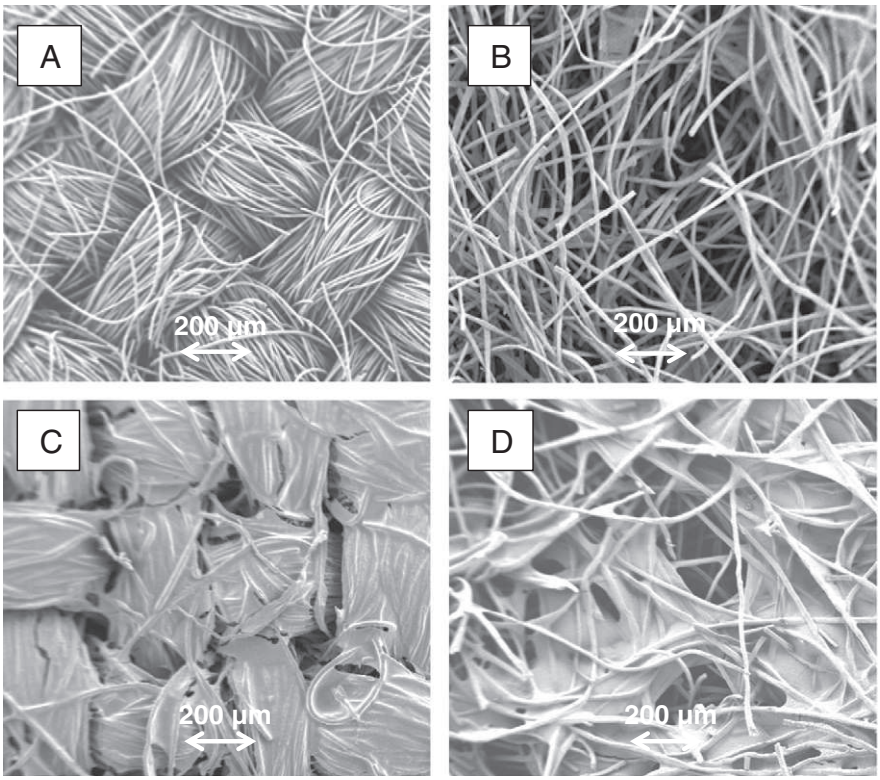

Fig. 2. SEM images of clean electrodes of A/carbon cloth, B/carbon felt, and secondary bioanodes on C/carbon cloth and D/carbon felt formed at +0.15 V/SCE (day 37).

from around 20 to 200 µm. At the end of the chronoamperometry (37 days) SEM showed an almost uniform biofilm on carbon cloth, while the carbon felt surface was only partly clogged (Fig. 2C-D).

Primary and secondary bioanodes were characterized by three-dimensional epifluorescent microscopy (Fig. 3A–B). The microbial volume ratios were 27 ± 6.8% and 12 ± 4.4% on carbon cloth and carbon felt, respectively, for the primary bioanodes. The secondary bioanodes showed a significant microbial volume increase to 39.3 ± 1.1% and 16.3 ± 2.7% on carbon cloth and carbon felt, respectively. Improvement of the current density from the primary to the secondary bioanodes corresponded to a significant microbial volume increase. The current density provided by the electrodes was straightforwardly linked to the microbial volume ratio.

Carbon cloth has a surface fully accessible for the microorganisms and led to an almost uniform biofilm with a thickness of the order of 80–120 µm and a microbial volume ratio of 39.3 ± 1.1%. Carbon felt led to microbial volume ratio of only 16.3 ± 2.7%. The epifluorescent images showed that the nucleic acids were mainly accumulated around the electrode fibers (Fig. 3). The microbial volumes were consequently higher on the cloth than on the felt structure because the fibers were denser in the cloth configuration.

Sectional cuts of the carbon felt bioanodes (Fig. 4) showed that fibers on the surface are completely covered with a biofilm wrapped around them, while the fibers in the center are not colonized at all. The biofilm penetration depth measured in several spots of the sectional cut view was in the range from 200 to 800 µm. As the biofilm was settled on the two sides of the felt, which has a total thickness of 5000 µm, it can be concluded that the biofilm colonized only 8%–32% of the total felt volume.

It must be recalled that the microbial volume ratios were measured only on the 117-µm-thick upper layer of the bioanodes (see Section 2.4.3). On the felt electrode, the 117-µm upper part of the biofilm had a lower microbial volume (16.3 ± 2.7%) than the biofilm formed on the cloth (39.3 ± 1.1%) but the biofilm penetrated 200–800 µm inside

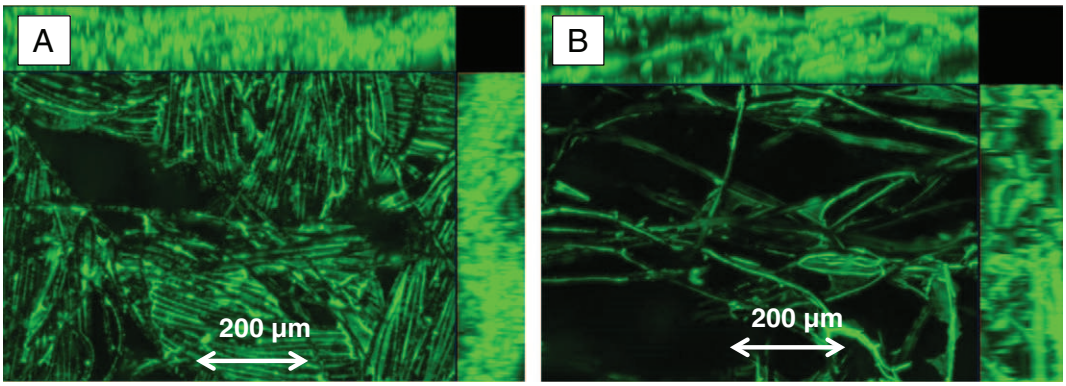

Fig. 3. 3D epifluorescent microscopy of secondary bioanodes formed under polarization at +0.15 V/SCE on A/carbon cloth and B/carbon felt (day 37).

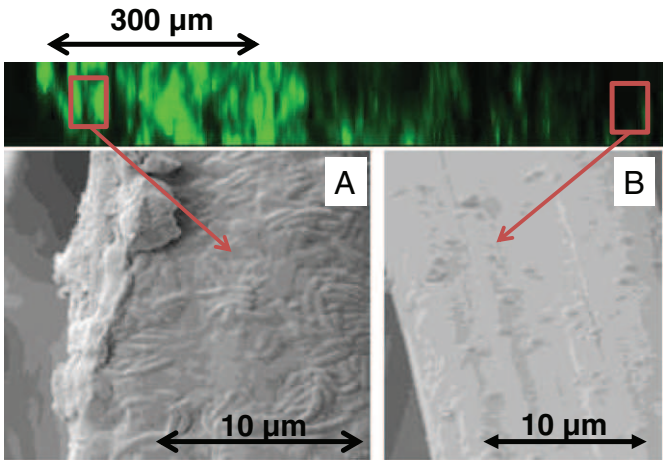

Fig. 4. Sectional views of a carbon felt secondaty bioanode (37 day) observed by epifluorescent microscopy (on top) and SEM of two different fibers (A) on the surface of the carbon felt (B) 1 mm deep in the carbon felt (B).

the felt electrode. The biofilm penetration inside the felt compensated for the lower microbial volume, and resulted in similar electrochemical performance for both structures.

For both structures, SEM showed a biofilm on the electrode surfaces (Fig. 2C-D), which partly clogged the porosity of the felt electrode. This surface biofilm can explain the weak biofilm penetration in the felt. A previous study has shown that bioanodes formed on the same carbon felt provided current density as high as 80 A/m^2 in highly saline media (45 g/L NaCl) [19]. Such hard conditions led to the strong selection of halotolerant species, resulting in a biofilm mainly wrapped around the fibers inside the felt structure [20]. In contrast, in the chemically rich medium used here, the non-selective conditions led to the easy formation of a surface biofilm that hindered deep internal colonization and explained the modest performance of the 3D felt.

This result is similar to that reported recently with pure cultures of *Geobacter sulfurreducens* [15]. Activated carbon was compared to graphite felt and, unexpectedly, the surface area of the electrode material proved to have negligible influence on the electrochemical performance. The thick biofilm formed by *G. sulfurreducens* was assumed to be an element of the explanation [15]. This previous study, performed with a model system, and the present work, carried out in close-to-industrial conditions, similarly evidenced that the biofilm pattern is a major parameter controlling the bioanode performance. The biofilm pattern can annihilate the advantage of 3D porous electrodes that has been postulated a priori so far. This issue now deserves dedicated investigations.

4. Conclusion

Bioanodes were formed in wastewater used as the medium and fed with food wastes. In this context, 2D cloth performed similarly to 3D felt. The higher efficiency of 3D electrodes, which has generally been postulated a priori so far, should now be called into question depending on the operating conditions. The study has shown that the biofilm pattern is a major parameter that controls the electrochemical performance and can annihilate the advantage of 3D porous electrodes. In industrially-scalable conditions, 2D electrodes should now be considered as a worthwhile solution, particularly with the objective of subsequently designing multilayer electrodes.

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