Introduction
High concentrations of planktonic bacterial cells (> $1 \times 10^4$ cells mL$^{-1}$) of broadly diverse populations are commonly found in both bottled mineral water and tap water. The conventional approach to characterize the general microbial quality of drinking water is the use of heterotrophic plate counts (HPC), which enumerate the bacterial cells which can grow up to form visible colonies on specific semi-solid media at selected incubation temperatures within a selected time period. On average, about 1% of the total bacteria in drinking water are culturable with standard HPC methods.

Importance
The value of accurate total cell counting is evident, e.g. when treatment plant operators aim to monitor the affectivity of specific processes such as membrane filtration or ozonation, or when regrowth during treatment (e.g. GAC filtration) is studied.

Approach
We have developed a protocol based on fluorescent staining of microbial nucleic acids with SYBR Green I, coupled with flow cytometric absolute cell counting, as a fast and accurate method to determine the total cell number in drinking water.

Result
The FCM method was shown to be fast, with 10 minutes required for the fluorescent staining step and 3 minutes for the analysis. The calibrated flow cytometer has an instrument error of 2%, which, combined with operator error gave an overall error of less than 5%. It is possible to detect cell concentrations as low as 200 cells/mL, but all evidence up to now suggests that this is well below the typical range of drinking water (c.a. $1 \times 10^5$ cells/mL).

More information
Full details on this deliverable can be found under D3.3.5. Further information can be requested from:
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Categorisation (i.e. classification, contains and constraints) of knowledge packages (KPs) can be carried out by ‘checking’ the appropriate boxes in the attached tables. For example, for a KP investigating point-of-use treatment suitable for a developing world country, the following might be checked:

**Classification**: Process chain – Tap (Customer) – Point-of-use (POU).

**Contains**: Report; Literature review.

**Constraints**: Low cost; Simple technology; No/low skill requirement; No/low energy requirement; No/low chemical requirement; No/low sludge production; Developing world location.

Note that only the lowest level classification needs to be checked, e.g. Point-of-use (POU) in the above example.

**Meta data** can be included under the ‘More Information’ section of the Executive Summary Report, i.e. Author(s), Organisation(s), Contact details (name and email), Quality controller (name and organisation) and Date prepared. (The TKI administrator can enter Source (= TECHNEAU), Date submitted (TKI) and Date revised (TKI)).
## TKI Categorisation

<table>
<thead>
<tr>
<th>Source</th>
<th>Process Chain</th>
<th>Process Chain (cont’d)</th>
<th>Water Quality</th>
<th>Water Quantity (cont’d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Catchment</td>
<td>Raw water storage</td>
<td>Sludge treatment</td>
<td>Legislation/regulation</td>
<td>- Leakage</td>
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<tr>
<td>Groundwater</td>
<td>- Supply reservoir</td>
<td>- Settlement</td>
<td>- Raw water (source)</td>
<td>- Recycle</td>
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<td>Surface water</td>
<td>- Pretreatment</td>
<td>- Dewatering</td>
<td>Chemical</td>
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<tr>
<td>Spring water</td>
<td>- Screening</td>
<td>- Disposal</td>
<td>- Organic compounds</td>
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<tr>
<td>Storm water</td>
<td>- Microstraining</td>
<td>Chemical dosing</td>
<td>- Inorganic compounds</td>
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<td>Primary treatment</td>
<td>- pH adjustment</td>
<td>- Disinfection by-products</td>
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<tr>
<td>Wastewater</td>
<td>- Sedimentation</td>
<td>- Coagulant</td>
<td>- Corrosion</td>
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<td>- Rapid filtration</td>
<td>- Polyelectrolyte</td>
<td>- Scaling</td>
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<td>- Slow sand filtration</td>
<td>- Disinfectant</td>
<td>- Chlorine decay</td>
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<tr>
<td>Bankside storage</td>
<td>- Bank filtration</td>
<td>- Lead/plumbosolvency</td>
<td>Microbiological</td>
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<tr>
<td>Water treatment</td>
<td>- Dune infiltration</td>
<td>Control/instrumentation</td>
<td>- Viruses</td>
<td>Consumers / Risk</td>
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<tr>
<td>- Pretreatment</td>
<td>Secondary treatment</td>
<td>- Flow</td>
<td>- Parasites</td>
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<td>Coagulation/flocculation</td>
<td>- Pressure</td>
<td>- Bacteria</td>
<td>Trust</td>
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<td>Sedimentation</td>
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<td>- Fungi</td>
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<tr>
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<td>Dissolved air flotation(DAF)</td>
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<td>- Hardness / alkalinity</td>
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<td>- Telemetry</td>
<td>- pH</td>
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<td>- Colour</td>
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<td>- Taste</td>
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- Internal plumbing - Service reservoir
- Internal storage Distribution
- Disinfection x
- Lead/plumbosolvency
- Manganese control
- Biofilm control x
Tap (Customer)
- Point-of-entry (POE) x
- Point-of-use (POU) x

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<td>Benchmarking</td>
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TKI Categorisation (continued)

- Water Quantity
- Risk Communication
- Source
- Potential pitfalls
- Source management
- Proven techniques
- Alternative source(s)
- Communication strategies
- Water balance
- Demand/supply trend(s)
- Demand reduction

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