and the base are dissolved in an appropriate solvent such as diethyleneglycol monoethyl ether. This is the titrating solution. The working medium is usually methanol, possibly with addition of a further solvent (see 4.6).

In the two-component technique, the reactants are distributed in two solutions: The titration component is a solution of iodine in methanol, the working medium is a solution of sulphur dioxide and the base in methanol, possibly with an additional solvent (see 4.6). The two-component technique needs two Karl Fischer components, but it guarantees an excess of sulphur dioxide which accelerates the reaction.

A further advantage is that the water equivalent of the titration solution is more stable than that of the one-component solution. The advantage of the one-component technique is that all the necessary reactants are added during titration whereas in the two-component technique the capacity of sulphur dioxide and base is limited and the solvent component becomes exhausted after several titrations.

For special applications like samples containing carbonyl compounds, special reagents exist.

### *3.2. Coulometric method*

In the coulometric method, iodine is not added but produced in the titration cell by anodic oxidation of iodide. The electrical charge necessary for the production of the amount of iodine consumed for the water in the sample is measured. The method is therefore “absolute”

and water equivalent is not relevant. This technique is very sensitive and the titration cell must not be opened after it has been titrated to dryness. Samples must be introduced in liquid or gaseous form through a septum. In special applications the water of solid samples is set free in an oven and the vapour driven into the cell by a stream of dried air.

The coulometric method is practically only used for the determination of very low water contents and is regarded as a trace method.

### *3.3. Automation*

Several techniques of sample changers have been developed.

This concerns “classical” volumetric titration. Sample beakers (closed by aluminium sheets) are brought to the titrating position where the sheets are penetrated by the electrodes and the burette. Blind values for the beakers and the solvent are subtracted in the calculation of the results.

Other techniques use the extraction of water by a solvent or a mixture of solvents before starting the analysis. Aliquots are taken from the assays and titrated in the titrating position.

In the “gas extraction”, the samples in vials are brought to the titrating position where a stream of dried air or nitrogen is led through the sample in the vial through a double needle. The gas with the water of the sample is led into the titration cell (volumetric or coulometric) where water is determined.

## **4. Availability of the sample water**

As the Karl Fischer method is based on a chemical reaction, the sample water must get in direct contact with the reactants. Ideally, the sample should therefore be soluble in the working medium. This condition is usually fulfilled in the coulometric method, because the sample is introduced in gaseous or liquid form. But many foodstuffs cannot be dissolved completely. In these situations the sample water is not immediately available but reaches the working medium only after some time by diffusion and extraction processes. The flow of water from the interior of the sample particles into the working medium becomes gradually slower because of the decreasing concentration gradient and is very slow at the end. There is therefore the risk of

stopping the titration before all the water is detected. Particularly in these cases, the recording of titration curves is helpful. The shape at the end usually shows a still decreasing slope, which indicates that there is still water coming into the working medium.

Several measures exist to accelerate the liberation of the sample water and to make a complete detection probable (Isengard 1995).

#### *4.1. Long titration time*

In order to provide a time long enough for practically all the water to reach the working medium, a minimal titration time can be enforced. This means that the analysis must not be ended before, even if the stop criteria should be fulfilled. This feature is often called extraction time, because during this time the sample water is extracted from the matrix.

#### *4.2. Long stop delay time*

When a stop delay time longer than usual is fixed (for instance 30 s instead of 5 or 10 s), a too early ending of the determination can be avoided. The same effect can be obtained by working with the drift as stop criterion.

#### *4.3. External extraction*

The extraction of the water can be carried out before the titration for a longer time. The extraction vessel can then be used as titration cell (in an automated system) or an aliquot can be taken for titration (also possible in an automated system).

#### *4.4. Particle size*

The smaller the particle size is, the shorter are the distances for the water to reach the working medium. The sample can therefore be cut, milled or ground before being put in the titration cell. It is, however, possible that by this procedure the original water content is changed, because water may be lost or adsorbed by the newly created surface. It is therefore preferable to process the sample within the titration cell. This can be done by using an internal homogeniser which is introduced through the lid through an additional opening (Isengard and Nowotny 1991).

#### *4.5. Elevated temperature*

The water migration can be accelerated by working at elevated temperatures (Isengard and Schmitt 1995). In special titration cells

with an additional reflux condenser closed by a drying tube, titrations are even possible at the boiling point of the working medium (Isengard and Striffler 1992). The water can also be evaporated in a tubular oven from where the vapour is led into the cell by a stream of dried gas. This procedure may, however, be critical, as additional water may be produced at high temperatures by decomposition reactions of components in the sample.

#### *4.6. Polarity of the working medium*

The main component of the working medium is methanol. When a polar sample like sugars is to be analysed, an additional solvent that increases the polarity can be added, such as formamide. In the opposite case a non-polar additional component is recommended, such as chloroform or a long-chained alcohol. Methanol can also be replaced by another alcohol, but it may then be necessary to change the stop criteria (polarising current of the electrodes and stop voltage) (Langer and Isengard 1998, Wünsch and Grünke 1998, Schöffski 1998).

In all these cases it must be checked if the Karl Fischer reaction is not disturbed. This can be done by measuring the water equivalent of the titrating solution. It must be the same as for the standard working medium.

#### *4.7. Combination of measures*

The above mentioned measures to accelerate the liberation of the water can usually be applied together. Thus, it is possible to titrate a sample using an internal homogeniser at the boiling point of the working medium consisting of methanol and chloroform.

#### *4.8. Special situations*

The above mentioned measures aim at a complete detection of the water in the sample. In some situations, however, not the total water content is to be determined but only the “free” surface water. In these cases the dissolution of the sample must be avoided. This means that just the opposite measures must be taken (Isengard and Heinze 2003).

### **The Karl Fischer titration as reference method**

Because of its selectivity for water the Karl Fischer titration is the ideal reference method for water determination (Isengard and Kerwin

2003). Nevertheless, many standards and reference methods are based on drying techniques. These methods do however not necessarily determine water specifically and completely. The result of drying, no matter which technique is applied, is not water content but the mass loss the sample undergoes under the conditions applied. No distinction is made between water and other volatile compounds. Volatile material can even be produced by chemical reactions during heating. Strongly bound water, on the other hand, may escape detection.

### References

- Fischer, K. (1935). Neues Verfahren zur maßanalytischen Bestimmung des Wassergehaltes von Flüssigkeiten und festen Körpern. *Angew. Chemie* 48, 394-396.
- Grünke, S., Wünsch, G. (2000). Kinetics and stoichiometry in the Karl Fischer solution. *Fresenius J. Anal. Chem.* 368, 139-147.
- Isengard, H.-D. (1995). Rapid water determination in foodstuffs. *Trends Food Sci. Technol.* 6, 155-162.
- Isengard, H.-D., Heinze, P. (2003). Determination of total water and surface water in sugars. *Food Chemistry* 82, 169-172
- Isengard, H.-D., Kerwin, H. (2003). Proposal of a new reference method for determining the water content of butteroil. *Food Chemistry* 82, 117-119
- Isengard, H.-D., Nowotny, M. (1991). Dispergierung als Vorbereitung für die Karl-Fischer-Titration. *Dtsch. Lebensm.-Rundsch.* 87, 176-180.
- Isengard, H.-D., Schmitt, K. (1995). Karl Fischer titration at elevated temperatures. *Mikrochim. Acta* 120, 329-337.
- Isengard, H.-D., Striffler, U. (1992). Karl Fischer titration in boiling methanol. *Fresenius J. Anal. Chem.* 342, 287-291
- Langer, N., Isengard, H.-D. (1998). 1-Propanol, a new working medium for the Karl Fischer titration. In *Papers and Abstracts from the Third International Symposium on Humidity and Moisture, 6-8 April 1998, London, England*, (Vol. 2, pp 143-144), Teddington, Middlesex, United Kingdom: National Physical Laboratory.
- Schöffski, K. (1998). Der lange Weg zur giftfreien Karl-Fischer-Titration. *GIT – Fachz. Lab.* 42, 681-684
- Scholz, E. (1984). *Karl Fischer titration*. Berlin: Springer-Verlag.
- Wünsch, G., Grünke, S. (1998). 1-Propanol als Reaktionsmedium für die Wasserbestimmung nach Karl Fischer. *GIT – Fachz. Lab.* 42, 504-507.