

# A modified method to determine biomass concentration as COD in pure cultures and in activated sludge systems

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## Abstract

A simple technique to determine biomass concentration as chemical oxygen demand (COD) was developed as an alternative to the standard volatile suspended solid (VSS) method. The proposed technique for biomass measurement as COD is based on the determination of the biomass COD ( $COD_B$ ) as the difference between total COD ( $COD_T$ ) and the soluble COD ( $COD_S$ ) of the sample. The obtained results show that this technique was quicker and simpler than the traditional VSS method.

The validity of the proposed methods was tested with pure cultures of a filamentous micro-organisms (*Sphaerotilus natans*), a floc-forming bacteria and activated sludges. The method was also used for estimating the conversion factor ( $f_{CV}$ ) from VSS to COD units.

A modification of the standard VSS technique was also proposed using two membranes in the filtration device; this technique allowed the biomass determination in 1  $\mu\text{m}$  size bacteria cultures that cannot be detected by the standard VSS method because cells are not retained by the 1.5  $\mu\text{m}$  diameter pore glass-fibre filter.

## Notation

BOD	=	biochemical oxygen demand ( $\text{mg}\cdot\text{t}^{-1}$ )
COD	=	chemical oxygen demand ( $\text{mg}\cdot\text{t}^{-1}$ )
VSS	=	volatile suspended solid ( $\text{mg}\cdot\text{t}^{-1}$ )
$f_{CV}$	=	conversion factor = ratio between $COD_B$ and VSS
$COD_B$	=	biomass as COD ( $\text{mg}\cdot\text{t}^{-1}$ )
$COD_T$	=	total COD of the sample containing the biomass ( $\text{mg}\cdot\text{t}^{-1}$ )
$COD_S$	=	soluble COD ( $\text{mg}\cdot\text{t}^{-1}$ )
CV	=	coefficient of variation

## Introduction

The most commonly used collective parameters in wastewater characterisation are BOD and COD (Wanner, 1994). BOD test indicates consumption of oxygen in receiving water bodies for the biochemical oxidation of organic matter and ammonia remaining in the effluent. Although important, BOD is a test of very little practical use as results are obtained at least in 5 d. In the COD test, organic compounds are not oxidised with molecular oxygen as in the BOD test; a much more aggressive oxidising agent is used. The electrons from organic matter are transferred to dichromate; the reaction is performed in hot sulphuric acid solution and catalysed by silver cations. Only carbonaceous compounds are completely oxidised, so that the COD value does not include ammonia. A few types of organic materials, such as aromatic hydrocarbons and pyridines, are resistant to the oxidising conditions of the test. However, the COD test is relatively easy to perform and the results are obtained in a few hours through the use of commercially available kits.

There are several methods to determine biomass concentration based on different types of measurements, such as mass, volume or linear extent, metabolic rates, light scattering, cell or organelle

count (Pirt, 1975). However, the simplicity of the VSS technique has established this method is one of the key parameters used in modelling activated sludge systems (Metcalf and Eddy, 1979). Volatile suspended solids are determined by measuring the mass of oven-dry solids retained by a 1.5  $\mu\text{m}$  glass-fibre filter, volatilised at 550 °C (*Standard Methods*, 1992). In spite of its simplicity, VSS determination has, in some cases, several problems related to the filtration stage such as the presence of filamentous micro-organisms that obstruct the filters or micro-organisms with a size smaller than 1 mm that cannot be retained by the filter.

In most of the mathematical models describing activated sludge systems, equations are expressed in oxygen units (Henze et al., 1987; Kappeler and Gujer, 1992; Henze et al., 1995; Keesman et al., 1998). Since biomass is usually determined as VSS, a conversion factor from VSS to COD units is needed. This conversion factor ( $f_{CV}$ ) is the COD per unit mass of VSS and it depends on the biomass composition. Due to the diversity of micro-organisms present in activated sludges, the assumption of a given  $f_{CV}$  constant could potentially lead to errors in estimation of the COD fraction for the particulate fraction and therefore, in the evaluation of the kinetic and stoichiometric growth parameters characterising the biodegradation of organic compounds in the residual water.

The objectives of this work were:

- to develop and evaluate a simple technique to determine biomass concentration as COD as an alternative to the standard VSS method;
- to propose a modification of the standard VSS method to allow the biomass determination in smaller than 1  $\mu\text{m}$  size bacteria cultures, that cannot be detected by the standard VSS method;
- to determine the conversion factor  $f_{CV}$  from VSS to COD units using the developed techniques.

## Materials and methods

### Micro-organisms

The evaluation of the technique was performed on:

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