

The influence of nitrogen on the development of biofilms

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A lot of information has been published about effects of nutrients on the development of biofilms – where as the influence of nitrogen has been neglected. This investigation demonstrates effects of eight different nitrogen concentrations (C:N:P; carbon:nitrogen:phosphorus) from a synthetic medium on the development of biofilms. The inoculation was performed using a mixed population from the activated sludge tank of a wastewater treatment plant. A biofilm-reactor arrangement was used to create the biofilms upon polyethylene immobilization units (IU). Multiple physical, chemical and biological parameters were monitored to characterize changes in bacterial biofilms.

Effects of available nitrogen on the composition of produced biofilms and their EPS (extracellular polymeric substances):
 - water, nitrogen (data not shown), carbohydrate, protein, CFU (colonized forming units) and calcium contents in biofilms and according to EPS (data not all shown) are depended on the available nitrogen
 - oxygen (35,1%±1,11), hydrogen (7%±0,19), carbon (46,9%±0,7) and phosphate (1,05%±0,14) were not influenced by the available nitrogen (comparable results, out of a natural biofilm, are described by Flemming, 1995)
 - synthetic medium is qualified for the application in corrosion investigations of metallic surfaces.

Biofilm reactor system

- 20 L synthetic medium
- pH-value 7
- temperature 30 °C
- oxygen: 9,2-10 mg/L (reactor 1)
- DOC (dissolved organic carbon) ~ 1020 mg/L
- flow-rate 4,8 L/h
- 700 IU (reactor 2)-surface 0,8 m²

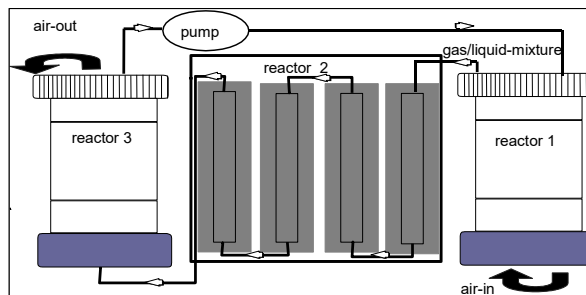


Figure 1: Biofilm-reactor-system (Scheen, 1998)

Synthetic medium

- C₁₂H₂₂O₁₁ = 4,37 mmol
- NH₄Cl = 0,71-3,53-5,64-7,05-8,46-9,87-12,68-14,1 mmol
- C₂H₆O = 1,05 ml/L
- K₂HPO₄ · 3 H₂O = 0,43 mmol
- FeSO₄ · 7 H₂O = 0,036 mmol
- MgSO₄ · 7 H₂O = 0,051 mmol
- CaCl₂ · 2 H₂O = 0,099 mmol

Results: Biofilms

Diagram 1: Water content in biofilms - 105 °C, over night – till constant of weight

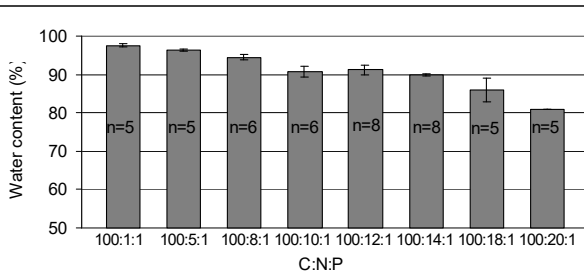


Diagram 2: CFU in biofilms - Agar, Merck 7881

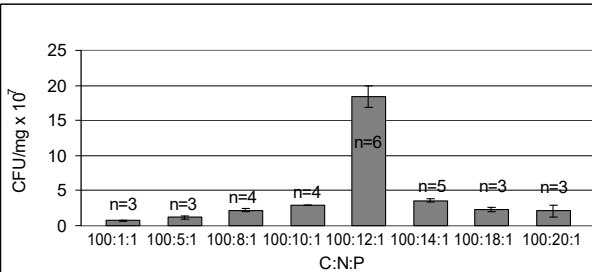


Diagram 3: Protein and Carbohydrate in biofilm-dry. Lowry (1951) Dubois(1956) Sum of protein and carbohydrate const. 70,8%±3,59

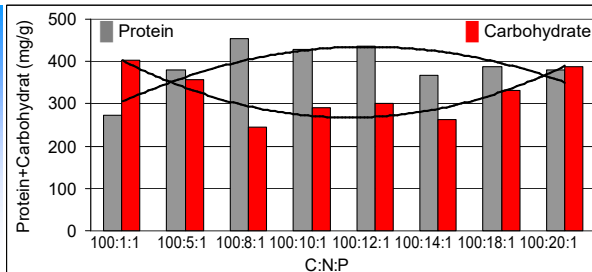
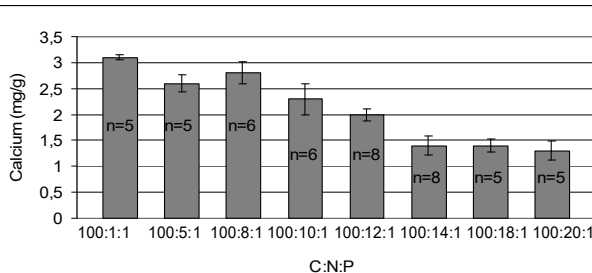


Diagram 4: Contents of calcium in biofilm-dry - ICP (inductively coupled plasma)



Results: EPS (extracted by cation-exchange-resin, CER - modified, Frølund et al., 1996)

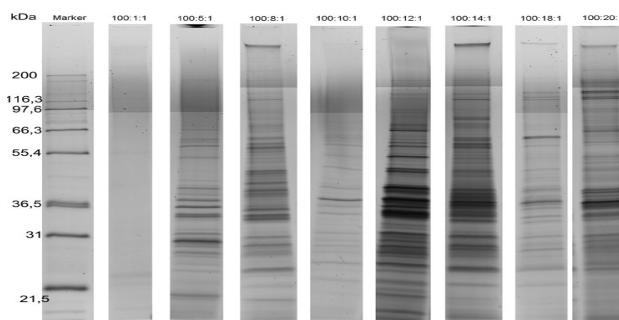


Figure 2: Protein in extracted EPS - SDS-PAGE (Sodium-dodecyl-sulphate-polyacrylamid-gel-electrophoresis)

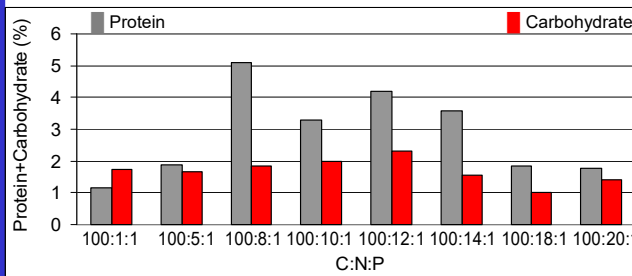
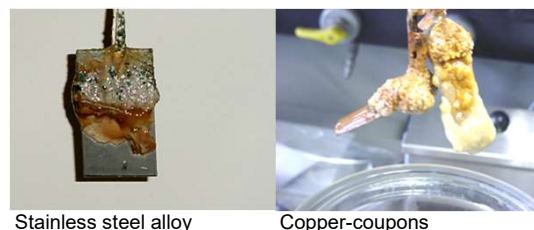


Diagram 5: Protein and Carbohydrate in EPS - calculated to biofilm-dry

Practical applications, examples:



Stainless steel alloy

Copper-coupons

Figure 3: Metal-coupons covered with biofilm

- synthetic medium
- batch – submersed
- duration time: 2000 h
- mixed-population

References

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