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Water quality — Sampling —

Part 3:

Guidance on the preservation and handling of
samples

Qualité de l'eau — Échantillonnage —

*Partie 3: Guide général pour la conservation et la manipulation des
échantillons*



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Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

International Standard ISO 5667-3 was prepared by Technical Committee ISO/TC 147, *Water quality*, Subcommittee SC 6, *Sampling (general methods)*.

This second edition cancels and replaces the first edition (ISO 5667-3:1985), of which it constitutes a technical revision.

ISO 5667 consists of the following parts, under the general title *Water quality — Sampling*:

- *Part 1: Guidance on the design of sampling programmes*
- *Part 2: Guidance on sampling techniques*
- *Part 3: Guidance on the preservation and handling of samples*
- *Part 4: Guidance on sampling from lakes, natural and man-made*
- *Part 5: Guidance on sampling of drinking water and water used for food and beverage processing*
- *Part 6: Guidance on sampling of rivers and streams*
- *Part 7: Guidance on sampling of water and steam in boiler plants*
- *Part 8: Guidance on the sampling of wet deposition*

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- *Part 9: Guidance on sampling from marine waters*
- *Part 10: Guidance on sampling of waste waters*
- *Part 11: Guidance on sampling of groundwaters*
- *Part 12: Guidance on sampling of bottom sediments*
- *Part 13: Guidance on sampling of sewage, waterworks and related sludges*

Annex A of this part of ISO 5667 is for information only.

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Introduction

This part of ISO 5667 is intended to be used in conjunction with ISO 5667-1 and ISO 5667-2 which deal respectively with the design of sampling programmes and sampling techniques.

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Water quality — Sampling —

Part 3:

Guidance on the preservation and handling of samples

1 Scope

This part of ISO 5667 gives general guidelines on the precautions to be taken to preserve and transport water samples.

These guidelines are particularly appropriate when a sample (spot or composite sample) cannot be analysed on site and has to be transported in order to be analysed in the laboratory.

2 Normative references

The following standards contain provisions which, through reference in this text, constitute provisions of this part of ISO 5667. At the time of publication, the editions indicated were valid. All standards are subject to revision, and parties to agreements based on this part of ISO 5667 are encouraged to investigate the possibility of applying the most recent editions of the standards indicated below. Members of IEC and ISO maintain registers of currently valid International Standards.

ISO 5667-1:1980, *Water quality — Sampling — Part 1: Guidance on the design of sampling programmes*.

ISO 5667-2:1991, *Water quality — Sampling — Part 2: Guidance on sampling techniques*.

ISO 5667-8:1993, *Water quality — Sampling — Part 8: Guidance on the sampling of wet deposition*.

3 Preservation of samples

3.1 General considerations

Waters, particularly surface waters and above all waste waters, are susceptible to being changed to differing extents as a result of physical, chemical or biological reactions which may take place between the time of sampling and the analysis. The nature and rate of these reactions are often such that, if the necessary precautions are not taken before and during transport as well as during the time in which the samples are preserved in the laboratory before being analysed, the concentrations determined will be different from those existing at the time of sampling.

It should be stressed that, particularly if there is any doubt, the analyst and the scientist interpreting the results should be consulted before deciding on the precise method of handling and preservation.

The causes of variations are numerous; some of these are as follows:

- Bacteria, algae and other organisms can consume certain constituents present in the samples; they can also modify the nature of the constituents to produce new constituents. This biological activity affects for example the contents of dissolved oxygen, carbon dioxide, nitrogen compounds, phosphorus and sometimes silicon.
- Certain compounds can be oxidized by the dissolved oxygen contained in the samples or by atmospheric oxygen [for example organic compounds, iron(II), sulfides].

- Certain substances can precipitate out [for example calcium carbonate, metals and metallic compounds such as $\text{Al}(\text{OH})_3$, $\text{Mg}_3(\text{PO}_4)_2$] or be lost to the vapour phase (for example oxygen, cyanides, mercury).
- The pH, conductivity, carbon dioxide content, etc. can be modified by the absorption of carbon dioxide from the air.
- Metals dissolved or in a colloidal state as well as certain organic compounds can be adsorbed or absorbed irreversibly on the surface of containers or solid materials contained in the samples.
- Polymerized products can depolymerize; conversely, simple compounds can polymerize.

The extent of these reactions is a function of the chemical and biological nature of the sample, its temperature, its exposure to light, the nature of the container in which it is placed, the time between sampling and analysis, the conditions (for example rest or agitation during transport) to which it is submitted, etc.

It follows that the variations relative to a particular constituent vary both in degree and rate, not only as a function of the type of water, but also, for the same type, as a function of seasonal conditions.

It must be emphasized moreover that these variations are often sufficiently rapid to modify the sample considerably in the space of several hours. In all cases, it is therefore essential to take the necessary precautions to minimize these reactions and, in the case of many parameters, to analyse the sample with a minimum of delay.

As the variations which take place in the water samples are due to a large extent to biological processes, it is generally necessary to choose, from the various possible methods of preservation, a method that does not introduce unacceptable contamination.

Even the time for which the preserved sample can be stored before being analysed may change.

As a guide, it can be said that methods of preservation tend to be less effective in the case of crude sewage than in the case of purified sewage (effluents from biological treatment plants). It has also been observed that the behaviour of various waste water samples during storage is different depending on whether the samples have been taken from municipal or industrial sewage-treatment plants.

On the other hand, surface waters and ground waters can in general be stored more effectively. In the case of potable waters, the problem of storage can be solved more easily because these waters are less susceptible to biological and chemical reactions.

Therefore, owing to these variations, which may affect the water samples, it may be necessary, in certain determinations, to take individual samples rather than collective samples and to analyse them immediately at the place of sampling. It should be remembered that the storage of samples for long periods is only possible for the determination of a limited number of parameters.

In spite of numerous investigations which have been carried out in order to recommend methods which will enable water samples to be stored without modification of their composition, it is impossible to give absolute rules in this context which will cover all cases and all situations and which do not have exceptions.

In every case, the method of storage should be compatible with the various analytical techniques for which it will be used. One object of this part of ISO 5667 is to describe the most commonly used techniques.

3.2 Feasible precautions

3.2.1 Filling the container

In the case of samples for the determination of physico-chemical parameters, one simple precaution, which is not however adequate in all cases, is to fill the flasks completely and stopper them in such a way that there is no air above the sample.

This limits interaction with the gas phase and agitation during transport (thus avoiding modifications in carbon dioxide content, and hence variations in pH; hydrogen carbonates are not converted into precipitable carbonates; iron has less tendency to be oxidized, thus limiting colour variations; etc.).

For microbiological examination, the sample container should not be filled to the brim so that an air space is left after insertion of the stopper. This aids mixing before examination and avoidance of accidental contamination.

Sample containers, whose contents are frozen as part of their preservation, should not be completely filled (see 3.2.4).

3.2.2 Use of appropriate containers

The choice and preparation of a container can be of major importance. ISO 5667-2 provides some guidance on this subject.

However, it is essential that the container in which the sample is stored and the stopper should not

- be a cause of contamination (for example borosilicate or soda-lime glass containers may increase the content of silica or sodium);
- absorb or adsorb the constituents to be determined (for example hydrocarbons may be absorbed in a polyethylene container, traces of metals may be adsorbed on the surface of a glass container, which can be prevented by acidifying the sample);
- react with certain constituents in the sample (for example fluorides reacting with glass).

It should be remembered that the use of opaque containers or brown (non-actinic) glass containers can reduce the photosensitive activities to a considerable extent.

It is preferable to reserve a set of containers for a particular determinand, thereby minimizing risks of cross-contamination. However, care is necessary in any case to prevent bottles which formerly held a high concentration of a determinand from contaminating subsequent low contamination samples. Disposable containers should be considered, if economic, to prevent this type of contamination, but they are not suitable for such specific parameters as organochlorine pesticides.

Blank samples containing distilled water should always be taken, preserved and analysed as a check on the suitability of the choice of container and cleaning procedure.

When sampling solid or semi-solid samples, jars or wide-mouthed bottles should be used.

3.2.3 Preparation of containers

3.2.3.1 For samples for chemical analysis

For analysis of trace quantities of chemical constituents of surface or waste water, it is usual to clean new containers thoroughly in order to minimize possible contamination of the sample; the type of cleaner used and the container material vary according to the constituents to be analysed.

In general, new glassware should be rinsed with water containing a detergent in order to remove dust and residues of packing material, followed by thorough rinsing with distilled or deionized water. For general trace analysis, the bottles should be filled with a 1 mol/l solution of nitric acid or hydrochloric acid and left to soak for at least one day, followed by rinsing with distilled or deionized water.

For the determination of phosphates, silicon, boron and surfactants, detergents should not be used for cleaning purposes. For trace analysis of organic material, special pretreatment of the bottles may be necessary and reference should be made to the relevant International Standard (also see 3.2.3.2).

3.2.3.2 For samples for determination of pesticides, herbicides and their residues

In general, glass (preferably brown) containers should be used because plastics, except polytetrafluoroethylene (PTFE), may introduce interferents which can be significant if trace analyses are to be performed.

All containers should be cleaned with water and detergent, followed by thorough rinsing with distilled or deionized water, then oven dried at 105 °C for 2 h and cooled before being rinsed with the extraction solvent used during the analysis. Finally they should be dried with a stream of carefully purified air or nitrogen.

In addition, for containers that have already been used, an extraction with acetone for 12 h, followed by a hexane rinse and drying as described in the previous paragraph, should also be used.

3.2.3.3 For samples for microbiological analysis

The containers should be able to withstand a sterilization temperature of 175 °C for 1 h and should not produce or release at this temperature any chemicals which would either inhibit biological activity, induce mortality or encourage growth.

When lower sterilization temperatures are used (e.g. steam sterilization), polycarbonate and heat-resistant polypropylene containers may be used. Caps or other stoppers should withstand the same sterilization temperatures as the containers.

It is essential that the containers be free of acidic, alkaline and toxic compounds. Glass containers should be cleaned with water and detergent, followed by thorough rinsing with distilled water. They should also be rinsed with nitric acid (HNO₃) 10 % (V/V) followed by thorough rinsing with distilled water in order to remove any heavy metals or chromate residues.

If the samples contain chlorine, sodium thiosulfate ($\text{Na}_2\text{S}_2\text{O}_3$) should be added, before sterilization (see table 3). This is to eliminate inactivation of bacteria by chlorine.

3.2.4 Cooling or freezing of the samples

The sample should be kept at a temperature lower than that during filling. Containers should be almost, but not completely, filled.

It should be emphasized that cooling or freezing of samples is only truly effective if it is applied immediately after the collection of the samples. When possible, this necessitates the use of cool-boxes or refrigerators in vehicles at the sampling site.

3.2.4.1 Simple cooling (in melting ice or in a refrigerator between 2 °C and 5 °C) and storage of the sample in the dark are, in most cases, sufficient to preserve the sample during transport to the laboratory and for a relatively short period of time before the analysis. Cooling cannot be considered as a means of long-term storage, particularly in the case of waste water samples (see table 1).

3.2.4.2 In general, freezing (– 20 °C) allows an increase in the period of storage. Nevertheless, it is necessary to control the freezing and thawing technique fully in order to return the sample to its initial equilibrium after thawing. In this case, the use of plastics containers (for example polyvinyl chloride) is strongly recommended.

Glass containers are not suitable for freezing. Samples for microbiological analysis should not be frozen.

3.2.5 Filtration or centrifuging of samples

Suspended matter, sediment, algae and other micro-organisms may be removed, either at the time of taking the sample or immediately afterwards, by filtration of the samples, through filter paper or membrane filter or by centrifuging. Filtration is, of course, not applicable if the filter is likely to retain one or more of the constituents to be analysed. Equally, it is essential that the filter is not a cause of contamination and is carefully washed before use, but in a manner consistent with the final method of analysis.

Alternatively, the reason for analysis may involve the separation of soluble and insoluble forms (for example of a metal) by filtration.

Membranes should be used with caution as various heavy metals and organic material may be adsorbed on the membrane surface, and soluble compounds

within the membrane can be leached out into the sample.

3.2.6 Addition of preservatives

Certain physical and chemical constituents can be stabilized by the addition of chemical compounds, either directly to the sample after taking it, or beforehand, to the container when it is still empty.

Various chemical compounds, at concentrations equally varied, have been proposed.

The most commonly used are

- acids;
- basic solutions;
- biocides;
- particular reagents, necessary for the specific preservation of certain constituents [for example the determination of oxygen, total cyanides and sulfides requires a previous fixation of the sample on site (see the relevant International Standards on analysis)].

WARNING — The use of mercury(II) chloride (HgCl_2) and phenyl mercury(II) acetate ($\text{CH}_3\text{CO}_2\text{HgC}_6\text{H}_5$) should be avoided.

It should be remembered that certain preservatives (for example acids, chloroform) need to be used with caution, considering the danger involved in their handling. Operators should be warned of these dangers and the ways of protecting themselves from them.

It is essential that the preservatives used do not interfere during the determination; tests intended to check their compatibility are necessary in cases of doubt. Any dilution of the sample with added preservatives should be taken into account during the analysis and the calculation of results.

It is preferable that the addition of preservatives be made using sufficiently concentrated solutions so that only small volumes are necessary. This enables the corresponding dilution to be disregarded in most cases.

The addition of these agents can also modify the chemical or physical nature of the constituents and it is therefore necessary that these modifications are not incompatible with the objects of later determinations. (For example, acidification can solubilize colloidal constituents or solids and should therefore only be used with caution if the aim of the measure-

ments is the determination of dissolved constituents. If the aim of the analysis is to determine the toxicity to aquatic animals, the solubilization of certain components, particularly heavy metals which are toxic in ionic form, has to be avoided. Samples should therefore be analysed as soon as possible.)

It is essential to carry out a blank test, particularly determinations of trace elements, to take into account possible introduction by the preservatives of an additional amount of the elements to be determined (for example acids can introduce a not insignificant amount of arsenic, lead and mercury). In such a case, samples of the preservatives used for the treatment of the water samples should be retained for use in the preparation of blank tests.

3.3 Recommendations

As stated in 3.1, it is impossible to give absolute rules for preservation; the duration of preservation, the nature of the container and the efficiency of the preservation processes depend not only on the constituents which have to be analysed and their levels, but also on the nature of the sample. The tables should therefore be considered as giving reasonable suggestions only.

In any case in question, there needs to be no significant difference between the results of a determination carried out immediately and the result obtained after preservation; each analyst should therefore verify, taking into account particularly the method of analysis which he intends to use, whether the suggestions in tables 1 to 5 are suitable for the sample with which he is concerned.

In addition, International Standards describing the methods of analysis do, wherever possible, indicate the recommended methods of preservation.

Moreover, given that incompatibility can exist between the analyses to be carried out and the various preservatives and containers possible, it is often necessary to take several samples of the same water and to treat each of them in relation to the analyses for which they are intended. This may result in a compromise between the techniques of preservation which would be most appropriate for each determination taken in isolation. The choice of sample preservation procedure should always be the subject of consultation with the analyst.

4 Identification of samples

Containers holding the samples should be marked in a clear and durable manner in order to permit identification without ambiguity in the laboratory.

Additionally, it is generally necessary to note, at the moment of sampling, numerous details which will permit a correct interpretation of the information obtained (date and hour of sampling, name of person sampling, nature and amount of preservatives added, etc.). Various processes (labels, forms, etc.) allow the practical attainment of these two objectives.

Special samples of anomalous material should be clearly marked and accompanied by a description of the observed anomaly. It is essential that samples containing hazardous or potentially hazardous materials, for example acids, are clearly identified as such.

5 Transport of samples

It is obvious that containers holding samples must be protected and sealed in such a way that they do not deteriorate and do not lose any part of their contents during transport. Packaging should protect the containers from possible external contamination and breakage, particularly near the opening, and should not itself be a source of contamination. During transportation, the samples should be kept as cool as practicable and protected from light, with each sample placed inside an individual waterproof container if possible.

If the time of travel exceeds the maximum recommended preservation time before analysis, then the samples should still be analysed, and the time between sampling and analysis reported, after consultation with the scientist interpreting the analytical results.

6 Reception of samples in the laboratory

On their arrival in the laboratory, the samples should, if their immediate analysis is impossible, be preserved under conditions such that any contamination of the outside of the containers is avoided and which prevent any change in their contents.

The use, for this purpose, of refrigerated cabinets or cool and dark places is highly recommended.

In all cases, and especially when a chain of custody needs to be established, it is recommended that the count of sample containers received be verified against the record of the number of sample bottles sent for each sample.

Table 1 — Techniques generally suitable for the preservation of samples — Physico-chemical and chemical analysis

The information in table 1 is only a general guide to the preservation of samples. The complex nature of natural and waste waters necessitates, before analysis, a verification of the stability of each type of sample treated according to the methods proposed in table 1.

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Acidity and alkalinity	P or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h	Samples should preferably be analysed at the spot where the sample is taken (particularly for samples high in dissolved gases).	
Aluminium dissolved ¹⁾	P	Filtration at the place of sampling and acidification of the filtrate to pH < 2	Laboratory	1 month	The dissolved ¹⁾ aluminium and that adhering to suspended matter can be determined from the same sample.	
total		Acidification to pH < 2	Laboratory	1 month		
Ammonia, free and ionized	P or G	Acidification to pH < 2 with H ₂ SO ₄ , cooling to between 2 °C and 5 °C	Laboratory	24 h		ISO 5664 ^[2] ISO 6778 ^[23] ISO 7150 ^[26] ^[27]
		Cooling to between 2 °C and 5 °C	Laboratory	6 h		
AOX (absorbable organic halides)	G	Acidification to pH < 2 with nitric acid, cooling to between 2 °C and 5 °C, storage in the dark	Laboratory	3 days	Analyse as soon as possible. Refer to relevant International Standard for details for particular types of water.	ISO 9562 ^[55]
Arsenic	P or G	Acidification to pH < 2	Laboratory	1 month	HCl should be used if the hydride technique is used for analysis.	ISO 6595 ^[19]
Barium	P or BG	See aluminium			Do not use H ₂ SO ₄ .	
BOD (biochemical oxygen demand)	P or G (Glass is preferable in the case of low BOD)	Cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	24 h		ISO 5815 ^[8]
Boron and borates	P		Laboratory	1 month		ISO 9390 ^[52]

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Bromides and bromine compounds	P or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h	Samples should be kept out of direct sunlight.	
Cadmium	P or BG	See aluminium				ISO 5961 [9]
Calcium	P or G	—	Laboratory	24 h	48 h may be possible but exercise caution for samples of conductivity above 70 mS/m.	ISO 6058 [10]
		Acidification to pH < 2	Laboratory	1 month	Acidification (do not use H ₂ SO ₄) permits determination of the calcium from the same sample as the other metals.	ISO 6059 [11] ISO 7980 [41]
Carbon dioxide	P or G	—	On site	—		
Carbon, organic	G	Acidification to pH < 2 with H ₂ SO ₄ , cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	1 week	The preservation technique will depend on the method of analysis to be used. The test should be carried out as soon as possible.	ISO 8245 [42]
	P	Freezing to – 20 °C	Laboratory	1 month	Freezing (– 20 °C) may be used in certain cases.	
Chlorides	P or G	—	Laboratory	1 month		ISO 9297 [50]
Chlorine, residual	P or G	—	On site	—	Transport in dark. The analysis should be carried out as soon as possible.	ISO 7393 [28] [29] [30]
Chlorophyll	P or G	Cooling to 4 °C	Laboratory	24 h	Transport in dark.	
		After filtration and freezing of the residue	Laboratory	1 month		
Chromium(VI)	P or BG	Cooling to between 2 °C and 5 °C	Laboratory	24 h		
Chromium, total	P or BG	See aluminium				ISO 9174 [48]
Cobalt	P or BG	See aluminium				ISO 8288 [44]

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
COD (chemical oxygen demand)	P or G (Glass is preferable in the case of low COD)	Acidification to pH < 2 with H ₂ SO ₄ , cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	5 days		ISO 6060 [12]
	P	Freezing to – 20 °C	Laboratory	1 month		
Colour	P or G	—	On site	—		ISO 7887 [34]
		Cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	24 h		
Conductivity	P or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h	The test should preferably be carried out on site.	ISO 7888 [35]
Copper	P or BG	See aluminium				ISO 8288 [44]
Cyanides, easily liberated	P	The preservation technique will depend on the method of analysis to be used.				ISO 6703-2 [21]
Cyanides, total	P	The preservation technique will depend on the method of analysis to be used.				ISO 6703-1 [20]
Detergents	See surfactants					
Dry residue	See total residue					
Fluorides	P but not PTFE	—	Laboratory	1 month		
Greases, oils, hydrocarbons	Glass washed with solvent (e.g. pentane) used for extraction	Extraction on site where practicable and cooling to between 2 °C and 5 °C	Laboratory	24 h	It is recommended that, immediately after sampling, the extraction agent used in the method of analysis or extraction be added, or the extraction be carried out, on site (but local safety regulations should be followed).	ISO 10359-1 [63]
Heavy metals (except mercury)	P or BG	See aluminium				ISO 8288 [44]
Hydrazine	G	Acidification with HCl to 1 mol/l (100 ml per litre of sample) and storage in the dark	Laboratory	24 h		
Hydrocarbons	See greases					
Hydrogencarbonates	See alkalinity					

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Iodides	Glass	Cooling to between 2 °C and 5 °C	Laboratory	24 h	Samples should be kept out of direct sunlight.	
		Alkalization to pH 11	Laboratory	1 month		
Iron(II)	P or BG	Acidification to pH < 2 with HCl and exclusion of atmospheric oxygen	On site or in laboratory	24 h		
Iron, total	P or BG	See aluminium				ISO 6332 [14]
Kjeldahl (nitrogen)	P or BG	Acidification to pH < 2 with H ₂ SO ₄ , cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	24 h	Do not acidify if free ammonia is to be measured on same sample.	ISO 5663 [1]
Lead	P or BG	See aluminium			Do not use H ₂ SO ₄ .	ISO 8288 [44]
Lithium	P	—	Laboratory	1 month		
		Acidification to pH < 2	Laboratory	1 month	Acidification permits determination of the lithium from the same sample as the other metals.	
Magnesium	P or BG	See calcium				ISO 6059 [11] ISO 7980 [41]
Manganese	P or BG	See aluminium				ISO 6333 [15]
Mercury, total	BG	Acidification to pH < 2 with HNO ₃ and addition of K ₂ Cr ₂ O ₇ [0,05 % (m/m) final concentration]	Laboratory	1 month	Particular care is needed to ensure that the sample containers are free from contamination.	ISO 5666 [3] [4] [5]
Nickel	P or BG	See aluminium				ISO 8288 [44]
Nitrate	P or G	Acidification to pH < 2 or cooling to between 2 °C and 5 °C	Laboratory	24 h		ISO 7890 [36] [37] [38]
		On-site filtration with a membrane filter of pore size 0,45 µm and cooling to between 2 °C and 5 °C	Laboratory	48 h	For ground and surface waters	
Nitrite	P or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h		ISO 6777 [22]

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Odour	G	Cooling to between 2 °C and 5 °C	Laboratory (for quantitative analysis)	6 h	The test can be carried out on site (qualitative analysis).	
Organic chlorine	See AOX					
Orthophosphates, total	B or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h	The analysis should be carried out as soon as possible.	ISO 6878-1 [24]
Orthophosphates, dissolved	B or G	The sample should be filtered on site at the time of sampling. Cooling to between 2 °C and 5 °C	Laboratory	24 h	The analysis should be carried out as soon as possible.	ISO 6878-1 [24]
Oxygen	P or G	—	On site	—		ISO 5813 [6] ISO 5814 [7]
	G	Fixing of the oxygen on site and storage in the dark	Laboratory	4 days at most	Fix the oxygen in accordance with the method of analysis used.	
Ozone	—	—	On site	—		
Permanganate index	G	Acidification to pH < 2 with H ₂ SO ₄ , cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	2 days	Analyse as soon as possible. Acidification in accordance with the start of the analytical method can be a useful preservation technique.	ISO 8467 [47]
	P	Freezing to -20 °C	Laboratory	1 month		
Pesticides, organochlorine	G (washed with solvent)	Cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	24 h	It is recommended that, immediately after sampling, the extraction agent used in the method of analysis be added, or that extraction be carried out on site.	
Pesticides, organophosphorus	G (washed with solvent)	Cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	24 h	Extraction should be commenced as soon as possible after sampling, preferably within 24 h.	
Petroleum and derivatives	See greases, oils and hydrocarbons					
pH	P or G	—	On site		The test should be carried out as soon as possible and preferably immediately on site after sampling.	
		Transportation at a lower temperature than the initial one	Laboratory	6 h		

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Phenol index	BG	Inhibition of biochemical oxidation by CuSO_4 and acidification with H_3PO_4 to $\text{pH} < 2$	Laboratory	24 h	The preservation technique will depend on the method of analysis to be used.	ISO 6439 [16]
Phenols	BG	Cooling to between 2 °C and 5 °C and storage in the dark	Laboratory	24 h	The extraction should be carried out as soon as possible.	
Phosphorus, dissolved	BG or G	Cooling to between 2 °C and 5 °C. Immediate on-site filtration necessary.	Laboratory	24 h	The use of iodized glass containers for samples is recommended when low concentrations are being examined. (A bottle can be iodized by placing a few crystals of iodine into the sealed container, which is then heated to 60 °C for 8 h.) It should be noted that the iodine can leach back into the sample, thus becoming an interferent in the analysis. It is recommended that consultation with the analyst be carried out prior to using this preservation technique.	ISO 6878-1 [24]
Phosphorus, total	BG or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h	See above	
		Acidification to $\text{pH} < 2$ with H_2SO_4	Laboratory	1 month	See above	
Potassium	See lithium					ISO 9964-2 [60] ISO 9964-3 [61]
Selenium	G or BG	Acidification to $\text{pH} < 1$, except if selenides are present. If present, alkalinize to $\text{pH} > 11$ with NaOH.	Laboratory	1 month		ISO 9965 [62]

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Silicates, dissolved	P	Filtration at the place of sampling, acidification to pH < 2 with H ₂ SO ₄ and cooling to between 2 °C and 5 °C	Laboratory	24 h		
Silicates, total	P	As for dissolved silicates	Laboratory	24 h		
Silver	P or BG	See aluminium			Do not use HCl. Some forms of silver need addition of cyanide for stabilization.	
Sodium	See lithium					ISO 9964-1 [59] ISO 9964-3 [61]
Sulfates	P or G	Cooling to between 2 °C and 5 °C	Laboratory	1 week	With waste water note that hydrogen sulfide may be formed; therefore hydrogen peroxide should be added to the sample. For samples with high BOD (e.g. > 200 mg/l) hydrochloric acid should be added instead, noting the possible hazard of released hydrogen sulfide.	ISO 9280 [49]
Sulfides (easily liberated)	P or G	Fix samples immediately on site by alkalization, if necessary, with sodium carbonate followed by addition of zinc acetate.	Laboratory	24 h	Stabilize according to the relevant International Standard.	
Sulfides	P or G	Fix samples immediately on site by alkalization, if necessary, with sodium carbonate followed by addition of zinc acetate. Fill sample bottle to exclude air completely.	Laboratory	—	Analyse as soon as possible. Stabilize according to the relevant International Standard.	

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Sulfites	P or G	Fixing on site by addition of 1 ml of a 2,5 % (m/m) solution of EDTA per 100 ml of sample	Laboratory	48 h		
Surfactants, cationic	G	Cooling to between 2 °C and 5 °C	Laboratory	48 h	Rinse glass containers as described in ISO 7875-1 and ISO 7875-2. Analyse samples as soon as possible. To prevent adsorption on the container wall, add on site 5 mg/l of a linear alkylethoxylated nonionic surfactant.	
Surfactants, anionic	G	Acidification to pH < 2 with H ₂ SO ₄ and cooling to between 2 °C and 5 °C	Laboratory	48 h	Rinse glass containers as described in ISO 7875-1. Analyse samples as soon as possible.	ISO 7875-1 [32]
Surfactants, nonionic	G	Addition of 40 % (V/V) formaldehyde to give a 1 % (V/V) solution; cool to between 2 °C and 5 °C and ensure sampling container is completely filled.	Laboratory	1 month	Rinse glass containers as described in ISO 7875-2. Analyse samples as soon as possible.	ISO 7875-2 [33]
Suspended and sedimentary matter	P or G	—	Laboratory	24 h	The test should be carried out as soon as possible and preferably on site.	
Tin	P or BG	See aluminium			Do not use HNO ₃ . If organo-tin is present, use acetic acid for preservation for total tin analysis, but if speciation is required then freeze and analyse as soon as possible.	
Total hardness	See calcium					

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Total residue (dry extract)	P or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h		
Turbidity	P or G	—	Laboratory	24 h	The test should preferably be carried out on site.	ISO 7027 [25]
Uranium	P or BG	See aluminium				
Zinc	P or BG	See aluminium				ISO 8288 [44]
1) Dissolved: Denotes that which passes through a filter of pore size 0,45 µm.						

Table 2 — Allocation of parameters to the types of preservation (annex to table 1)

Allocating the corresponding parameters to one of the non-specific types of preservation given in table 2 is intended to help the user to select a type of preservation which is effective for several parameters simultaneously, if necessary. However, limited applicability should be checked in each individual case on the basis of the data for individual materials. Parameters not listed in table 2 should not normally be preserved using these methods.

Preservation by	Suitable for	Not suitable for
Acidification to pH < 2	Alkaline metals Aluminium Ammonia (but not if separate free and total ammonia analyses are required) Arsenic Alkaline earth metals Nitrate Total hardness Phosphorus, total Heavy metals	Cyanides Sulfides Carbonates, bicarbonates, carbon dioxide Sulfites, sulfur dioxide Thiosulfates Nitrites Phosphonates (if speciation is required) Soaps and esters Hexamethylenetetramine Do not use sulfuric acid for calcium, strontium, barium, radium, lead. Do not use hydrochloric acid for silver, thallium, lead, bismuth, mercury(I) and antimony. Do not use nitric acid for tin.

Preservation by	Suitable for	Not suitable for
Alkalization to pH > 11	Iodides	Most organic compounds, heavy metals, especially in lower valency states. Some metals form soluble anions at higher valency states. (Depending on the anion present consult solubility tables.) Ammonia/ammonium Amines and amides Phosphorus, total Hydrazine Hydroxylamine
Cooling to 2 °C to 5 °C	Acidity, alkalinity Ammonium Bromide and bromine compounds Chlorophyll Iodides Kjeldahl (nitrogen) Conductivity Nitrate Nitrite Odour Orthophosphates Phosphorus Sulfates Surfactants, cationic Dry residue Total residue Biological tests	
Deep freezing (– 20 °C)	Chlorophyll COD Biological tests, toxicity tests Organic carbon Permanganate index	Not suitable for biota if a distinction is to be made between liquid content and biota cell content Dissolved gases Microorganisms for identification Changes can also occur in many solutes requiring homogenization after thawing. Precipitation (and polymerization) can occur making resolution difficult. Conversely some polyacids depolymerize. Suitability should be evaluated before routine use.

Table 3 — Techniques generally suitable for the preservation of samples — Microbiological analysis

Parameter to be studied	Type of container	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis	Comments	International Standard (The numbers refer to annex A.)
Enumeration of total bacteria Total coliforms Thermotolerant coliforms Faecal streptococci Salmonella Shigella etc.	Sterile container	Cooling to between 2 °C and 5 °C	Laboratory	8 h (drinking water, surface water, ground water and sludges)	For chlorinated or brominated water the sample should be collected in a flask containing (before it is sterilized) sodium thiosulfate [in general 0,1 ml of a 10 % (m/m) solution of Na ₂ S ₂ O ₃ per 125 ml of sample]. For waters containing heavy metal concentrations greater than 0,01 mg/l, add to the container (before it is sterilized) 0,3 ml of 15 % (m/m) NTA per 500 ml of sample. (Also see 3.2.6.)	ISO 6222 [13] ISO 6461 [17] [18] ISO 7899 [39] [40] ISO 8360 [45] [46] ISO 9308 [51] [52]

Table 4 — Techniques generally suitable for the preservation of samples — Biological analysis

The biological parameters to be determined are generally numerous and may sometimes vary from one biological species to another. For this reason, it is impossible to draw up an exhaustive check-list of all the precautions that should be taken to preserve samples for this type of analysis. The information in table 4 therefore only relates to certain parameters generally studied for various animal or vegetable groups.

It should be noted that before carrying out any detailed study, it is essential to choose the parameters of interest.

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Counting and identification Benthic macro-invertebrates — large samples	P or G	Addition of 70 % (V/V) ethanol	Laboratory	1 year	Water in samples should first be decanted to maximize the preservative concentration.	ISO 7828 [31] ISO 8265 [43] ISO 9391 [54]
— small samples (for example reference collections)	G	Addition of appropriate volume of 40 % (V/V) formaldehyde neutralized with sodium borate to give a 2 % (V/V) to 5 % (V/V) solution Transfer to a preservative solution consisting of 70 % (V/V) ethanol, 40 % (V/V) formaldehyde and glycerol (in the proportions 100 + 2 + 1 respectively).	Laboratory	Indefinitely		

WARNING — Beware of formaldehyde vapours — Do not store large numbers of samples in work areas.

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Periphyton	G	Addition of 1 part by volume of Lugol's solution to 100 parts by volume of sample. (Lugol's solution is made up of 20 g of potassium iodide and 10 g of iodine per litre, stored in a dark glass bottle.)	Laboratory	1 year	Store samples in the dark.	
Phytoplankton	G	See periphyton	Laboratory	1 year	Store samples in the dark.	
Zooplankton	G	Addition of 40 % (V/V) formaldehyde to give 4 % (V/V) formalin, or addition of Lugol's solution as for periphyton	Laboratory	1 year	Addition of more Lugol's solution may be necessary if decolorization occurs.	
Fresh and dry mass						
Benthic macro-invertebrates	P or G	Cooling to between 2 °C and 5 °C	On site or in the laboratory	24 h	Do not freeze to – 20 °C. The analysis should be carried out as soon as possible and not later than 24 h.	
Macrophytes						
Periphyton						
Phytoplankton						
Zooplankton						
Fish		—	On site			
Mass of ash						
Benthic macro-invertebrates	P or G	Filtration and cooling to between 2 °C and 5 °C	Laboratory	2 weeks		
Macrophytes						
Periphyton						
Phytoplankton						

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Toxicity tests	P or G	Cooling to between 2 °C and 5 °C	Laboratory	24 h	The preservation period will vary according to the method of analysis to be used.	
		Freezing to - 20 °C	Laboratory	2 weeks		

3:1994

1) Schwoerbel, J. *Methoden der Hydrobiologie. "Süßwasserbiologie"*, 3rd edn., Fischer Verlag, Stuttgart, 1980.

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Table 5 — Techniques generally suitable for the preservation of samples — Radiochemical parameters

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Alpha activity Beta activity (except radio-iodine)	P	1. If it is desired to determine the soluble and suspended activity separately, filter immediately. 2. Add 20 ml $\pm$ 1 ml of 50 % (V/V) nitric acid per litre of sample. The pH-value should be less than 1. 3. Store in a dark place at 2 °C to 5 °C.	Laboratory	As soon as possible	Safety precautions and shielding depend on the activity of the sample. WARNING — It is essential that radioactive dust is not inhaled or left on the body or clothing.	ISO 9696 [56] ISO 9697 [57]
Radio-iodine	P Either the bottle should be pre-treated with solid nonradioactive iodine at a minimum of 60 °C until completely coated, then rinsed with ethanol, followed by washing with water until no more iodine is leached out, or sodium iodide should be added as a carrier.	1. Adjust the pH-value to $8,0 \pm 0,1$ with sodium hydroxide solution. 2. Add 0,1 g $\pm$ 0,01 g of nonradioactive sodium iodide per litre of sample. 3. Add 2 ml to 4ml of sodium hypochlorite [10 % (m/m)] per litre of sample, ensuring an excess of free chlorine.	Laboratory	As soon as possible	Samples should not be acidic when the iodide is added. (This is especially important if a joint sample is taken for alpha and beta activity measurement.) Do not use ammonia to make alkaline.	

Parameter to be studied	Type of container P = Plastics (e.g. polyethylene, PTFE, PVC, PET) G = Glass BG = Borosilicate glass	Preservation technique	Place of analysis	Maximum recommended preservation time before analysis (If a preservation period is not specified, it is generally unimportant. The indication "1 month" represents preservations without particular difficulty.)	Comments	International Standard (The numbers refer to annex A.)
Gamma activity (For radon isotopes and radio-iodine, see the separate entry)	P	1. If suspended matter is present and a separate measurement of its activity is required, or the solids are not readily dissolved, filter the sample and treat as two separate samples. 2. Add quantitatively to the sample a known amount of solution containing non-radioactive isotopes of interest. For samples containing metals, the solution is usually acidified to a pH-value of less than 2; the acid used should not precipitate or volatilize the elements of interest. Special requirements are necessary for radon isotopes (see the separate entry). 3. Store in tightly stoppered bottles in the dark at 2 °C to 5 °C.	Laboratory	Depends on the half-life of the radionuclides of interest. The shorter the half-life the sooner the sample needs to be analysed.	Safety precautions and shielding depend on the activity of the sample. WARNING — It is essential that radioactive dust is not inhaled or left on the body or clothing.	