



**International
Standard**

ISO 13647

**Water quality — Enumeration
of culturable microorganisms
— Colony count by spread plate
inoculation on R2A medium**

*Qualité de l'eau — Dénombrement des micro-organismes
revivifiants — Comptage des colonies parensemencement par
étalement sur un milieu gélosé R2A*

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Foreword

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Introduction

Treated drinking water contains a variety of microorganisms derived from various sources, such as raw water resource, different treatment steps within drinking water production and drinking water distribution networks.

Enumeration of the overall colony count of culturable microorganisms provides useful information for the assessment and surveillance of water quality. Colony counts are useful for assessing the raw water quality and efficiency of water treatment processes and provide an indication of the cleanliness and integrity of the drinking water distribution systems.

However, the main value of colony counts lies in the detection of changes from the expected values, based on representative monitoring. The change in colony counts can be caused for various reasons like biofilm formation, increase in water temperature, presence of microbially available nutrients or decrease in chlorine concentration. Any substantial increase in count can also be either a warning of potential treatment failure or contamination.

Nutrient rich tryptone yeast extract agar medium has been used for long time in water microbiology. ISO 6222 provides a monitoring method for heterotrophic plate counts in tap water quality verification. However, ISO 6222 only applies to the detection of microorganisms in nutrient rich conditions at 22 °C in 3 d or at 36 °C in 2 d.

Reasoner's 2 agar (R2A) medium, which has a low-nutrient composition and long incubation time (7 d at 22 °C), enables the determination of micro-organisms forming colonies after a longer incubation time. Furthermore, the low level of nutrients in the medium minimizes the colony size and overgrowth. Therefore, the R2A method is especially suitable for providing more information on the enumeration of heterotrophic colony counts in water with low colony counts.

The method is well suited for monitoring water for human consumption which is low in nutrients and is distributed in temperatures below 20 °C (see Reference [1]), for example, to measure the operational efficiency of the treatment process of public drinking water supplies. The low-nutrient agar specified in this document for enumeration of culturable microorganisms usually gives higher colony counts from water samples than typically used nutrient-rich formulations of culture media (see References [2], [3] and [4]).