AQUA CHEM

Reference Guide of Compendial Water Standards

Compendial Water Standards

	Purified Water (PW)		Water for Injection (WFI)	
MONOGRAPH	USP 29 <1231>	PhEur 9.1 <0169>	USP 29 <1231>	PhEur 9.1 <0169>
PROCESS*	Distillation, RO and any other suitable process	Distillation, ion exchange, RO & other suitable process	Distillation or RO	Distillation or a Purification Process Equivalent to Distillation
CONDUCTIVITY	< 1.3 µS/cm² @ 25°C	< 4.3 µS/cm² @ 20°C	< 1.3 µS/cm² @ 25°C	< 1.1 µS/cm² @ 20°C
AEROBIC BACTERIA	100 cfu/ml (suggested)	< 100 cfu/ml	< 10 cfu/100 ml (suggested)	< 10 cfu/100 ml
BACTERIAL ENDOTOXIN	N/A	< 0.25 IU/ml (bulk water for dialysis)	< 0.25 EU / ml	< 0.25 IU / ml
TOC	500 ppb	< 0.5 mg/l	500 ppb	< 0.5 mg/l
PH	5.0 - 7.0	5.0 - 7.0	5.0 - 7.0	5.0 - 7.0
NITRATES	N/A	< 0.2 ppm	N/A	< 0.2 ppm
HEAVY METALS	N/A	< 0.1 ppm	N/A	< 0.1 ppm
ALUMINUM	N/A	< 10 ppb (dialysis solutions)	N/A	< 10 ppb (dialysis solutions)

** The water source used for treatment / production must be obtained from potable waters complying with the "U.S. Environmental Protection Agency National Primary Drinking Water Regulations" or equivalent regulations of the European Union or Japan.*

Common Production Methods

Many factors must be considered when selecting an appropriate treatment process for process water; required volume, capital cost, operating cost, maintenance downtime, raw water quality, sanitization methods, and so on. Excluding pre-treatment, common water processes include:

PURIFIED WATER (PW)

- Ion Exchange + 185nm UV + Microbial Filter (0.2 µm)
- Reverse Osmosis + Ion Exchange or Continuous Electrodeionization
- Two-Pass Reverse Osmosis
- Vapor Compression Distillation

WATER FOR INJECTION (WFI)

- Single or Two-Pass Reverse Osmosis + Continuous Electrodeionization + Ultra Filtration
- Reverse Osmosis + Ion Exchange or Continuous Electrodeionization + Multi-Effect Distillation
- Two-Pass Reverse Osmosis + Multi-Effect Distillation
- Vapor Compression Distillation

Bacteria / Bioburden

Bioburden is the contaminant of most concern in a pharmaceutical water system and is quantified in **colony forming units (CFU) per mL**. There is no single method for determining all potential types of microbial contaminants and the distribution and concentration of bacteria can be unpredictable. Left unchecked, **bacteria** will adhere to surfaces and form a **biofilm**, which is difficult to remove and becomes a source of continuous contamination. Counts are representative of 'loose' **bacteria** across key points on a system and are typically cultured in a laboratory on an agar plate. New online instrument approaches include automated laser scanning techniques for faster reporting.

Common Disinfection Methods

- Hot Water Sanitization (72-80 °C for 60 min)
- UV Disinfection (30,000 µW-s/cm2 dose @ 254nm)
- Ozone (50-100 ppb continuous residual, never w/ RO)
- Peracetic Acid (1% solution for 36 min)
- Biofilm Removal (10-12 pH + 30-45 °C for 1-8 hours)

PURIFIED WATER	WATER FOR INJECTION
100 CFU / mL	10 CFU / 100 mL
USP 29 Bioburden Limits	

Endotoxin (<0.25 EU / mL)

Endotoxins are lipopolysaccharides (LPS) from Gram negative bacterial cell-walls that are released upon death/lysis. While they are commonly found everywhere in nature, **endotoxins** generate a fever (pyrogenic) reaction when present in the blood stream making them a concern for parenteral Water for Injection (WFI) systems. The LAL method described in USP 29 <85> is commonly used for testing grab samples for **endotoxin** and reports in **endotoxin units (EU) per mL**.

Endotoxin contamination is managed using strict microbial prevention and may be directly filtered using 10-20 kDa MWCO or better ultrafiltration. Infrequent sanitization (>1-week intervals) increases risk of endotoxin release from established biofilms.

Pyrogen producing Bacteria Examples

- Escherichia Coli
- Pseudomonas
- Enterobacter

PURIFIED WATER	WATER FOR INJECTION
none	0.25 EU / mL
USP 29 Endotoxin Limits	

Nitrates / Heavy metals / Aluminum

While the European Monograph still includes test requirements for **Nitrates**, **Heavy Metals**, and **Aluminum** for dialysis use, there has never been a Nitrates requirement for USP waters and the requirement for **heavy metals** testing was removed in 1996. **Heavy metals** are toxic to humans but as source water must be compliant with EPA primary drinking water standards and common water system construction materials do not leach contaminants, **heavy metals** testing is recognized to be redundant to the **USP 29 <645> conductivity test**.

Where required to prove absence of contaminants, laboratory grab-samples are commonly tested using the inductively coupled plasma mass spectrometry (ICP-MS) method and reported as **part-per-million (PPM)** or **part-per-billion (PPB)**.

Conductivity (< 1.3 µS/cm² @ 25°C)

Conductivity is a qualitative measurement for the concentration of dissolved salts (minerals) in a water solution and is a measure of the ability of water to pass an electrical current. Because dissolved salts and other inorganic chemicals conduct an electrical current, **conductivity** increases as salinity increases. **Conductivity** is reported in micro-Siemens (µS/cm²) and is described in USP 29 <645>.

Conductivity is affected by temperature: the warmer the **water**, the higher the **conductivity**. Temperature compensation can be used to normalize values for process monitoring and diagnostics but all compendial product waters must be reported using uncompensated values.

Purified water readily absorbs atmospheric **carbon dioxide (CO_2)** which increases its **conductivity** and is not a contaminant of concern. Compendial specifications take this into account and allow for pH-neutral, demineralized waters given enough uptake time to saturate with dissolved **CO_2**.

Temp. (°C)	Cond. (µS/cm²)
0	0.6
5	0.8
10	0.9
15	1.0
20	1.1
25	1.3
30	1.4
35	1.5
40	1.7
45	1.8
50	1.9
55	2.1
60	2.2
65	2.4
70	2.5
75	2.7
80	2.7
85	2.7
90	2.7
95	2.9
100	3.1

USP <645> Stage 1: Temperature & Non-Compensated Conductivity

pH (5.0 – 7.0 Standard Units)

pH is measurement expressing the acidity or alkalinity of a solution. Theoretically pure water has a balanced **pH** of exactly 7.0 at 25 °C which indicates a neutral solution. The **pH** scale is logarithmic, and values less than neutral are more acidic and higher values are more basic. As purified water in inherently neutral, **pH** testing is recognized to be redundant to the USP 29 <645> conductivity test and was deleted in 1998.

Purified water readily absorbs atmospheric **carbon dioxide (CO_2)** which lowers its **pH** and is not a contaminant of concern. Compendial specifications take this into account and allow for pH-neutral, demineralized waters given enough uptake time to saturate with dissolved **CO_2**.

pH measurement of purified water is difficult due to the absence of ions that normally allow for electrical measurement across sensor electrodes. The USP recommends addition of potassium chloride (KCl) electrolyte to stabilize measurements and USP 29 <791> describes sensor and standardization requirements.

pH (S.U.)	Cond. (µS/cm²)
5.0	4.7
5.1	4.1
5.2	3.6
5.3	3.3
5.4	3.0
5.5	2.8
5.6	2.6
5.7	2.5
5.8	2.4
5.9	2.4
6.0	2.4
6.1	2.4
6.2	2.5
6.3	2.4
6.4	2.3
6.5	2.2
6.6	2.1
6.7	2.6
6.8	3.1
6.9	3.8
7.0	4.6

USP <645> Stage 3: KCl + Atmosphere / Temperature Equilibrated Conductivity Samples

TOC (<500 ppb)

Total Organic Carbon (TOC) is the amount of carbon found in an organic compound and is often used as a non-specific indicator of water quality or cleanliness of pharmaceutical manufacturing equipment. In general, **TOC** is considered a qualitative analysis of the amount of biomass and food available for the reproduction of bacteria in a water system and increasing numbers indicate filter integrity failure or non-sanitary process conditions.

TOC is reported as mg/L, ppm, or ppb and is determined using online analysis or by processing grab samples. Analysis methods measure Total Carbon, oxidize the sample, then report based on the difference of oxidizable/non-oxidizable carbon.