1

2

Sulfate reducing ammonium oxidation (SULFAMMOX) process under anaerobic conditions

3 Grubba Dominika[a], Majtacz Joanna[a], Mąkinia Jacek[a1]

4 [a]Gdansk University of Technology, Department of Sanitary Engineering, Faculty of Civil and Envi-
5 ronmental Engineering, Narutowicza Street 11/12, 80-233 Gdansk, Poland; (domini-
6 ka.grubba@pg.edu.pl; joamajta@pg.edu.pl; jmakinia@pg.edu.pl)
7
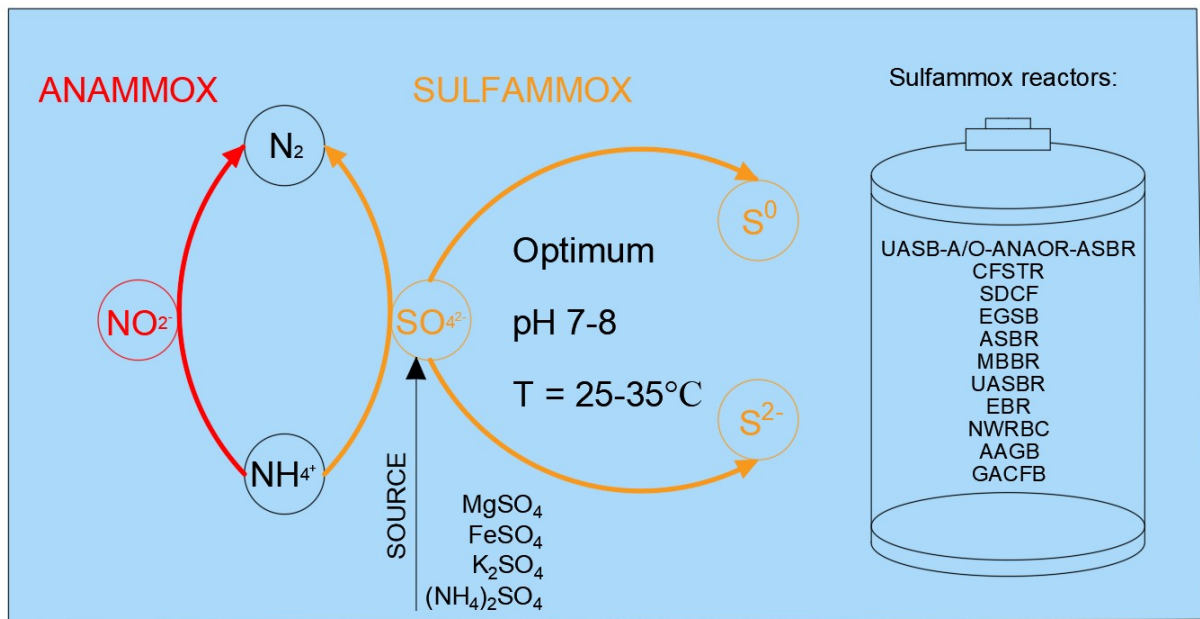8 [1]Correspondence: jmakinia@pg.edu.pl

9

10 **Highlights**

11 • Sulfate may be an additional electron acceptor in the anaerobic ammonium oxidation.

12 • *Bacillus Benzoevorans* and *Brocadia Anammoxoglobus Sulfate* can perform sulfammox.

13 • The optimal conditions for the sulfammox is 25°C -35°C and pH - 7.0 – 8.

14 • Sulfammox is a viable option for specific industrial wastewater with high NH_4^+ and SO_4^{2-}.

15 • Sulfammox has been studied in suspended growth, biofilm, granular and hybrid reactors.

16 **Abstract**

17 Sulfate (SO_4^{2-}) can be an electron acceptor for ammonium nitrogen (NH_4^+) oxidation under anaerobic

18 conditions. The process is known as sulfammox and can be a viable alternative to conventional, nitrite

19 (NO_2^-) dependent, anammox. Two bacterial species, including *Bacillus Benzoevorans* and *Brocadia*

20 *Anammoxoglobus Sulfate*, can perform that process. With sulfammox, an economically inefficient pre-

21 nitration step (due to aeration) is not required. There are more than 10 different systems in which

22 sulfammox has been studied, including suspended growth, biofilm, granular and hybrid reactors. A

23 combination of anammox and sulfur related processes (sulfammox and autotrophic denitrification)

24 would especially be appropriate for specific industrial wastewater with high content of nitrogen

25 compounds and SO_4^{2-}. The results of recent studies suggest that very high removal efficiencies could

26 simultaneously be achieved with respect to both NH_4^+ (92-99%) and SO_4^{2-} (53-60%).

27   **Graphical abstract**

ANAMMOX SULFAMMOX

Sulfammox reactors:
UASB-A/O-ANAOR-ASBR
CFSTR
SDCF
EGSB
ASBR
MBBR
UASBR
EBR
NWRBC
AAGB
GACFB

N_2

NO_2^-

SO_4^{2-}

NH_4^+

Optimum
pH 7-8
T = 25-35°C

S^0

S^{2-}

SOURCE

$MgSO_4$
$FeSO_4$
K_2SO_4
$(NH_4)_2SO_4$

28

29   **Keywords**: sulfammox, anammox, autotrophic denitrification, sulfate, sulfur cycle

32
33
34

1. Introduction

Nitrification and denitrification are the most common processes responsible for nitrogen (N) conversions in wastewater treatment systems. A viable alternative to that pathway of nitrogen removal is the "anaerobic" ammonium oxidation (anammox) process. "Anaerobic" because it is actually an anoxic process due to the presence of nitrite (NO_2^-). It is generally accepted that anaerobic ammonia oxidizing bacteria (AAOB) oxidize ammonia (NH_4^+) to N_2 with NO_2^- as an electron acceptor. In fact, however, AAOB have a more comprehensive metabolism than initially assumed and other phenomena of "anaerobic" NH_4^+ oxidation have been discovered (Kartal et al. 2012; Liu et al. 2008). In addition to NO_2^-, there may be other electron acceptors, including sulfate (SO_4^{2-}), for NH_4^+ oxidation under "anaerobic" conditions (Zandt et al. 2018). This process is known as sulfate reducing ammonium oxidation (SRAO) or sulfammox (Bi et al. 2020).

In addition to anaerobic sludge digester liquors, the sulfammox process may especially be appropriate for treatment of some industrial wastewater, containing high concentrations (>1000 mg/l) of both NH_4^+ and SO_4^{2-}. Such characteristics are typical for the effluents from seafood, chemical, textile, paper, fermentation and sugar production (Rikmann et al. 2016).

In comparison with the conventional anammox, sulfammox is easier to control as nitritation becomes unnecessary (SO_4^{2-} instead of NO_2^- serves as the electron acceptor) (Zhang et al. 2009). Besides, as a reducing process of SO_4^{2-}, it is also free of secondary pollution caused by sulphide (S^{2-}), which is toxic and harmful to human health and aquatic ecosystems (Zhang et al. 2019a). Sulfammox can also prevent interference with the conventional anammox process caused by inhibition of S^{2-} (Xu et al. 2020) or hydrogen sulfide (H_2S) (Wiśniewski et al. 2019). Moreover, elemental sulfur (S^0) is formed and its recovery provides a valuable by-product (Rios-Del Toro and Cervantes 2019). The recovered S^0 could be used as electron donor for autotrophic denitrification as reported by Ucar et al. (2020). Moreover, recovering S^0 from wastewater is also essential to reuse it as fertilizer or to re-enter production lines in other industries. The combination of the technology based on the anammox process with the technology based on the sulfammox process would enable a balanced approach to the

61   problem of specific industrial wastewater with high content of nitrogen compounds and SO_4^{2-} through

62   their co-treatment in combined processes.

63   The sulfammox process may occur either independently or in conjunction with the conventional

64   annamox process. The combination of both processes can increase the overall nitrogen removal

65   efficiency. Recent studies (Zhang et al. 2019a; Wu et al. 2020) have shown a high degree of

66   simultaneous removal of NH_4^+ and SO_4^{2-}, i.e. in the range 92-99% and 53-60%, respectively, with

67   NO_2^- and SO_4^{2-} as electron acceptors.

68   The sulfammox process has briefly been addressed in reviews on anammox in marine environments

69   (Rios-Del Toro and Cervantes 2019) and in the state of anammox research in China (Ali et al. 2013).

70   There are still a few publications on this process and finding them is not straightforward, as

71   sulfammox also appears as SRAO or sulfate-dependent anammox. Based on the Web of

72   Science database, using the keywords "sulfate", "anammox" and "wastewater", a number of

73   publications and their citations appearing in 1999-2020 years are presented in Fig. 1.
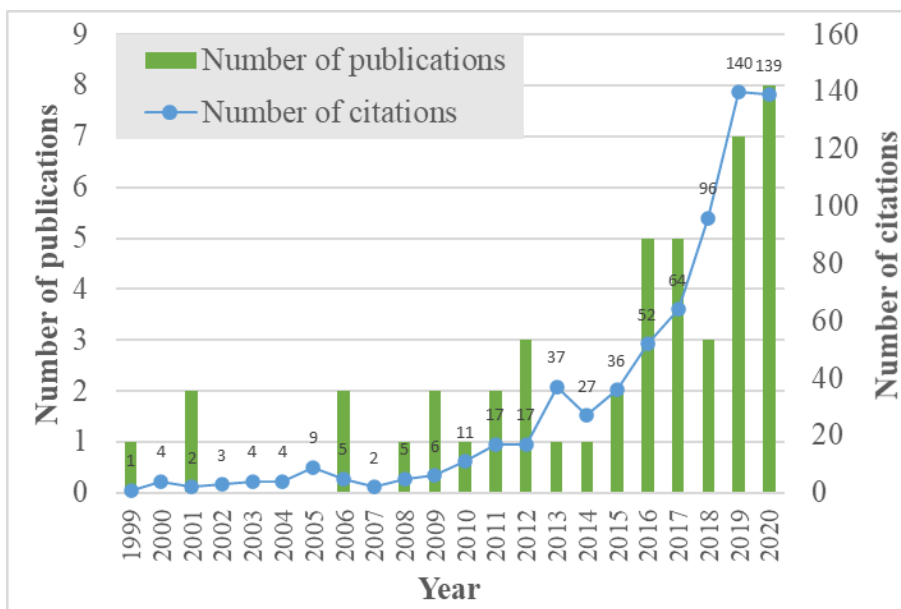
74

75   **Fig 1** Number of publications based on keywords "sulfate", "anammox" and "wastewater" and their citations in

76   the Web of Science database in 1999-2020

4

77   Based on the data in Fig. 1, it can be projected that the number of publications on sulfammox and their

78   citations will be increasing fast over the next few years. Very recently, Liu et al. (2021) have

79   published the first review paper especially dedicated to sulfammox. However, the paper does not

80   incorporate a few studies, Zhang et al. (2019a) or Wang et al. (2017a), that have a significant effect on

81   the process understanding.. In particular, the latter paper describes the NH_4^+/SO_4^{2-} ratio which plays a

82   key role in the sulfammox process. Other issues, omitted or not sufficiently addressed in the study of

83   Liu et a. (2021), comprised the spontaneity of the process, effect of COD on sulfammox, and feeding

84   options (NO_2^- and SO_4^{2-} together and separately).

85   In the present study, the combination of several processes influencing removal of NH_4^+ and SO_4^{2-} from

86   wastewater was addressed, including sulfammox, anammox, sulfide-dependent autotrophic

87   denitrification, sulfur-dependent autrotrophic denitrification, nitrification, denitrification, and

88   heterotrophic sulfate reduction. A special attention was given to linking the sulfammox process with

89   sulfur-dependent autotrophic denitrification. Moreover, a wide variety of sulfammox reactors was

90   presented and discussed in terms of the operating conditions and performance efficiency.

91   Both soil, air and water are exposed to the influence of toxic sulfur compounds - H_2S and S^{2-}.

92   Recognition of the sulfammox process may lead to the development of research on this process, and

93   hence to environmental protection, thanks to the decomposition of these compounds into S^0 and

94   reduction of energy consumption by limiting two separate processes of removing NH_4^+ and SO_4^{2-} to

95   one co-treatment. Therefore, the aim of this mini review is to characterize the sulfammox process,

96   indicate the operational conditions in which it can be carried out, and compare the examined

97   sulfammox reactors.

98   **2.   The characteristics of the sulfammox process**

99   Sulfammox was first reported by Fdz-Polanco et al. (2001b) in a granular activated carbon anaerobic

100   fluidized bed reactor treating vinasse from an ethanol distillery of sugar beet molasses. The authors

101   observed that approximately 80% of SO_4^{2-} was converted to S^0 with simultaneous oxidation of NH_4^+ to

102   N_2. The combined process for removal of NH_4^+ and SO_4^{2-} was described as follows:

103     $SO_4^{2-} + 2NH_4^+ \rightarrow S^0 + N_2 + 4H_2O$ (1)

104 In the follow-up studies, Liu et al. (2008) and Yang et al. (2009) identified SO_4^{2-} as a potential electron

105 acceptor as it was the feed component. They investigated the process of simultaneous removal of

106 $NH4^+$ and SO_4^{2-} under anaerobic conditions. The ratio of NH_4^+ to NO_2^- consumption was

107 approximately 1.1 : 1 and 1 : 1.15, respectively, in a non-woven rotating biological contactor (NRBC)

108 and upflow anaerobic sludge blanket reactor (UASBR) (see: Table 2). These values were significantly

109 higher in comparison with 1 : 1.32, which is the theoretical ratio for the conventional, NO_2^--dependent,

110 anammox process (Xie et al. 2017).

111 Based on the literature (Strous et al. 2006; Zhang et al. 2009; Schrum et al. 2009), formation of HS^- in

112 the sulfammox process may also be considered:

113     $8NH_4^+ + 3SO_4^{2-} \rightarrow 4N_2 + 3HS^- + 12H_2O + 5H^+$ (2)

114 Alternatively, formation of HS^- may be associated with oxidation of NH_4^+ to NO_3^- (Schrum et al.

115 2009):

116 $NH_4^+ + SO_4^{2-} \rightarrow NO_3^- + HS^- + H_2O + H^+$ (3)

117 The consumption rate of SO_4^{2-} can be estimated based on the corresponding consumption rate of N-

118 NH_4^+ and the theoretical stoichiometric consumption ratio (= 2) of NH_4^+ to SO_4^{2-} in sulfammox (see:

119 reaction 1). An inadequate influent ratio of NH_4^+/SO_4^{2-} - different than 2 (see: reaction 2,3), may result

120 in the formation of HS^- in the sulfammox process according to reactions (2,3). The NH_4^+/SO_4^{2-} ratio of

121 2 was indeed found in the studies of Zhang et al. (2009), Yang et al. (2009) and Cai et al. (2010). In

122 other studies, the reported ratios were lower, i.e. 1.71 - 1.75 (Liu et al. 2008) and 1.65 (Bi et al. 2020).

123 Rikmann et al. (2012) noted that the stoichiometric ratio of NH_4^+ moles consumed per mole of reduced

124 SO_4^{2-} was higher than could be expected from the amount of SO_4^{2-} reduced. This implicitly indicated

125 the presence of additional electron acceptors, other than SO_4^{2-} (like humic matter) coupled with NH_4^+

126 oxidation or reoxidation of reduced sulfur compounds into SO_4^{2-}. The high efficiency of NH_4^+ removal

127 may result from complex interactions between organic compounds, nitrogen and sulfur like

6

128 sulfammox, anammox, autotrophic denitrification, heterotrophic denitrification (Rikmann et al. 2012,

129 2014, 2016).

130 The newly discovered, SO_4^{2-} dependent, AAOB species have been found to be responsible for carrying

131 out the above reactions (2-4). The first one was *Brocadia Anammoxoglobus Sulfate* (Liu et al. 2008),

132 which was a functional microorganism in the simultaneous removal of NH_4^+ and SO_4^{2-} and ended the

133 conversion of NH_4^+ and SO_4^{2-} by producing NO_2^- as an intermediate. The second isolated species,

134 *Bacillus Benzoevorans* (Cai et al. 2010), was responsible for carrying out the entire sulfammox

135 reaction. In the study of Liu et al. (2015b), the dominant bacteria changed from *Candidatus* Brocadia

136 to *Bacillus Benzoevorans* when the process transformed from the conventional anammox to

137 sulfammox. Sulfammox bacteria and AAOB combine the N and S cycles, increasing the range of N-S

138 transformations as shown in Fig. 2.
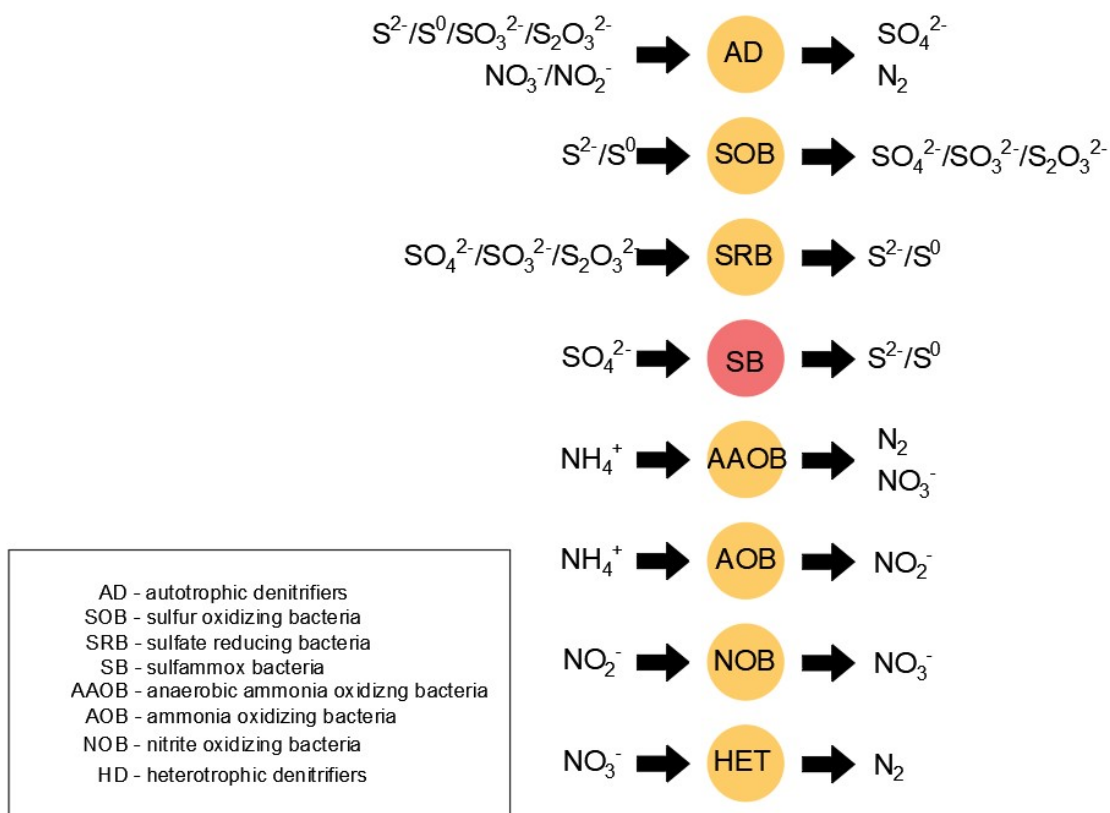
139
140 **Fig 2** Bacteria responsible for the specific N and S transformations

141

142 The SO_4^{2-} dependent AAOB are rodshaped with flagellum and spore, having a size of (0.7-1.0) × (2.4-

143 3.5) μm. The colony on the plate was round with a diameter of about 1 mm with a light yellow color,

144 and its surface was smooth and wet. The cultivated biomass was dominated by chains of bacilli and

145 cocci. Cocci generally had a diameter of 0.9 μm, whereas bacilli varied around 0.8 μm and 1-1.2 μm in

146 width and length, respectively (Zhang et al. 2009; Cai et al. 2010; Ali et al. 2013).

147 Some Proteobacteria, which may potentially perform sulfammox, include the following species: *Sulfu-*

148 *rimonas, Desulfuromonadales, Desulfovibrio, Desulfuromonas, Desulfobulbus, norank Rhodobacter-*

149 *aceae* and *Thiobacillus* (Rios-Del Toro et al. 2017; Wang et al. 2017a; Rios-Del Toro et al. 2018).

150 A syntrophic relationship between ammonia oxidizing bacteria (AOB), sulfate reducing bacteria

151 (SRB) and AAOB could make thermodynamically not favorable oxidation of NH_4^+ to NO_2^- coupled

152 with a possible reduction of SO_4^{2-} to S^0 (Rikmann et al. 2014). A pure chemical reaction between NH_4^+

153 and SO_4^{2-} without microorganisms is not possible (Yang et al. 2009).

154 **3. Relationship between sulfammox and sulfide-dependent autotrophic denitrification**

155 The overall sulfammox reaction (reaction (1)) has been shown to occur in three consecutive

156 biochemical reactions (reactions (4-6)) (Fdz-Polanco et al. 2001b; Zhang 2019a; Bi et al. 2020):

157 $3SO_4^{2-} + 4NH_4^+ \rightarrow 4NO_2^- + 3S^{2-} + 4H_2O + 8H^+$ (4)

158 $3S^{2-} + 2NO_2^- + 8H^+ \rightarrow N_2 + 3S^0 + 4H_2O$ (5)

159 $2NO_2^- + 2NH_4^+ \rightarrow 2N_2 + 4H_2O$ (6)

160 In reaction (4), NH_4^+ reacts with SO_4^{2-} and is oxidized to NO_2^- (intermediate) inside the bacterial cell

161 and SO_4^{2-} is simultaneously deoxygenated to S^{2-}. The NO_2^- produced diffuses outside of the bacterial

162 cell. In reaction (5), part of NO_2^- is reduced with S^{2-}, which leads to production of N_2 and S^0. Finally,

163 reaction (6) is the conventional anammox process carried out by *Planctomycetes* (Van der Star et al.

164 2007). Yang et al. (2009) described reaction (5) as the denitrification process that occur through

165 reduction of NO_2^- to N_2 with simultaneous oxidation of S^{2-} by autotrophic denitrifiers, where the

166 electron donor is S^{2-} and the electron acceptor is NO_2^-. For better understanding, reactions 4-6 are

167 shown in Figure 3a.
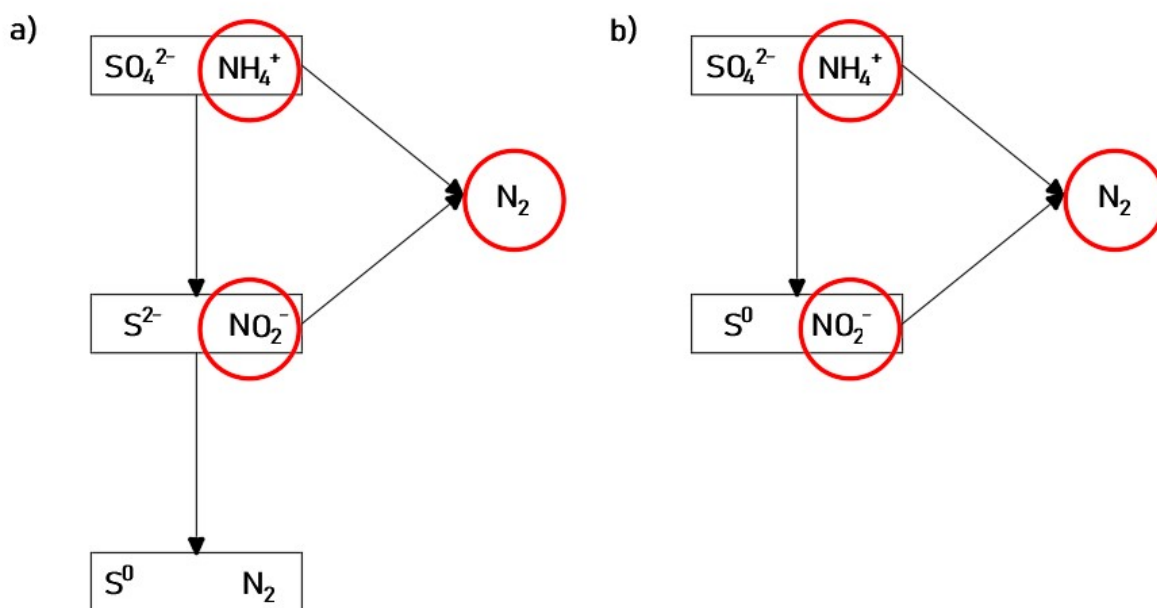
168
169 **Fig 3** Reactions involved in the sulfammox process as proposed by Yang et al. (2009) (a), and Liu et al. (2008)

170 (b)

171 **It should be emphasized that sulfide-dependent autotrophic denitrification is one of the reactions**

172 **involved in the overall sulfammox process, according to Fig 3a. Therefore, it is difficult to**

173 **distinguish a strict boundary between sulfammox and sulfide-dependent autotrophic**

174 **denitrification. In fact, the denitrification reaction is one of the components of sulfammox and**

175 **without it sulfammox cannot occur, as shown in reactions (4-6).** Therefore, some researchers do

176 not distinguish the efficiency of NH_4^+ removal in the sulfammox process at all, but only report the total

177 efficiency of NH_4^+ removal under anaerobic conditions in the presence of SO_4^{2-} (Wu et al 2020; Bi et

178 al. 2020; Zhang et al 2019a).

179 On the contrary, Liu et al. (2008) explained the sulfammox process as a combination of two reactions

180 as shown earlier in Fig. 3b. According to that concept, NH_4^+ would be partially converted to NO_2^- and

181 coupled with a conversion of SO_4^{2-} (electron acceptor) to S_0. Then NH_4^+ would be oxidized to N_2 by

182 NO_2^- in the conventional anammox process. Currently, the exact pathway of sulfammox remains

183 largely unknown. More detailed microbiological tests are needed to check which microorganisms and

184 genes are involved in that process.

9

185   It should be emphasized that S^{2-} in the sulfammox process (see: reaction 4) can be oxidized to either S^0

186   or SO_4^{2-}, depending on the initial S^{2-} to NO_2^- ratio. Therefore, that ratio must be strictly controlled to

187   avoid re-oxidation to SO_4^{2-}. For sulfammox, it is important to reduce SO_4^{2-} to S^0. When S^{2-} is oxidized

188   back to SO_4^{2-}, the total reduction of SO_4^{2-} in the sulfammox process decreases.

189   When the stoichiometric ratio of NH_4^+ moles consumed per mole of reduced SO_4^{2-} is higher than

190   might be expected from the degree of SO_4^{2-} reduction, this may also be due to the partial reoxidation

191   of S^0 or HS^- to SO_4^{2-} by sulfur-related autotrophic denitrification (Rikmann et al. 2012, 2014; Qin et al.

192   2019; Wang et al. 2020). Some chemolithotrophic denitrifiers, such as *Thiobacillus denitriticans*, are

193   capable of performing sulfur-related autotrophic denitrification.

194   The autotrophic denitrification reactions can occur with S^{2-}, sulphite (SO_3^{2-}), thiosulphate ($S_2O_3^{2-}$) or

195   S^0 as electron donors, and either NO_3^- or NO_2^- as electron acceptors (Guo et al. 2013; Xu et al. 2013;

196   Yu et al. 2013; Wang et al. 2017b; Di Capua et al. 2019). Then, either SO_4^{2-} or S^0 is formed depending

197   on the sulfur to nitrogen ratio (Kalyuzhnyi et al. 2006; Liu et al. 2015a). The following reactions

198   describe those complex phenomena (Li i wsp. 2009; Wang et al. 2017b):

199    $5S^{2-} + 2NO_3^- + 12H^+ \rightarrow 5S + N_2 + 6H_2O$ (7)

200    $5S^0 + 6NO_3^- + 2H_2O \rightarrow 5SO_4^{2-} + 3N_2 + 4H^+$ (8)

201    $5S^{2-} + 8NO_3^- + 8H^+ \rightarrow 5SO_4^{2-} + 4N_2 + 4H_2O$ (9)

202    $3S^{2-} + 2NO_2^- + 8H^+ \rightarrow 3S^0 + N_2 + 4H_2O$ (10)

203    $3S^0 + 6NO_2^- \rightarrow 3SO_4^{2-} + 3N_2$ (11)

204    $3S^{2-} + 8NO_2^- + 8H^+ \rightarrow 3SO_4^{2-} + 4N_2 + 4H_2O$ (12)

205   A simplified relationship of the sulfammox process with the nitrogen and sulfur cycles is presented in

206   Fig. 4.
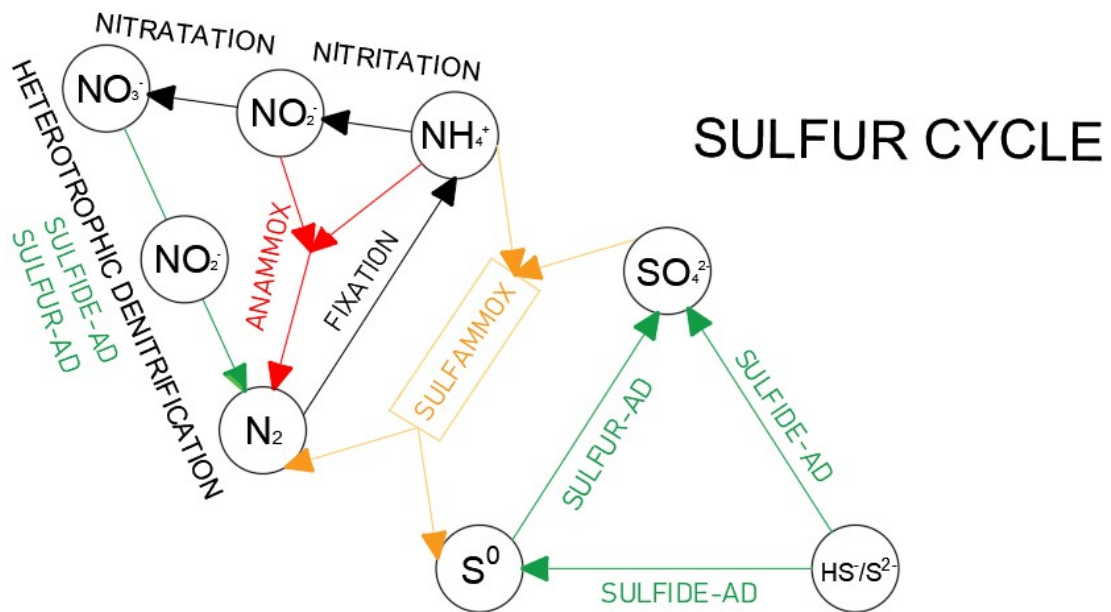
207
208 **Fig 4** Coupling the nitrogen and sulfur cycles in sulfammox, sulfur-dependent autotrophic denitrification (sulfur-

209 AD) and sulfide-dependent autotrophic denitrification (sulfide-AD)

4. Environmental factors and operational conditions affecting sulfammox

4.1. Process medium and feeding options

Most of the sulfammox studies have been carried out with synthetic wastewater or growth media (Zhao et al. 2006; Liu et al. 2008; Yang et al. 2009; Cai et al. 2010; Bi et al. 2020), but there have also been a few studies using real wastewater (Rikmann et al. 2012, 2014, 2016). Different compounds have been used as the SO_4^{2-} source in the medium (see: Table 1). The inoculum biomass originated from various sources (see: Table 2), including long-term operated anammox reactors and anaerobic digesters. The cultivation experiments have been carried out with three feeding options:

a) conventional anammox was run at the beginning, and then NO_2^- was replaced with SO_4^{2-} as a new electron acceptor (Yang et al. 2009; Rikmann et al. 2012, 2016; Zhang et al. 2019a, b; Bi et al. 2020);

b) SO_4^{2-} was used since the beginning without any addition of NO_2^- (Zhang et al. 2009; Wang et al. 2017a; Zhang et al. 2019b; Bi et al. 2020);

223   c) SO_4^{2-} and NO_2^- were simultaneously used as electron acceptors during the whole study period (Zhao

224   et al. 2006; Liu et al. 2008; Zhang et al. 2019a; Wu et al. 2020).

225   4.2. Temperature

226   The process temperature set point normally ranged from 30°C to 36°C as shown in Table 1. Cai et al.

227   (2010) tested the sulfammox process efficiencies at the following series of temperatures: 15°C, 25°C,

228   30°C, 35°C, 45°C and 55°C. The NH_4^+ and overall SO_4^{2-} removal efficiencies were approximately

229   37.5% and 35%, 36% and 30%, respectively, at T = 15°C and T = 55°C. The highest NH_4^+ and overall

230   SO_4^{2-} removal efficiencies were observed at T = 30°C, i.e. 44.4% and 40%, respectively. The removal

231   rate of NH_4^+ and SO_4^{2-} at that temperature was 0.168 mg N/l/h (R^2 = 0.98) and 0.191 mg S/l/h (R^2 =

232   0.95), respectively. The optimal temperature range for the sulfammox process is 25°C -35°C (Cai et

233   al., 2010).

234   The sulfammox process was also studied at lower temperatures, e.g. 20°C (Rikmann et al. 2016) and

235   14-15°C (Wu et al. 2020). In the latter case, despite such a low temperature, the NH_4^+ and overall

236   SO_4^{2-} removal efficiencies remained at a high level, i.e. 98.5% and 52.8%, respectively (Wu et al.

237   2020). Due to the combination of anaerobic-aerobic, continuous and batch processes adopted in this

238   process, the anammox and sulfammox coupled to remove nitrogen. Rikmann et al. (2016) studied two

239   reactors at different temperatures, i.e. MBBR (20°C) and UASBR (36°C). That approach was not

240   clearly explained, but apparently resulted from the use of different sludges in both reactors. In the

241   UASBR, the inoculum originated from an anaerobic reactor for treatment of industrial wastewater

242   (yeast production), whereas the MBBR was inoculated with carriers with a well-deposited anammox

243   biofilm developed in a conventional laboratory-scale anammox reactor. The TN removal efficiencies

244   were in the range 5 - 72% for the MBBR and 10 - 75% for the UASBR, respectively. Despite the use

245   of different temperatures, the TN removal rates were similar, i.e. 0.05 kg N/m³/d for the MBBR and

246   0.04 kg N/m³/d for the UASBR.

247   4.3. pH

248   The optimal pH value is 7.0 - 8.5 for both conventional anammox and sulfammox (Wu et al. 2020),

249   therefore, many studies on sulfammox have been carried out in that pH range (Yang et al. 2009; Zhang

250   et al. 2009; Zhang et al. 2019a,b; Bi et al. 2020; Wu et al. 2020). Cai et al. (2010) studied the effect of

251   pH on the efficiency of NH_4^+ and overall SO_4^{2-} removal. The following pH values were considered:

252   6.5, 7.5, 8.5, 9.5 and 10.5, and the optimum pH was found at 8.5. On the contrary, Zhao et al. (2006)

253   found the optimum pH = 7.8.

254   **Tab. 1** Environmental factors and operational conditions in the sulfammox studies

Source of SO_4^{2-}	COD addition	pH [-]	Temperature [°C]	Reference
$MgSO_4$ / $FeSO_4$	No	7.8	35	Bi et al. [2020]
$(NH_4)_2SO_4$	No	8.1-8.3	35	Zhang et al. [2019a]
$(NH_4)_2SO_4$	No	8.1-8.6	30	Zhang et al. [2019b]
K_2SO_4	No	8.5	30	Cai et al. [2010]
Na_2SO_4	No	7.5-8.5	35	Yang et al. [2009]
Na_2SO_4	No	7.5	30	Zhang et al. [2009]
$(NH_4)_2SO_4$	No	8-8.2	35	Liu et al. [2008]
n.a.	Yes	7-8.5	14-15	Wu et al. [2020]
n.a.	Yes	6.9-8.1	36	Wang et al. [2017a]
K_2SO_4	Yes	8.4	20	Rikmann et al. [2016]
K_2SO_4	Yes	8.11	36	Rikmann et al. [2014]
n.a.	Yes	7.8-8.3	36	Fdz-Polanco et al. [2001b]

255   n.a.: not available

256   4.4. COD addition

257   Even though COD is not required for the sulfammox process (Zhang et al. 2009), the experiments

258   were performed either without COD addition (Liu et al. 2008; Cai et al. 2010; Prachakittikul et al.

259   2016; Zhang et al. 2019a,b; Bi et al. 2020) or with COD addition (Fdz-Polanco et al. 2001a,b;

260   Rikmann et al. 2012, 2014, 2016; Wang et al. 2017a; Wu et al., 2020). When COD is present in

261   wastewater, the sulfammox process can be coupled with subsequent heterotrophic denitrification

262 (Zhang et al. 2019b). In the studies of Zhang et al. (2019b) sulfammox was mainly due to the high

263 proportion of Proteobacteria, but approximately 12.4% of denitrifiers were also found in the sediment.

264 This indicates that nitrification, denitrification and the traditional anammox with sulfammox may

265 simultaneously occur in oxidation of NH_4^+. This allows for simultaneous removal of NH_4^+, SO_4^{2-} and

266 COD from wastewater (Wang et al. 2017a). Kosugi et al. (2019) proposed a combined SO_4^{2-} reduction,

267 denitrification/anammox and partial nitrification process in an anaerobic-anoxic reactor. The authors

268 confirmed the coexistence of heterotrophic denitrifying bacteria, sulfur denitrifying bacteria and

269 anammox Candidatus Brocadia bacteria. They also recognized that heterotrophic and autotrophic

270 denitrifying bacteria, competing for NO_3^- and NO_2^-, can be used to oxidize S^{2-} to S^0 prior to oxidation

271 of organic carbon.

272 Yin et al. (2017) showed that sulfur-based autotrophic denitrification occurred with heterotrophic

273 denitrification in an anaerobic baffled reactor. The authors also indicated that without addition of S^{2-} a

274 significant amount of NO_3^- was reduced heterotrophically to N_2 (76.6%). However, the addition of S^{2-}

275 stimulated autotrophic denitrification (from 19.7% to 40.8%) and inhibited heterotrophic

276 denitrification (decreased to 46.9%), thereby resulting in a shift (8%) in the NO_3^- reduction pathway

277 from denitrification to dissimilatory NO_3^- reduction to NH_4^+. The addition of S^{2-} caused a proportional

278 increase in the population of sulfur-oxidizing nitrate-reducing bacteria (mainly Paracoccus) from

279 18.6% to 27.2% and suppressed heterotrophic nitrate-reducing bacteria (mainly Pseudoxanthomonas

280 and Pseudomonas), which caused a decrease (25.5%) in their population.

281 On the contrary, Zhao et al. (2006) found that more efficient removal of NH_4^+ was obtained when the

282 COD concentration was lower. In the studies of Wu et al. (2020), organic matter (300 mg COD/l in the

283 influent) negatively affected conventional anammox, but sulfammox was not affected. As a

284 consequence, the concentration of the dominant potential sulfammox bacteria (*Sulfurimonas,*

285 *Desulfovibrio, Desulfuromonas, Desulfobulbus, norank Rhodobacteraceae* and *Thiobacillus*) was

286 higher than the concentration of *Candidatus* Kuenenia performing conventional anammox.

287 4.5. Spontaneity and oxidation-reduction potential

288   Zhang et al. (2009) described the spontaneity of the sulfammox reaction. ΔG^θ of the sulfammox is

289   −45.35 kJ/mol. The reaction is obviously more difficult to proceed than conventional anammox, which

290   has ΔG^θ = -357 kJ/mol.

291   As the SO_4^{2-} dependent AAOB are obligate anaerobic bacteria, high substrate concentrations and a low

292   oxidation-reduction potential (ORP) (< -100 mV) can intensify the sulfammox process (Zhang et al.

293   2009; Ali et al. 2013). Fdz-Polanco et al. (2001a) found that the calculated values of redox potential

294   for the half reactions of reduction of N_2 to NH_4^+ and SO_4^{2-} to S^0 at pH = 8 was in the narrow range

295   from -330 to -360 mV. Those results suggested that SO_4^{2-} reduction and NH_4^+ oxidation could coexist

296   together under anaerobic conditions. Similar to conventional anammox, hydrazine injections have also

297   been reported to improve the sulfammox activity (Rikmann et al. 2012, 2014, 2016).

298   4.6. Other factors influencing the sulfammox process

299   In contrast, there are also several factors that may negatively affect the sulfammox process. Wu et al.

300   (2020) found that DO levels >0.3 - 0.5 mg/l could have a negative effect on sulfammox, as this leads

301   to partial nitration and the production of NO_2^-. DO inhibits the enrichment of the dominant bacteria

302   of both sulfammox and anammox and leads to the growth of AOB, competing with AAOB for NH_4^+.

303   High concentrations of NO_2^- and NO_3^- also favor SO_4^{2-} resynthesis as a result of sulfur-related

304   autotrophic denitrification. Rikmann et al. (2016) also pointed out that NO_2^- and HCO_3^- concentrations

305   exceeding 10 mg N/l and 1000 mg/l, respectively, disrupted sulfammox. - the latter because it

306   affected TN removal efficiency.

307   Yang et al. (2009) noted that NH_4^+ and SO_4^{2-} removal efficiencies could negatively be affected by the

308   presence of H_2S and S^{2-}. However, the authors did not provide the exact thresholds at which

309   sulfammox could be inhibited.

310   In the study of Zhao et al. (2006), the obtained efficiencies of NH_4^+ = 43% (low) and SO_4^{2-} = 59%

311   (high) implied a competition between SRB and not identified microorganisms responsible for

312   simultaneous removal of NH_4^+ and SO_4^{2-}. Therefore, it is worth of paying attention to the participation

313   of SRB in the sulfammox process, as they are responsible for the reduction of SO_4^{2-} to S^{2-} under

314   anaerobic conditions. High COD concentration increases the growth of SRB.

5. Sulfammox based reactors and reported efficiencies

316   Until now, the sulfammox process has been studied in different reactors in terms of the flow

317   conditions and biomass retention method (see: Table 2).

318

Tab. 2 Sulfammox based reactors and efficiency of NH_4^+, SO_4^{2-} removal

Reactor	Origin of biomass	Influent NH_4^+ [mg/l]	Influent SO_4^{2-} [mg/l]	NH_4^+ removal efficiency [%]	SO_4^{2-} removal efficiency [%]	Highlights of the study	Reference
Combining system: Upflow Anaerobic Sludge Blanket (UASB), Anoxic/Oxic Reactor (A/O), Anammox and Sulfammox Reactor (ANAOR), Anaerobic Sequencing Batch Reactor (ASBR)	landfill leachate	610-700	1870-1920	ca. 98	ca. 53	Landfill leachate was used as a substrate. The tests were carried out at a low temperature (14-15°C). The relative abundances of dominant sulfammox bacteria were 10-20 times higher that of *Candidatus* Kuenenia (anammox). **Reduction of SO_4^{2-} and NH_4^+ was considered as a combination of anammox, sulfammox, nitrification and denitrification processes.**	Wu et al. [2020]
Continuous Flow Stirred Tank Reactor (CFSTR)	long-term operation anammox up-flow reactor	110	0-110	ca. 40	ca. 0	SRAO occurred only in the cases of high amounts of inoculum biomass at DO = 0.2 - 0.5 mg/L). When DO<0.2 mg/L, the process was not observed. **SRAO was considered as a combination of aerobic ammonium oxidation, anammox, and heterotrophic sulfate reduction processes.**	Bi et al.. [2020]
		60	90	ca. 30	ca. 10		
		60	90	ca. 55	ca. 0		
Self-Designed Circulating Flowreactor (SDCF)	n.a.	120	183	ca. 30	ca. 40	NH_4^+ oxidization and SO_4^{2-} reduction efficiencies increased in the presence of NO_2^- and NO_3^-. *Proteobacteria, Chloroflexi, Bacteroidetes, Chlorobi, Acidobacteria, Planctomycetes* and *Nitrospirae* were detected. *Proteobacteria* were the dominant functional microorganisms removing nitrogen. **These results showed that nitrogen was converted by nitrification, denitrification, and conventional anammox, simultaneously with SRAO. The sulfur-based autotrophic denitration and denitrification in the reactor were caused by the influent NO_2^- and NO_3^-.**	Zhang et al.. [2019a]
		160	216	ca. 55	ca. 0		
		110	116	ca. 75	ca. 30		
		80	100	ca. 100	ca. 45		
		120	183	30	40		
		160	216	11	11		
		160	216	ca. 15	ca. 25		
		90	133	ca. 100	ca. 70		
Self-Designed Circulating Flowreactor (SDCF)	mixed sludge, which consisted anaerobic granular sludge from a municipal wastewater plant and denitrification sludge from a continuous stirred-tank reactor	50	90	ca. 40	ca. 30	The increasing ratio of N/S in the influent resulted in higher NO_2^- concentrations in the effluent. The microbial community comprised *Proteobacteria, Chloroflexi, Bacteroidetes, Chlorobi, Acidobacteria* and *Planctomycetes*. SRAO was mainly due to the high performance of Proteobacteria (12.4% of denitrifying bacteria were found in the biomass). **Part of nitrogen was converted by nitrification-denitrification, and conventional anammox, simultaneously with SRAO.**	Zhang et al.. [2019b]
		120	170	ca. 90	ca. 30		
		180	360	ca. 20	ca. 5		
Expanded Granular Sludge Bed (EGSB)	anaerobic hydrolysis acidification reactor	166-666	3600	40-58	64-71	The removal efficiency of SO_4^{2-} gradually improved as the influent NH_4^+ concentrations increased from 166-666 mg N/l to 1000-2000 mg N/l. At the same time, 71% NH_4^+ was removed. After increasing the NH_4^+ concentration to > 3000 mg N/l, the SO_4^{2-} reduction efficiency was reduced to 28%. **SRB and**	Wang et al. [2017a]
		1000-2000		40-70	66-82		
		>3000		10-25	28		

Reactor	Source					Description	Reference
						denitrifying bacteria were mainly responsible for SO$_4^{2-}$ and nitrogen removal.	
Anaerobic Sequencing Batch Reactor (ASBR)	activated sludge from the aerobic tank of digested liquor	97	261	ca. 88	ca. 19	**The presence of Planctomycetes revealed that anammox was a highly involved pathway in NH$_4^+$ removal, even without NO$_2^-$ in the feed. Other autotrophic denitrifying bacteria, related to species the** *Paracoccus Denitrificans*, were also present. These bacteria utilize S^0 as an electron donor and produce SO$_4^{2-}$, and competitively use NO$_2^-$ with anammox.	Pra-chakittikul et al.. [2016]
Moving Bed Biofilm Reactor (MBBR)	well-established attached anammox biofilm withdrawn from a lab-scale conventional anammox reactor treating reject water	69	ca. 70	ca. 30	ca. 10	SRAO tests were performed in MBBR at 20°C and UASBR at 36°C. Very similar results of NH$_4^+$ and overall SO$_4^{2-}$ removal were obtained in both reactors. **The SRAO process took place as one reaction of the multiple complex interactions between N-compounds, S-compounds, and organics (primarily humic matter) resulting in a significantly higher removal ratio of NH$_4^+$ than the SRAO stoichiometry predicts. It was postulated that the phylum Verrucomicrobia could also be involved in sulfammox.**	Rikmann et al.. [2016]
Upflow Anaerobic Sludge Blanket Reactor (UASBR)	anaerobic sludge from a yeast factory wastewater treating facility (Salutaguse, Estonia)	69	ca. 70	ca. 25	ca. 10		
Upflow Anaerobic Sludge Blanket Reactor (UASBR)	reject water from anaerobic digestion of municipal wastewater sludge	221	193	ca. 30	ca. 20	Sulfammox and anammox tests were carried out at 36°C and 20°C, respectively. NO$_2^-$ was proved to be a more efficient electron acceptor than SO$_4^{2-}$. **The reduction of SO$_4^{2-}$ and NH$_4^+$ was considered as a combination of sulfammox and denitrification processes.**	Rikmann et al.. [2014]
Expanded Bed Reactor (EBR)	lab-scale reactor treating N-NH$_4^+$ and SO$_4$-S simultaneously for more than two years	229	163	ca. 44	40	*Bacillus Benzoevorans* was isolated. Its optimum pH and temperature were 8.5 and 30°C, respectively. **The reduction of SO$_4^{2-}$ and NH$_4^+$ was considered as as sulfammox only.**	Cai et al.. [2010]
Upflow Anaerobic Sludge Blanket Reactor (UASBR)	nitrifying sludge in a municipal wastewater treatment plant	60	240	40	30	Sulfammox was successfully performed by changing NO$_2^-$ into SO$_4^{2-}$ as an electron acceptor. **The reduction of SO$_4^{2-}$ and NH$_4^+$ was considered as sulfammox only.**	Yang et al.. [2009]
Expanded Bed Reactor (EBR)	anaerobic digester in a municipal wastewater treatment plant	84-270 / 30-90	450-740 / 80-200	ca. 40 / ca. 55	ca. 10 / ca. 43	Sulfate-dependent anaerobic ammonium oxidation occurs with acclimated anaerobic digested sludge in the absence of organic matter. Anaerobic ammonium oxidation with sulfate does not tend to occur spontaneously due to its low ΔG^e value. The experiment demonstrated that high substrate concentrations and low ORP may be favorable for sulfammox. **The reduction of SO$_4^{2-}$ and NH$_4^+$ was as a sulfammox only.**	Zhang et al.. [2009]
Non-Woven Rotating Biological Contactor (NWRBC)	long-term operation anammox up-flow reactor	ca. 198	ca. 528	ca. 100	ca. 70	Bacteria belonging to *Planctomycetales*, especially the new species '*Anammoxoglobus Sulfate*, were identified as the functional community. **The reduction of SO$_4^{2-}$ and NH$_4^+$ was considered as a sulfammox only.**	Liu et al.. [2008]
Anaerobic Attached-Growth Bioreactor (AAGB)	anaerobic activated sludge collected from an anaerobic continuous stirred tank reactor	50	57	ca. 43	ca. 59	Low removal of NH$_4^+$ was obtained with high removal of SO$_4^{2-}$, implying the existence of competition between SRB) and microorganisms responsible for using SO$_4^{2-}$ and NH$_4^+$. Low COD, high SO$_4^{2-}$ and high NH$_4^+$ loadings at pH = 7.8 could	Zhao et al.. [2006]

						promoted sulfammox. **The reduction of SO_4^{2-} and NH_4^+ was considered as a sulfammox only.**	
Granular Activated Carbon Fluidized-Bed (GACFB)	diluted vinasse originating from an ethanol distillery plant processing beet sugar molasses	<10	1000	50	80	The first report on the sulfammox process. The "anomalous" NH_4^+ removal was obtained in a granular activated carbon (GAC) anaerobic fluidized-bed reactor. The reactor treated vinasse from an ethanol distillery of sugar beet molasses. About 50% of the influent nitrogen load was removed from the liquid phase appearing as N_2 in the gas phase. Simultaneously, only 20% of the SO_4^{2-} initially present in the influent appears as S^{2-} in the effluent or H_2S in the biogas, indicating that 80% of the sulfur was removed in sulfammox. **The reduction of SO_4^{2-} and NH_4^+ was considered as a combination of sulfammox and denitrification processes.**	Fdz-Polanco et al.. [2001b]

268   Zhang et al. (2019a) studied the effects of NO_2^- and NO_3^- on sulfammox and found that the removal

269   efficiencies of both NH_4^+ and SO_4^{2-} increased from 30% to 100% and from 40% to 70%, respectively,

270   while increasing NOx concentrations. Autotrophic denitrification had a large share in the removal.

271   With the influent NH_4^+ concentration of 80 mg N/l, SO_4^{2-} of 100 mg S/l and NO_2^- of 28 mg N/l, the

272   NH_4^+ removal efficiency reached almost 100%, while the overall SO_4^{2-} removal efficiency was only

273   45%. Similarly, with the influent NH_4^+ concentration of 90 mg N/l and SO_4^{2-} of 133 mg S/l and NO_3^-

274   of 90 mg N/l, the NH_4^+ and overall SO_4^{2-} removal efficiencies were approximately 100% and 70%,

275   respectively.

276   On the contrary, there have been studies indicating a lower efficiency of sulfammox for reject water,

277   i.e. approximately 30% and 10% for NH_4^+ and SO_4^{2-}, respectively (Rikmann et al. 2016). The influent

278   ratio of NH_4^+/SO_4^{2-} was implicitly a key factor as studied by Wang et al. (2017a). When the SO_4^{2-}

279   /NH_4^+ ratio was close to 2, the process efficiency was highest, while too low or too high ratios resulted

280   in lower efficiencies.

281   In an Expanded Granular Sludge Bed Reactor (EGSBR) performing sulfammox (Wang et al. 2017a),

282   the removal efficiency of SO_4^{2-} and organic compounds gradually improved from 64% to 71% and

283   66% to 82%, respectively, as the influent NH_4^+ concentrations increased from 166-666 mg N/l

284   (NH_4^+/SO_4^{2-} = 0.25-0.99) to 1000-2000 mg N/l (NH_4^+/SO_4^{2-} = 1.48-2.96). At the same time,

285   approximately 71% NH_4^+ was removed. However, after increasing the NH_4^+ concentration to >3000

286   mg N/l (NH_4^+/SO_4^{2-} > 4.44), the SO_4^{2-} reduction efficiency was reduced to approximately 28%. Zhao

287   et al. (2006) also reported that the volumetric NH_4^+ removal rates could reach the highest level when

288   the concentration of NH_4^+ was 450 mg N/l (37.5 g N/m^3/d), compared to 50 mg N/l (4.17 g N/m^3/d)

289   and 250 mg N/L (20.8 g N/m^3/d).

290   Wu et al. (2020) investigated the sulfammox process in a system consisting of four types of reactors

291   connected in series, including a UASBR, an anoxic/oxic reactor (A/O), an Anammox and Sulfammox

292   reactor (ANAOR), and an ASBR. In the first reactor (UASBR), the NH_4^+ concentration decreased

293   mainly due to dilution, while NO_2^- and NO_3^- (from nitrification solution recycle) were reduced by

294   denitrification. Partial nitrification was carried out at the A/O reactor, while anammox and sulfammox

295   were performed in the ANAOR and ASBR. In the ANAOR, NH_4^+ was removed by anammox (38 mg

296   NH_4^+/l) and sulfammox (148 mg NH_4^+/l). Those results indicated that the sulfammox share in the

297   NH_4^+ removal was more than 3 times higher than conventional anammox. Moreover, relatively high

298   amounts of NH_4^+ and SO_4^{2-} were removed in the ANAOR compared to other reactors. These amounts

299   were 187 mg N/l, 52 mg N/l and 35 mg N/l of NH_4^+ in the ANAOR, A/O and ASBR respectively. The

300   corresponding amounts for SO_4^{2-} were 393 mg S/1, 73.5 mg S/l and 42.3 mg S/l. The mass balance

301   calculations revealed that the combined system allowed to achieve the NH_4^+ removal efficiency at

302   98.5%, including 44.2% removed by sulfammox, whereas the overall SO_4^{2-} removal efficiency was

303   52.8%.

304   Rikmann et al. (2012) found that changing the electron acceptor from NO_2^- to SO_4^{2-} resulted in

305   reduction of the anammox efficiency. The efficiency of TN removal with NO_2^- was 85%, whereas after

306   changing to SO_4^{2-}, the average TN removal efficiency was only 23-24% in two different reactors

307   (MBBR and UASBR).

308   In order to compare sulfammox and conventional anammox, these processes were run in two parallel

309   UASBRs (Rikmann et al. 2014). It was assumed that a higher temperature could promote sulfammox,

310   partially compensating for its thermodynamic deficiency. Therefore, sulfammox and anammox

311   reactors were carried out at 36°C and 20°C, respectively. The use of NO_2^- as an electron acceptor was

312   still much more efficient than SO_4^{2-} as evidenced by the TN removal efficiency, i.e. 75% (conventional

313   anammox) and 17% (sulfammox), despite the significant temperature difference.

314   In the most recent study of Rikmann et al. (2016), sulfammox was carried out in a MBBR at 20°C and

315   a UASBR at 36°C. Very similar NH_4^+ and overall SO_4^{2-} removal efficiencies were obtained in both

316   reactors, i.e. 30% and 25% for NH_4^+ in the MBBR and UASBR, respectively, and 10% for SO_4^{2-} in

317   both reactors.

318   One of the principal drawbacks of sulfammox is the start-up time of the process. The sulfammox

319   reactor start-up takes even more time than conventional anammox due to the fact that the growth rate

320   of SO_4^{2-} dependent AAOB is very slow (Ali et al. 2013). For example, Zhang et al. (2009) found that

321   the cultivated sludge became capable of sulfammox reaction after 3 years of the operation under

322 anaerobic conditions. This makes sulfammox impossible to implement in the mainstream reactor. In

323 addition, when undesirable process disturbances occur, slow growth causes a long period of bacterial

324 regeneration. However, this disadvantage (slow start-up) can be partially overcome by enriching the

325 reactor by sulfammox consortia from marine sediments (Ali et al. 2013). Figure 5 summarizes all the

326 major research and discoveries related to the development of the sulfammox process.
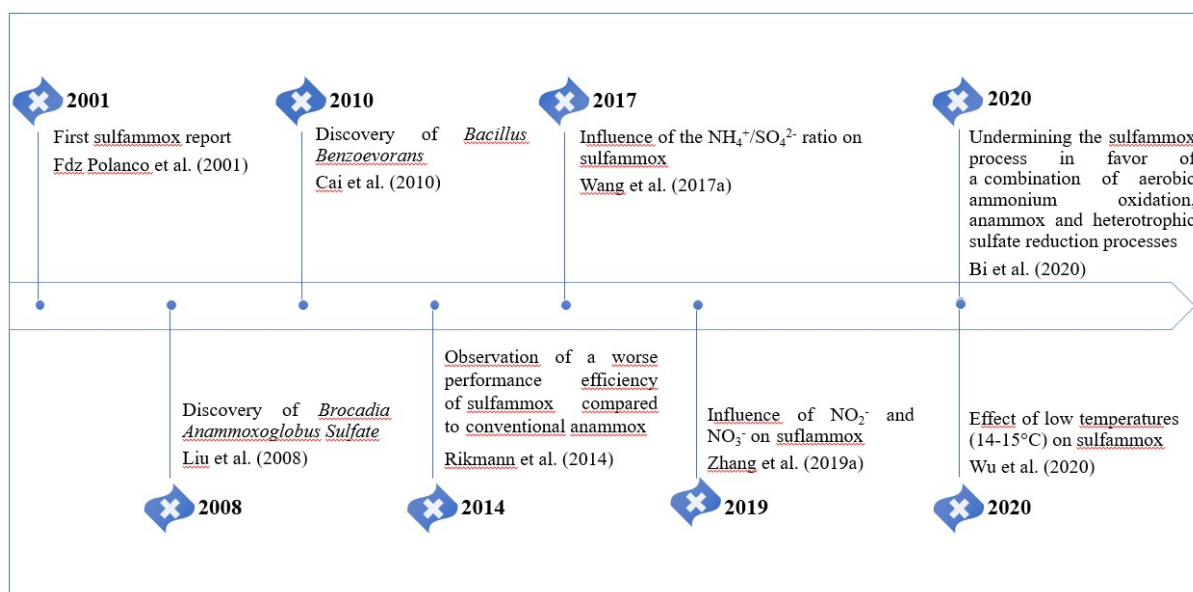
328 **Fig 5** Sulfammox process development timeline

6. Perspectives and conclusions

330 The conventional anammox process appears to be more advantageous than sulfammox for treatment of

331 nitrogen rich wastewater. With sulfammox, however, an economically inefficient pre-nitration step

332 (due to aeration) is not required and formation of toxic sulphide (S^{2-}) could be avoided. The main

333 disadvantage of SO_4^{2-} dependent AAOB, which is a very slow doubling time, could partially be

334 overcome by enriching inoculum biomass with marine sediments.

335 A combination of anammox and sulfur related processes (sulfammox and autotrophic denitrification)

336 would be a viable option for specific industrial wastewater with high content of nitrogen compounds

337 and SO_4^{2-}. There are more than 10 different novel systems in which sulfammox has been studied,

338 including suspended growth, biofilm, granular and hybrid reactors. Evidence suggests that high

339 removal efficiencies could be achieved with respect to both NH_4^+ (>90%) and SO_4^{2-} (>50%).

340

References

Ali M, Chai LY, Tang CJ, Zheng P, Min XB, Yang ZH, Xiong L, Song YX (2013) The increasing interest of ANAMMOX research in china: Bacteria, process development, and application. BioMed Res Int 2013, 134914. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1155/2013/134914

Bi Z, Wanyan D, Li X, Huang Y (2020) Biological conversion pathways of sulfate reduction ammonium oxidation in anammox consortia. Front Environ Sci Eng 14. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1007/s11783-019-1217-1

Cai J, Jiang JX, Zheng P (2010) Isolation and identification of bacteria responsible for simultaneous anaerobic ammonium and sulfate removal. Sci China Chem 53:645-650. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1007/s11426-010-0053-8

Di Capua F, Pirozzi F, Lens PNL, Esposito G (2019) Electron donors for autotrophic denitrification. Chem Eng J 362:922-937. https://doi.org/10.1016/j.cej.2019.01.069

Fdz-Polanco F, Fdz-Polanco M, Fernandez N, Urueña MA, Garcia PA, Villaverde S (2001b) New process for simultaneous removal of nitrogen and sulphur under anaerobic conditions. Water Res 35:1111-1114. https://doi.org/10.1016/S0043-1354(00)00474-7

Fdz-Polanco F, Fdz-Polanco M, Fernández N, Urueña MA, García PA, Villaverde S (2001a) Combining the biological nitrogen and sulfur cycles in anaerobic conditions. Water Sci Technol 44:77-84. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.2166/wst.2001.0469

Guo H, Chen C, Lee DJ, Wang A, Ren N (2013) Sulfur-nitrogen-carbon removal of Pseudomonas sp. C27 under sulfide stress. Enzyme Microb Technol 53:6-12. https://doi.org/10.1016/j.enzmictec.2013.04.002

In 't Zandt MH, de Jong AEE, Slomp CP, Jetten MSM (2018) The hunt for the most-wanted chemolithoautotrophic spookmicrobes. FEMS Microbiol Ecol 94: fiy064. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1093/femsec/fiy064

365 Kalyuzhnyi S, Gladchenko M, Mulder A, Versprille B (2006) DEAMOX-New biological nitrogen

366 removal process based on anaerobic ammonia oxidation coupled to sulphide-driven conversion of

367 nitrate into nitrite. Water Res 40:3637-3645. https://doi.org/10.1016/j.watres.2006.06.010

368 Kartal B, van Niftrik L, Keltjens JT, Op den Camp HJM, Jetten MSM (2012) Anammox-Growth

369 Physiology, Cell Biology, and Metabolism. Adv Microb Physiol 60: 211-262.

370 https://doi.org/10.1016/B978-0-12-398264-3.00003-6

371 Kosugi Y, Matsuura N, Liang Q, Yamamoto-Ikemoto R (2019) Nitrogen flow and microbial

372 community in the anoxic reactor of "Sulfate Reduction, Denitrification/Anammox and Partial

373 Nitrification" process. Biochem Eng J 151. https://doi.org/10.1016/j.bej.2019.107304

374 Li W, Zhao QI, Liu H (2009) Sulfide removal by simultaneous autotrophic and heterotrophic

375 desulfurization-denitrification process. J Hazard Mater 162: 848-853.

376 https://doi.org/10.1016/j.jhazmat.2008.05.108

377 Liu C, Zhao D, Yan L, Wang A, Gu Y, Lee DJ (2015a) Elemental sulfur formation and nitrogen

378 removal from wastewaters by autotrophic denitrifiers and anammox bacteria. Bioresour Technol

379 191:332-336. https://doi.org/10.1016/j.biortech.2015.05.027

380 Liu Y, Xi GJ, Xing DF, Liu BF, Ding J, Cao GL, Ren NQ (2021) Sulfate dependent ammonium

381 oxidation: A microbial process linked nitrogen with sulfur cycle and potential

382 application. Environmental Research, 192. https://doi.org/10.1016/j.envres.2020.110282

383 Liu S, Yang F, Gong Z, Meng F, Chen H, Xue Y, Furukawa K (2008) Application of anaerobic

384 ammonium-oxidizing consortium to achieve completely autotrophic ammonium and sulfate removal.

385 Bioresour Technol 99:6817-6825. https://doi.org/10.1016/j.biortech.2008.01.054

386 Liu ZC, Yuan LJ, Zhou GB, Li J (2015b) Achievement of sulfate-reducing anaerobic ammonium

387 oxidation reactor started with nitrate-reducing anaerobic ammonium oxidation. Huanjing Kexue

388 36:3345-3351.

389 Prachakittikul P, Wantawin C, Noophan P, Boonapatcharoen N (2016) ANAMMOX-like performanc-

390 es for nitrogen removal from ammonium-sulfate-rich wastewater in an anaerobic sequencing batch

reactor. J Environ Sci Health Part A Toxic Hazard Subst Environ Eng 51:220-228. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1080/10934529.2015.1094336

Qin Y, Wu C, Chen B, Ren J, Chen L (2019) Short term performance and microbial community of a sulfide-based denitrification and Anammox coupling system at different N/S ratios. Bioresour Technol 294, 122130. https://doi.org/10.1016/j.biortech.2019.122130

Rikmann E, Zekker I, Tomingas M, Tenno T, Loorits L, Vabamäe P, Mandel A, Raudkivi M, Daija L, Kroon K, Tenno T (2016) Sulfate-reducing anammox for sulfate and nitrogen containing wastewaters. Desalin Water Treat 57:3132-3141. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1080/19443994.2014.984339

Rikmann E, Zekker I, Tomingas M, Tenno T, Menert A, Loorits L, Tenno T (2012) Sulfate-reducing anaerobic ammonium oxidation as a potential treatment method for high nitrogen-content wastewater. Biodegradation 23:509-524. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1007/s10532-011-9529-2

Rikmann E, Zekker I, Tomingas M, Vabamäe P, Kroon K, Saluste A, Tenno T, Menert A, Loorits L, dC Rubin SSC, Tenno T (2014) Comparison of sulfate-reducing and conventional Anammox upflow anaerobic sludge blanket reactors. J Biosci Bioeng 118:426-433. https://doi.org/10.1016/j.jbiosc.2014.03.012

Rios del Toro EE (2017) Anaerobic ammonium oxidation in marine environments: Contribution to biogeochemical cycles and biotechnological developments for wastewater treatment. Dissertation. Instituto Potosino De Investigación Científica Y Tecnológica, A.C.

Rios-Del Toro EE, Cervantes FJ (2019) Anaerobic ammonium oxidation in marine environments: contribution to biogeochemical cycles and biotechnological developments for wastewater treatment. Rev Environ Sci Biotechnol 18:11-27. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1007/s11157-018-09489-3

Rios-Del Toro EE, Valenzuela EI, López-Lozano NE, Cortés-Martínez MG, Sánchez-Rodríguez MA, Calvario-Martínez O, Sánchez-Carrillo S, Cervantes FJ (2018) Anaerobic ammonium oxidation linked

to sulfate and ferric iron reduction fuels nitrogen loss in marine sediments. Biodegradation 29:429-442. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1007/s10532-018-9839-8

Schrum HN, Spivack AJ, Kastner M, D'Hondt S (2009) Sulfate-reducing ammonium oxidation: A thermodynamically feasible metabolic pathway in subseafloor sediment. Geology 37:939-942. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1130/G30238A.1

Strous M, Pelletier E, Mangenot S, Rattei T, Lehner A, Taylor MW, Horn M, Daims H, Bartol-Mavel D, Wincker P, Barbe V, Fonknechten N, Vallenet D, Segurens B, Schenowitz-Truong C, Médigue C, Collingro A, Snel B, Dutilh BE, Op Den Camp HJM, Van Der Drift C, Cirpus I, Van De Pas-Schoonen KT, Harhangi HR, Van Niftrik L, Schmid M, Keltjens J, Van De Vossenberg J, Kartal B, Meier H, Frishman D, Huynen MA, Mewes HW, Weissenbach J, Jetten MSM, Wagner M, Le Paslier D (2006) Deciphering the evolution and metabolism of an anammox bacterium from a community genome, Nature 440:790-794. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1038/nature04647

Ucar D, Yilmaz T, Di Capua F, Esposito G, Sahinkaya E (2020) Comparison of biogenic and chemical sulfur as electron donors for autotrophic denitrification in sulfur- fed membrane bioreactor (SMBR). Bioresour Technol 299. https://doi.org/10.1016/j.biortech.2019.122574van der Star WRL, Abma WR, Blommers D, Mulder JW, Tokutomi T, Strous M, Picioreanu C, van Loosdrecht MCM (2007) Startup of reactors for anoxic ammonium oxidation: Experiences from the first full-scale anammox reactor in Rotterdam. Water Res 41:4149-4163. https://doi.org/10.1016/j.watres.2007.03.044

Wang D, Liu B, Ding X, Sun X, Liang Z, Sheng S, Du L (2017a) Performance evaluation and microbial community analysis of the function and fate of ammonia in a sulfate-reducing EGSB reactor. Appl Microbiol Biotechnol 101:7729-7739. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1007/s00253-017-8514-z

Wang T, Guo J, Lu C, Li H, Han Y, Song Y, Hou Y, Zhang J (2020) Faster removal of nitrite than nitrate in sulfur-based autotrophic denitrification coupled with anammox, affected by the anammox effluent. Environ Sci Water Res Technol 6:916-924. https://doi-1org-10000044b05c9.han.bg.pg.edu.pl/10.1039/D0EW00065E

443 Wang XN, Sun GX, Zhu YG (2017b) Thermodynamic energy of anaerobic microbial redox reactions

444 couples elemental biogeochemical cycles. J Soils Sed 17:2831-2846. https://doi-1org-

445 10000044b05c9.han.bg.pg.edu.pl/10.1007/s11368-017-1767-4

446 Wisniewski K, di Biase A, Munz G, Oleszkiewicz JA, Makinia J (2019) Kinetic characterization of

447 hydrogen sulfide inhibition of suspended anammox biomass from a membrane bioreactor. Biochem

448 Eng J 143:48-57. https://doi.org/10.1016/j.bej.2018.12.015

449 Wu L, Yan Z, Li J, Huang S, Li Z, Shen M, Peng Y (2020) Low temperature advanced nitrogen and

450 sulfate removal from landfill leachate by nitrite-anammox and sulfate-anammox. Environ Pollut 259,

451 113763. https://doi.org/10.1016/j.envpol.2019.113763

452 Xie GJ, Cai C, Hu S, Yuan Z (2017) Complete nitrogen removal from synthetic anaerobic sludge

453 digestion liquor through integrating anammox and denitrifying anaerobic methane oxidation in a

454 membrane biofilm reactor. Environ Sci Technol 5 :819-827.

455 https://doi-1org-10000044b0f55.han.bg.pg.edu.pl/10.1021/acs.est.6b04500

456 Xu LZJ, Zhang Q, Fu JJ, Zhang JT, Zhao YH, Jin LY, Fan NS, Huang BC, Jin RC (2020) Deciphering

457 the microbial community and functional genes response of anammox sludge to sulfide stress.

458 Bioresour Technol 302, 122885. https://doi.org/10.1016/j.biortech.2020.122885

459 Xu X, Chen C, Lee DJ, Wang A, Guo W, Zhou X, Guo H, Yuan Y, Ren N, Chang JS (2013) Sulfate-

460 reduction, sulfide-oxidation and elemental sulfur bioreduction process: Modeling and experimental

461 validation. Bioresour Technol 147:202-211. https://doi.org/10.1016/j.biortech.2013.07.113

462 Yang Z, Zhou S, Sun Y (2009) Start-up of simultaneous removal of ammonium and sulfate from an

463 anaerobic ammonium oxidation (anammox) process in an anaerobic up-flow bioreactor. J Hazard

464 Mater 169:113-118. https://doi.org/10.1016/j.jhazmat.2009.03.067

465 Yin Z, Xie L, Cui X, Zhou Q (2017) Effective carbon and nitrogen removal with reduced sulfur

466 oxidation in an anaerobic baffled reactor for fresh leachate treatment. J Biosci Bioeng 123(1):84-90.

467 https://doi.org/10.1016/j.jbiosc.2016.07.004

468   Yu H, Wang AJ, Chen C (2013) Structure and dynamics of microbial community in the denitrifying

469   sulfide removal process. Huanjing Kexue 34:1190-1195.

470   Zhang D, Cui L, Madani RMA, Wang H, Zhu H, Liang J (2019a) Effect of nitrite and nitrate on

471   sulfate reducing ammonium oxidation. Water Sci Technol 80:634-643. https://doi-1org-

472   10000044b05c9.han.bg.pg.edu.pl/10.2166/wst.2019.277

473   Zhang D, Cui L, Wang H, Liang J (2019b) Study of sulfate-reducing ammonium oxidation process

474   and its microbial community composition. Water Sci Technol 79:137-144. https://doi-1org-

475   10000044b05c9.han.bg.pg.edu.pl/10.2166/wst.2019.027

476   Zhang L, Zheng P, He Y, Jin R (2009) Performance of sulfate-dependent anaerobic ammonium

477   oxidation.   Sci   China   Ser   B   Chem   52(1):86-92.   https://doi-1org-

478   10000044b05c9.han.bg.pg.edu.pl/10.1007/s11426-008-0161-x

479   Zhao QI, Li W, You SJ (2006) Simultaneous removal of ammonium-nitrogen and sulphate from

480   wastewaters with an anaerobic attached-growth bioreactor. Water Sci Technol 54:27-35. https://doi-

481   1org-10000044b05c9.han.bg.pg.edu.pl/10.2166/wst.2006.762

482