

TRACE METALS REQUIREMENTS

Heavy Metals Requirement

Metals which are considered to be most important for anaerobic digestion are Fe, Ni, Co and Mo. These heavy metals were found as components of the essential enzymes that drive numerous anaerobic reactions. These trace elements have a significant enhancement effect on granulation [Zeeuw and Lettinga, 1980, Riera *et al.*, 1985, Murry and Berg, 1981, Yoda *et al.*, 1991]. COD conversion and bacterial growth are reported to be limited at iron deficient concentration. Bivalent ions (Ca, Fe, Ba) are also found to be important in microbial aggregation [Shen *et al.*, 1993]. Iron seems to be involved in energy metabolism as a cytochrome and ferredoxin in methylotrophic methanogens. Iron is a growth-limiting factor for M-thermoautophicum and omission of iron prevents in increase in the final biomass. Minimum amount of iron needed to complete conversion of acetate is 0.1 mM, and addition of Fe at a concentration as low as 0.01 mM causes a significant increase in CH₄ production [Fatherpure, 1987]. The limiting concentrations of these metals are reported to be: Fe = 5 mg/L , Ni = 0.25 mg/L and Co = 0.10 mg/L [Callander *et al.*, 1987].

Nickel is an essential growth factor for many methanogens. Nickel is also a component of the carbon monoxide dehydrogenase in *Methanosarcina barkeri* [Speece *et al.*, 1983]. Nickel was shown to be essential in the growth medium of *Methanobacterium bryantii* to prevent rapid cell lysis. Addition of Mo enhances the rate of CH₄ production during the early growth phase. Addition of Ni and Mo to the medium may not be always necessary because low levels of these metals in the medium as contaminants may be sufficient for the growth of *Methanotherix sohengenii* [Fatherpure, 1987]. It was also found that bacteria in granules are efficient in collecting trace elements, which are in very low concentration, even from the impurity of feed chemicals.

It was found that extracellular polymers prefer to bind heavy metals when they are available due to more stable complexes and addition of yeast extract enhances the ability of bacteria to collect trace elements [Shen *et al.*, 1993]. The cobalt and iron concentration in Extracellular Polymeric Substances (EPS) may suggest that cobalt and iron bind to EPS competitively [Shen *et al.*, 1993]. Inhibition of CH₄ production at elevated concentrations of trace elements may be due to nonspecific binding of trace elements with the carrier proteins that are involved in their uptake and incorporation. An excess of a particular element may saturate the carrier molecules and thereby restrict the uptake of other essential metal ions. A metal ion in excess may also replace the essential metal of an enzyme. This would result in decrease in methanogenesis at high levels of these metals [Fatherpure, 1987].

Trace elements such as Fe, Ni, Co, and Mo can considerably affect the duration of start-up because these elements are essential for methanogenic growth. The approximate trace metals demand for low and high strength wastewaters are given below [Weiland and Rozzi, 1991]:

Trace metal requirements

Trace Element	Concentration range	
	10 g COD/L	50 g COD/L
Fe	0.5-20.0	3.0-100.0
Ni	0.05-3.0	0.3-15.0
Co	0.05-2.0	0.3-10.0
Mo	0.01-0.05	0.05-0.2