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Performance of different *Sporomusa* species for the microbial electrosynthesis of acetate from carbon dioxide

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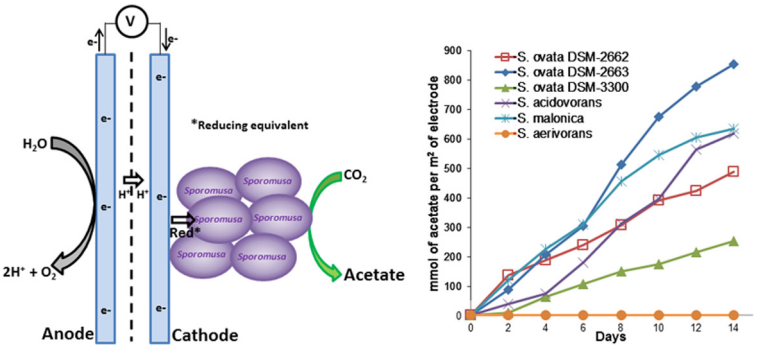
HIGHLIGHTS

- Six *Sporomusa* species were evaluated for microbial electrosynthesis activity.
- *S. ovata* DSM-2663 was the best microbial catalyst for MES among the tested species.
- MES performance varies greatly among the six *Sporomusa* species.
- *Sporomusa* oxidizing H_2 efficiently are not necessarily good MES microbial catalysts.

GRAPHICAL ABSTRACT



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ABSTRACT

Sporomusa ovata DSM-2662 produces high rate of acetate during microbial electrosynthesis (MES) by reducing CO_2 with electrons coming from a cathode. Here, we investigated other *Sporomusa* for MES with cathode potential set at −690 mV vs SHE to establish if this capacity is conserved among this genus and to identify more performant strains. *S. ovata* DSM-2663 produced acetate 1.8-fold faster than *S. ovata* DSM-2662. On the contrary, *S. ovata* DSM-3300 was 2.7-fold slower whereas *Sporomusa aerivorans* had no MES activity. These results indicate that MES performance varies among *Sporomusa*. During MES, electron transfer from cathode to microbes often occurs *via* H_2. To establish if efficient coupling between H_2 oxidation and CO_2 reduction may explain why specific acetogens are more productive MES catalysts, the metabolisms of the investigated *Sporomusa* were characterized under H_2:CO_2. Results suggest that other phenotypic traits besides the capacity to oxidize H_2 efficiently are involved.

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1. Introduction

Microbial electrosynthesis (MES) technology is being developed for the production of multicarbon commodities such as acetate and biofuels from the greenhouse gas CO_2 and from electrons coming from the cathode of a bioelectrochemical system (BES) (Lovley and Nevin, 2013 and Rabaey and Rozendal, 2010). One of the main features of MES is that electrical energy powers directly the biological synthesis of specific chemicals (Tremblay and Zhang, 2015). For instance, MES can be driven with electricity surplus from the power grid to store electrical energy into the chemical bonds of products of interest (Jürgensen et al., 2014 and Tremblay and Zhang, 2015). MES can also be coupled with photoelectrodes or photovoltaics to carry out artificial bioinorganic photosynthesis, a

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bioproduction process that has the potential to be significantly more efficient than natural photosynthesis (Liu et al., 2015, 2016; Lovley and Nevin, 2013; Nichols et al., 2015 and Zhang, 2015).

BES reactors employed for MES often consist of an anodic chamber and a cathodic chamber separated by an ion-exchange membrane (IEM) (Jourdin et al., 2014; LaBelle et al., 2014; Nevin et al., 2010 and Nevin et al., 2011). Protons and electrons required for CO_2 reduction are generated at the anode *via* oxidation reactions that can be abiotic or biologically-driven (Rabaey and Rozendal, 2010 and Villano et al., 2010). The microbial catalyst grown in the cathodic chamber reduces CO_2 to chemicals with electrons from the cathode and protons that passed through the IEM. The whole process requires an external source of power to drive both the anodic and cathodic reactions.

Compared to other autotrophic microbes, acetogens are microbial catalysts of choice for MES or for other autotrophic bioprocess such as gas fermentation because CO_2 is their final electron acceptor (Drake et al., 1997, 2008 and Ragsdale and Pierce, 2008). This means that most electrons coming from a cathode or other electron donors end up in the reduced products resulting in high coulombic efficiency (Tremblay and Zhang, 2015). Acetogens reduce CO_2 via the Wood-Ljungdahl pathway mainly into acetate but also into ethanol, butyrate, 2,3-butanediol, and 1-butanol (Schiel-Bengelsdorf and Dürre, 2012). In MES reactors inoculated with environmental samples and treated to inhibit methanogenesis, acetogens such as *Acetobacterium* sp. usually become predominant and produce primarily acetate from CO_2 and electricity (Bajracharya et al., 2015, 2016, 2017; Jourdin et al., 2014, 2015; LaBelle et al., 2014; Marshall et al., 2012, 2013; Mohanakrishna et al., 2015). Furthermore, pure culture-driven MES system has been demonstrated with different acetogenic species including *Sporomusa ovata*, *Sporomusa sphaeroides*, *Sporomusa silvacetica*, *Acetobacterium woodii*, *Clostridium ljungdahlii*, *Clostridium aceticum*, and *Moorella thermoacetica* (Arends, 2013; Gong et al., 2013; Nevin et al., 2010, 2011; Nie et al., 2013 and Zhang et al., 2013).

Sporomusa ovata DSM-2662 and strains derived from it are some of the most productive and efficient acetogenic bacteria capable of driving MES (Aryal et al., 2016; Chen et al., 2016 and Tremblay et al., 2015). The affinity of the *Sporomusa* genus for the cathode as a source of electrons is also highlighted by the fact that a microbial community enriched in a MES system from an environmental inoculum has been shown to contain a large population of *Sporomusa* spp. (Zhu et al., 2015). It has been proposed that one of the major routes for electron transfer from the cathode of a MES reactor to acetogens is *via* molecular H_2 (May et al., 2016 and Tremblay et al., 2016). Besides MES, *S. ovata* DSM-2662 is also an efficient strain for H_2:CO_2 gas fermentation for which it has been shown to have a high acetate production rate compared to other acetogens (Groher and Weuster-Botz, 2016). The performance of *S. ovata* DSM-2662 in MES reactor as well as during H_2:CO_2 fermentation could be explained in part by a high metabolic capacity for the usage of H_2 as an electron donor.

The main purpose of the study presented here was to determine if the capacity of reducing CO_2 *via* MES is conserved throughout the *Sporomusa* genus and if *S. ovata* DSM-2662 is really the most productive *Sporomusa* wild type strain. Thus, different *Sporomusa* never evaluated before with a cathode as the electron source were screened for MES performance and compared with *S. ovata* DSM-2662. This includes *S. ovata* DSM-2663, *S. ovata* DSM-3300, *Sporomusa acidovorans*, *Sporomusa malonica* and *Sporomusa aerivorans*. Additionally, in order to establish if acetogens efficient at reducing CO_2 to acetate with H_2 as the electron donor are automatically good microbial catalyst for MES, the growth and acetate production rate of *Sporomusa* spp. were studied under a H_2:CO_2 atmosphere.

2. Materials and methods

2.1. Strains and culture conditions

S. ovata DSM-2662, *S. ovata* DSM-2663, *S. ovata* DSM-3300, *S. acidovorans* DSM-3132, *S. malonica* DSM-5090 and *S. aerivorans* DSM-13326 were obtained from the Deutsche Sammlung Mikroorganismen und Zellkulturen (DSMZ) (Table 1). Under a N_2:CO_2 (80:20) atmosphere, *S. ovata* strains, *S. acidovorans* and *S. malonica* were routinely maintained in the DSM 311 medium of the Deutsche Sammlung Mikroorganismen (DSM) with 40 mM betaine as substrate, whereas *S. aerivorans* was maintained in the DSM 503a medium in the presence of 5 g l^{-1} Na-DL-lactate. Each culture was transferred at least three times under autotrophic conditions with a H_2:CO_2 (80:20) atmosphere (1.7 atm) before subsequent experiments. Casitone, sodium sulfide, and resazurin were omitted for all experiments with DSM-311 medium. Yeast extract was also omitted for experiments with *S. ovata* strains whereas 0.3 g l^{-1} yeast extract was added for experiments with *S. acidovorans* or *S. malonica*. Casamino acids, resazurin and dithiothreitol were omitted and 0.3 g l^{-1} yeast extract was added for experiments with *S. aerivorans* in DSM 503a medium. *S. acidovorans*, *S. malonica* and *S. aerivorans* cannot grow without yeast extract addition. For MES experiments, cysteine was also omitted from all cultivation media. For all the different *Sporomusa* species tested in this study and for every media employed here, no growth or acetate production was observed in the absence of either H_2 or the poised cathode of a MES reactor as electron donor, indicating that small amount of yeast extract or other components of the culture media were not used as growth substrates.

2.2. Microbial electrosynthesis of acetate from CO_2

H-cell type three-electrode MES reactors were operated at 25 °C with different *Sporomusa* grown in the cathode chamber as described previously (Tremblay et al., 2015). Briefly, the graphite stick cathode (36 cm^2) and anode (36 cm^2) were suspended in 250 ml of cultivation medium in two chambers separated by a Nafion 115 ion-exchange membrane (Ion Power, Inc., New Castle, DE, USA). All experiments repeated in triplicate were carried out with a CH Instrument potentiostat (CH Instruments, Inc, USA) and the cathode potential was set at −690 mV versus Standard Hydrogen Electrode (SHE). 100 ml of *Sporomusa* cultures grown on H_2:CO_2 at an optical density (545 nm) of ca. 0.1 were used to inoculate the cathode chamber. *Sporomusa* cultures were further established in the cathode chamber by bubbling with a hydrogen-containing gas mix N_2:CO_2:H_2 (83:10:7) and by swapping the medium three times. Each medium swap was performed after the *Sporomusa* cultures reached an OD_{545} of ca. 0.1. After the third medium swap, the gas mix was switched to N_2:CO_2 (80:20) for few hours before data start being collected for the MES experiments. During the whole experiment, the anode chamber was continuously bubbled with N_2:CO_2. Electrochemical data were collected for a period of two weeks with the potentiostat and analyzed with EC-Lab v.10.2 software (BioLogic, France).

2.3. Acetate concentration measurement

Acetate was quantified by High Performance Liquid Chromatography (HPLC) as previously described (Tremblay et al., 2015). Briefly, a HPX-87 H anion exchange column (Bio-Red Laboratories Inc., California, USA) at 30 °C was used with 5 mM H_2SO_4 as the mobile phase at a flow rate of 0.6 ml min^{-1}. A refractive index detector was used for detection and the results were analyzed with the Chromeleon 7 software (ThermoFisher Scientific, Denmark).

Table 1
Important characteristics of *Sporomusa* strains investigated in this study.

Strain	Other designation	Isolation source	Autotrophic growth mode	Other characteristic	Reference
S. ovata DSM-2662	Type strain, H1	Sugar beet leaf silage	MES, H_2:CO_2, formate, methanol	Gram−, Spore-forming	Möller et al. (1984) and Nevin et al. (2010)
S. ovata DSM-2663	H6	Mud, Leine river	MES, H_2:CO_2, formate, methanol	Gram−, Spore never observed	Möller et al. (1984) This study
S. ovata DSM-3300	Nile	Mud, Nile river	MES, H_2:CO_2, formate, methanol	Gram -, Spore-forming	Möller et al. (1984) This study
S. acidovorans DSM-3132	Type strain, Mol	Alcohol distillation wastewater fermenter	MES, H_2:CO_2, formate, methanol	Gram−, Spore-forming, yeast extract required for growth	Ollivier et al. (1985) This study
S. malonica DSM-5090	Type strain, Wo Gl2	Mud of polluted freshwater	MES, H_2:CO_2, formate, methanol	Gram−, Spore-forming, yeast extract required for growth	Dehning et al. (1989) This study
S. aerivorans DSM-13326	Type strain, TmAO3	Gut of soil-feeding termite	H_2:CO_2, formate, methanol	Gram−, Spore-forming, High capacity for oxygen scavenging, yeast extract required for growth	Boga et al. (2003) This study

Unpaired *t*-test was used to evaluate the statistical significance of difference observed between *Sporomusa* in acetate production rate as well as in current density and doubling time.

2.4. Confocal Laser Scanning Microscopy

For Confocal Laser Scanning Microscopy (CLSM) image, cathodes were removed from MES reactors after two weeks of operation and treated with LIVE/DEAD® BacLight™ Bacterial Viability Kit (ThermoFisher Scientific) to stain *Sporomusa* cells as described previously (Aryal et al., 2016). CLSM images were taken with a LSM5 Pascal laser scanning microscope (Zeiss) and the images were further analyzed with the ZEN imaging software (Zeiss, Germany). Z-stack composite images formed with multiple images taken at different focal distances are presented here to render the many layers of bacterial cells present at the surface of the cathode.

2.5. Whole-genome sequencing

Genomes of *S. ovata* DSM-2663 and *S. ovata* DSM-3300 were sequenced as described previously (Tremblay et al., 2015). Briefly, Genomic DNA of both strains was extracted with Easy-DNA gDNA purification Kit (Life Technologies, Carlsbad, CA). Genomic libraries were generated with the TruSeq Nano DNA LT Sample Preparation Kit (Illumina Inc., San Diego CA). Average size of dsDNA fragments in the libraries was determined with an Agilent 2100 Bioanalyzer. Sequencing was carried out with a MiSeq Reagent kit v2 (300 cycles) on a MiSeq (Illumina) platform with a paired-end protocol and read lengths of 151 nucleotides. The sequencing reads were trimmed with Trimmomatic (Bolger et al., 2014) and variant calling was done with breseq (Deatherage and Barrick, 2014). The reference genome was *S. ovata* DSM 2662 (NCBI accession ASXP00000000.1) (Poehlein et al., 2013). All the samples had an average coverage of at least 30X.

3. Results and discussion

3.1. MES with Sporomusa ovata strains

Until now, three different strains of *S. ovata* have been isolated and deposited into culture collections (Table 1). Of those three strains, only *S. ovata* DSM-2662 (ATCC 35899) has been tested and shown to drive MES with cathodes made of different materials and poised at potentials ranging from −400 to −690 mV vs SHE (Aryal et al., 2016; Chen et al., 2016; Nevin et al., 2010; Nie et al., 2013; Tremblay et al., 2015 and Zhang et al., 2013). In an effort to identify better microbial catalysts for pure culture-driven MES and to establish if the capacity to drive MES is conserved in all *S. ovata* strains, the three strains were tested in a H-cell MES reactor (Fig. 1). The acetate production rate was increased 1.8-fold (p-value of 0.07) and the current density was enhanced 1.7-fold (p-value of 0.07) with *S. ovata* DSM-2663 compared to that of *S. ovata* DSM-2662 during MES (Fig. 1A and B; Table 2). On the contrary, *S. ovata* DSM-3300 was significantly less performant for MES with a current density 2.4 times lower (p-value of 0.03) and an acetate production rate 2.6 times lower (p-value of 0.03) than *S. ovata* DSM-2662 (Fig. 1A and C; Table 2). Compared to *S. ovata* DSM-2662 and *S. ovata* DSM-3300, *S. ovata* DSM-2663 had lower coulombic efficiency (Table 2). These results showed that although *S. ovata* DSM-2663 generated acetate faster than the two other strains, it was less efficient at converting electrons from the cathode specifically into acetate molecules.

Images of confocal laser scanning microscopy (CLSM) showed substantial cell attachment by *S. ovata* DSM-2662 and *S. ovata*

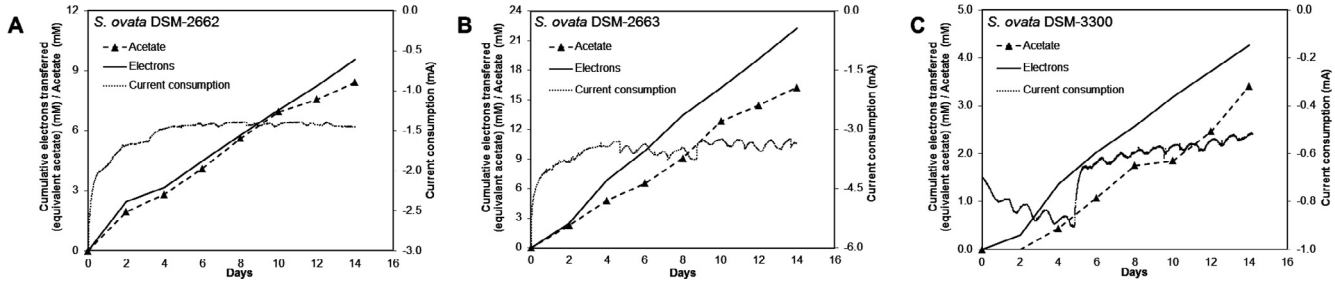

Fig. 1. Acetate production, current consumption and electron transferred during MES with (A) *S. ovata* DSM-2662, (B) *S. ovata* DSM-2663 and (C) *S. ovata* DSM-3300. Electron transferred curves correspond to the acetate concentration in mM if all the electrons transferred were converted to acetate. Acetate production curves in mM correspond to the real progression of acetate concentration in the MES reactor detected by high-pressure liquid chromatography. Results shown are from a representative example of three replicate MES reactors.

Table 2
The average acetate production and current consumption density of MES with different *Sporomusa*.

Strain	Acetate production rate[a]		Current density[a] (mA m^{-2})	Coulombic efficiency[a] (%)
	mmol m^{-2} day^{-1}[b]	mM day^{-1}		
S. ovata DSM-2662	34.3 ± 7.0	0.49 ± 0.10	454.2 ± 150.4	91.8 ± 5.3
S. ovata DSM-2663	61.1 ± 18.1	0.88 ± 0.26	782.5 ± 187.1	61.1 ± 12.6
S. ovata DSM-3300	12.9 ± 5.6	0.19 ± 0.08	191.2 ± 5.2	84.9 ± 4.0
S. acidovorans	44.1 ± 14.2	0.63 ± 0.20	572.1 ± 210.6	69.9 ± 0.9
S. malonica	45.4 ± 4.9	0.65 ± 0.07	703.2 ± 168.4	90.8 ± 13.9
S. aerivorans	N.D.[c]	N.D.	25.6 ± 2.1	N.A.[d]
Sterile control	N.D.	N.D.	16.4 ± 3.0	N.A.

[a] Each value is the mean and standard deviation of three replicates.
[b] Values normalized with respect to the graphite stick cathode projected surface area.
[c] Not detected.
[d] Not applicable.

DSM-2663 at the surface of the graphite cathode. In the case of *S. ovata* DSM-3300, less cell attachment was observed at the surface of the electrode correlating well with its lower MES performance. The observations made here with the three tested *S. ovata* strains indicate that different "wild type" strains of the same acetogenic species do not have similar performance for MES.

3.2. Genome variations between the S. ovata strains

The three *S. ovata* strains tested here were isolated by the same research group from three different sources (Möller et al., 1984). In Möller et al. study, *S. ovata* DSM-2662, *S. ovata* DSM-2663 and *S. ovata* DSM-3300 were shown to have similar growth conditions and physiological characteristics. To ensure that the *S. ovata* cultures used here are really three distinct strains and to evaluate the level of difference between them, the genomes of *S. ovata* DSM-2663 and *S. ovata* DSM-3300 were sequenced and compared to the genome sequence of *S. ovata* DSM-2662 (Poehlein et al., 2013). *S. ovata* DSM-2663 had 69 genome variations including a base substitution in a subunit of a NADP-reducing dehydrogenase (*hndD2*) coupling the reduction of NADP to NADPH with the oxidation of molecular H_2 (Dermoun et al., 2002). This enzyme may be involved in energy conservation during autotrophic growth by *S. ovata*. Additionally, genes coding for regulatory proteins (*baeS*, SOV_1c09220) presented variations with possible effects on the expression of other genes in *S. ovata* DSM-2663, which could be related to the better MES performance of this strain. *S. ovata* DSM-3300 was significantly more distant from both *S. ovata* DSM-2662 and *S. ovata* DSM-2663 with ca. 36800 genome variations in genes coding for proteins with multiple functions such as regulation, H_2 metabolism, transport, energy conservation and CO_2 reduction via the Wood-Ljungdahl pathway. Genome sequencing results presented here confirmed that the studied strains were different genetically, which is probably related to the observed variation in MES performance.

3.3. Growth of S. ovata strains with H_2:CO_2

Below a potential of −590 mV vs SHE, carbonaceous cathodes evolve H_2 abiotically (Aulenta et al., 2008). Furthermore, the presence of microbial cells at the surface of the cathode has been shown to accelerate H_2 evolution (Deutzmann et al., 2015; Jourdin et al., 2016 and May et al., 2016). Therefore, in the MES system employed here with a graphite cathode set at potential of −690 mV vs SHE, it could be hypothesized that any acetogenic species has the potential to be a good candidate for the electrosynthesis of acetate if they are capable of growing efficiently with H_2 as the electron donor and CO_2 as the electron acceptor. To verify this hypothesis, the growth and acetate production of *S. ovata* DSM-2662, DSM-3300, and DSM-2663 was evaluated in batch mode under a H_2:CO_2 atmosphere (Fig. 2 and Table 3). The three strains had similar doubling times and acetate production rates, and thus, no relation could be established between their metabolism efficiency under a H_2:CO_2 atmosphere in batch mode and their MES performance. More specifically, results obtained with *S. ovata* DSM-3300 demonstrate that good fitness with H_2:CO_2 does not translate into good performance in the MES system (Figs. 1C and 2C).

3.4. MES and H_2:CO_2 growth with other Sporomusa species

Until now, all the *Sporomusa* species tested in the MES system were shown to be capable of reducing CO_2 with electrons coming from a cathode (Tremblay and Zhang, 2015). This also includes both *S. silvacetica* and *S. sphaeroides*, which are known to electrosynthesize acetate but significantly slower than *S. ovata* DSM-2662 (Nevin et al., 2010 and Nevin et al., 2011). To establish if

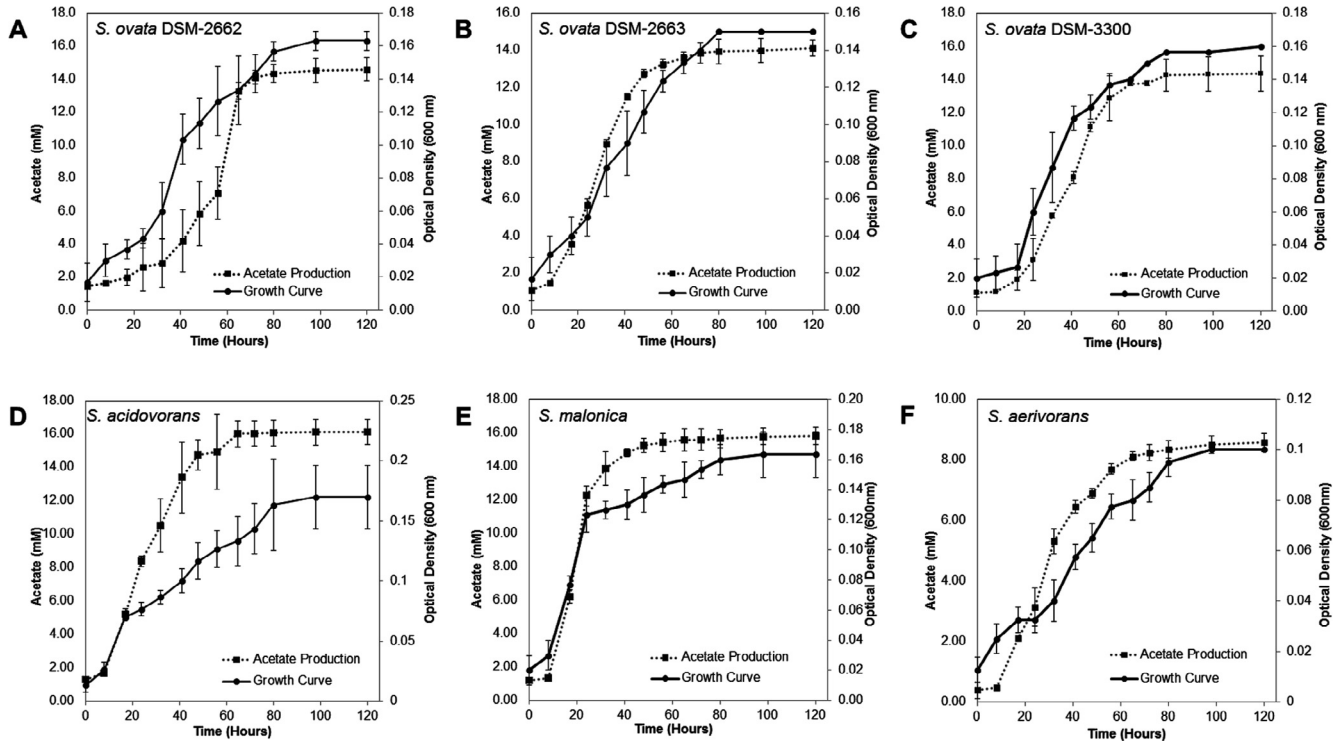

Fig. 2. Growth on H_2:CO_2 of (A) *S. ovata* DSM-2662, (B) *S. ovata* DSM-2663, (C) *S. ovata* DSM-3300, (D) *S. acidovorans*, (E) *S. malonica* and (F) *S. aerivorans*. Each curve is the mean and standard deviation of three replicates.

Table 3
Growth on H_2:CO_2 of different *Sporomusa*.

Strain	Doubling time[a] (Hour)	Acetate production[a] rate (mM day^{-1})
S. ovata DSM-2662	24.8 ± 4.0	6.7 ± 1.3
S. ovata DSM-2663	25.7 ± 2.9	7.2 ± 0.1
S. ovata DSM-3300	25.4 ± 0.7	6.5 ± 0.3
S. acidovorans	27.0 ± 4.4	7.9 ± 1.0
S. malonica	9.7 ± 1.9	10.2 ± 0.3
S. aerivorans	30.2 ± 3.1	3.2 ± 0.1

[a] Each value is the mean and standard deviation of three replicates.

the capacity to perform MES is a widespread characteristic within the *Sporomusa* genus, three other *Sporomusa* species including *S. acidovorans*, *S. malonica*, and *S. aerivorans* were tested in the MES reactor (Fig 3 and Table 2). *S. acidovorans* and *S. malonica* were drawing current and producing acetate from CO_2 at levels comparable to what has been observed with *S. ovata* DSM-2662 and *S. ovata* DSM-2663 (Fig. 3A and B). However, *S. acidovorans* had significantly lower coulombic efficiency than *S. malonica* and *S. ovata* DSM-2662. *S. aerivorans* did not electrosynthesize acetate from CO_2 and the observed current density was of the same order of magnitude as the abiotic control MES system (Fig. 3C). CLSM images showed substantial cell attachment of both *S. acidovorans* and *S. malonica* on the graphite cathode during MES.

Growth and acetate production of *S. acidovorans*, *S. malonica*, and *S. aerivorans* were also evaluated in batch mode under a H_2:CO_2 atmosphere (Fig. 2 and Table 3). *S. acidovorans* had a doubling time and an acetate production rate comparable to the three strains of *S. ovata*, whereas *S. malonica* exhibited the best growth performance among all the tested strains (Fig. 2 and Table 3). Compared to *S. ovata* DSM-2663, *S. malonica* was slightly less performant for MES but it grew 2.6 times faster (p-value of 0.001) and had an acetate production rate 1.4 times higher (p-value of 0.0001) under a H_2:CO_2 atmosphere (Fig. 2E and Table 3). This indicates that a more efficient metabolism with H_2:CO_2 does not necessarily translate into better MES performance. On the other hand, *S. aerivorans* was unable to perform MES but it was still

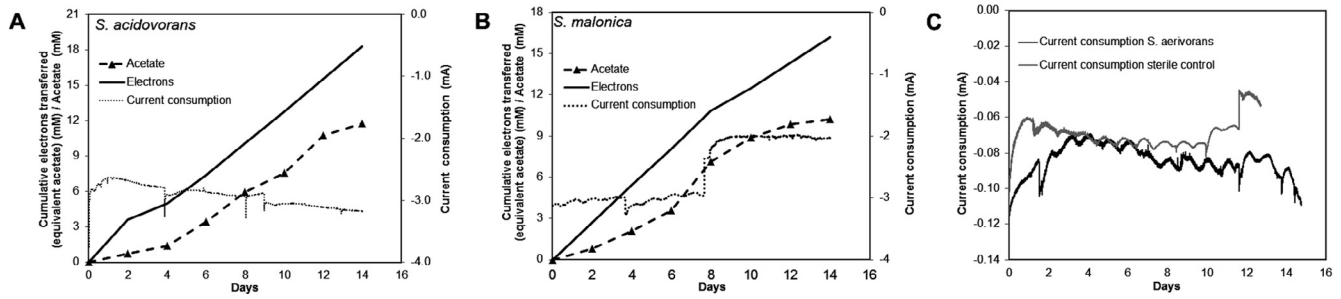

Fig. 3. Acetate production, current consumption and electron transferred during MES with (A) *S. acidovorans* and (B) *S. malonica*. (C) Current with *S. aerivorans* and the sterile medium control. No acetate was produced with *S. aerivorans* or the sterile medium control. Electron transferred curves correspond to the acetate concentration in mM if all the electrons transferred were converted to acetate. Acetate production curves in mM correspond to the real progression of acetate concentration in the MES reactor detected by high-pressure liquid chromatography. Results shown are from a representative example of three replicate MES reactors.

metabolically active under a H_2:CO_2 atmosphere and capable of generating biomass despite its significantly slower growth and acetate production than the other tested *Sporomusa* species (Fig. 2F and Table 3). These observations showed that the capacity of oxidizing H_2 for the reduction of CO_2 *via* the Wood-Ljungdahl pathway is not sufficient for an acetogenic species to be able to drive MES.

This study strongly suggested that acetogens performing well in MES reactors must have other characteristics than the ability to oxidize H_2 efficiently for the biological reduction of CO_2. For example, performant acetogenic MES microbial catalyst may 1) be more efficient at forming biofilm on electrode surface to increase the volume of electrons exchange, 2) have the capacity to accelerate the cathode-mediated H_2 evolution, 3) have uptake hydrogenases more suitable for H_2 oxidation in closed proximity to a cathode, or 4) be capable of acquiring electrons either directly or *via* alternative electron shuttles (Deutzmann et al., 2015; Jourdin et al., 2016; Patil et al., 2015; Tremblay et al., 2016).

In this work, MES acetate production rates for *Sporomusa* strains including *S. ovata* DSM-2663 were at least one order of magnitude slower than for H_2:CO_2 grown cultures (Tables 2 and 3). Previous studies have shown that using graphene-modified cathodes instead of the graphite cathode is sufficient to increase acetate production by *Sporomusa*-driven MES by one order of magnitude (Aryal et al., 2016; Chen et al., 2016). Further research is required to establish if employing *S. ovata* DSM-2663 in combination with graphene-modified cathodes or with other bioelectrochemical reactor improvements could have a synergistic effect on MES performance.

4. Conclusion

Among the tested *Sporomusa*, only *S. aerivorans* could not perform MES. This result demonstrates that the capacity of catalyzing MES is not conserved among the *Sporomusa* genus. *S. ovata* DSM-3300 catalyzed MES, but not as efficiently as other *Sporomusa*. Under a H_2:CO_2 atmosphere, *S. ovata* DSM-3300 reduced CO_2 similarly to the better MES microbial catalysts indicating that there is no correlation between fitness on H_2:CO_2 and MES performance. *S. ovata* DSM-2663 was the best MES microbial catalyst in this study. Although it had lower coulombic efficiency, *S. ovata* DSM-2663 could be a promising microbial catalyst for MES applications.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.biortech.2017.02.128.

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