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Photocatalytic inactivation of *Clostridium perfringens* spores on TiO_2 electrodes

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Abstract

Disinfection of drinking water is commonly carried out by chlorination, however research has shown this method to be ineffective against certain protozoan, viral and biofilm forming microorganisms. Furthermore, chlorination can result in the formation of mutagenic disinfection by-products. Semiconductor photocatalysis may be a possible alternative to chlorination for point-of-use drinking water disinfection. In this work TiO_2 electrodes were fabricated using electrophoretic immobilisation of commercially available TiO_2 powders onto conducting supports, i.e. indium-doped tin oxide-coated glass, titanium metal and titanium alloy. Photocatalytic inactivation of *Escherichia coli* and *Clostridium perfringens* spores in water was demonstrated on all immobilised TiO_2 films. The rate of photocatalytic inactivation of *E. coli* was one order of magnitude greater than that of *C. perfringens* spores. The application of an external electrical bias significantly increased the rate of photocatalytic disinfection of *C. perfringens* spores. The effect of incident light intensity and initial spore loading were investigated and disinfection kinetics determined as pseudo-first order.

Keywords: Photocatalysis; Titanium dioxide; Disinfection; *Escherichia coli*; *Clostridium perfringens*; Spores; Photo-electrochemistry

1. Introduction

In the past decade outbreaks of waterborne gastro-enteritis have increased across the developed world with incidences attributed to both drinking and recreational waters [1]. Chemical disinfection (e.g. chlorination and ozonation) is commonly used to protect potable water against microbial contamination, however, the efficacy of conventional disinfection strategies against certain protozoan, viral and biofilm forming microorganisms, has been questioned [2]. Furthermore, chlorination can result in the formation of disinfection by-products, many of which have mutagenic properties [3].

Advanced oxidation technologies are potential alternative and/or complementary water purification methods [4]. UV-semiconductor photocatalysis is an AOP which operates under ambient temperatures and pressures. Titanium dioxide (TiO_2) is the photocatalyst of choice for water purification applications as it is non-soluble, non-toxic and photochemically stable. When TiO_2 is irradiated with UV light ($\lambda < 400$ nm), an electron is promoted from the valence band to the conduction band, leaving a positive hole in the valence band. The holes can migrate to the particle–solution interface where they can oxidise hydroxyl ions or water to form hydroxyl radicals. In order to maintain electrical neutrality, the photogenerated electrons must be removed from the conduction band. Dissolved oxygen is a readily available electron acceptor, but, under conditions, e.g. high-organic loading, high-light intensity or low DO, this reduction step may be rate limiting [5]. Reduction of dissolved oxygen generates additional reactive oxygen species including superoxide radical anion ($O_2^{\bullet -}$), hydroperoxyl radical ($HO_2^{\bullet}$) and hydrogen peroxide (H_2O_2). These species can feed into the radical attack mechanisms responsible for the oxidation of organic and inorganic species present in the water. Photocatalysis has been reported to completely mineralise organic pollutants to carbon dioxide, water and mineral acids (treatment time-dependent) [4]. Furthermore, photocatalysis has been reported to degrade problematic drinking water pollutants including endocrine disrupting chemicals [6] and pharmaceutical residues [7].

The photocatalytic inactivation of microorganisms in water has been widely reported. Bacterial organisms studies include

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indicator organisms, *Escherichia coli* [8] and coliforms [9], and a wide range of both Gram-positive and Gram-negative species [10]. The photocatalytic inactivation of viruses [11,12] and protozoan organisms [13,14] has also been reported. Given *E. coli*'s role as an indicator organism in water quality, it has been widely used in photocatalytic disinfection experiments. Reactor systems using slurry and immobilised TiO_2, with *E. coli* cells suspended in distilled water, tap water, buffer solutions and river/stream water have been reported, and recently reviewed [15]. Bacterial cells have, however been described as a rather easy target for disinfectants, with bacterial spores and protozoa suggested as more robust target organisms. *Clostridium perfringens* spores have been reported to be chlorine resistant at levels used in potable water supplies [16].

The mechanism of photocatalytic disinfection is still not fully understood [17–20] however, hydroxyl radical-mediated lipid peroxidation of the outer cell wall components has been proposed as a key process [20].

Photocatalytic reactors using immobilised catalysts have a number of practical and financial advantages in comparison to slurry-based systems. Immobilisation of TiO_2 powder onto a conducting support produces a microporous, nanocrystalline electrode [21]. However, the decrease in catalyst surface area requires strategies to improve the mass transfer of pollutants to the catalyst surface. Electrochemically assisted photocatalysis (EAP), utilises the application of a small potential bias to a TiO_2 electrode and has been reported to increase the rate of photocatalytic degradation of chemical and microbiological pollutants [8,22]. EAP decreases electron–hole recombination and physically separates charge carrier redox sites by removing photogenerated electrons to the counter electrode [22].

In this work TiO_2 electrodes were used to investigate the photocatalytic and the EAP disinfection of *E. coli* cells and chlorine resistant *C. perfringens* spores in water.

2. Experimental

2.1. Photocatalytic disinfection

Immobilised TiO_2 electrodes were prepared using the electrophoretic deposition method described by Byrne et al. [23]. TiO_2 powder, *Degussa P25* (80% anatase, 20% rutile, primary particle size 30 nm) and *Aldrich* (Cat. No. 232033, 99% anatase, primary particle size <1 μm) were suspended in methanol (1%, w/v) and coated onto 2 cm × 1 cm supporting substrates by the application of 25 V vs. a stainless steel counter electrode. Electrically conducting substrates used included Ti–4Al–6V alloy (Shorts, Belfast), Ti foil (Aldrich) and indium-doped tin oxide-coated (ITO) borosilicate glass (Instrument Glasses/Donnely Co.). The average weight of TiO_2 deposited onto each substrate was determined by gravimetric analysis to be $1.0 \pm 0.2\ mg\ cm^{-2}$. The coated substrates were annealed at 500 °C in air for 1 h. Electrical contact was made using a copper wire and conducting expoxy (Circuit Works). Any area not coated with TiO_2 was insulated using negative photoresist (Casio Chemicals).

In a typical experiment a TiO_2 electrode was positioned in the centre of a quartz water-jacketed reactor [8] and a Philips 125 W HPR lamp used as the irradiation source (mainline emission 360 nm). The UVA incident on the reactor was determined to be 5.8×10^{-8} Einstein $s^{-1}\ cm^{-2}$ using chemical actinometry [24]. Irradiation of the ITO glass films was possible in two orientations. Light directed onto the TiO_2-coated side is described as front-face irradiation and irradiation of the TiO_2 layer through the glass support is referred to as back-face irradiation. The reactor was thermostatically controlled at 20 ± 2 °C and agitation of the bacterial suspensions was provided by a magnetic flea with stirrer. The solution was air-sparged using an aquarium pump at a flow rate of $900\ cm^3\ min^{-1}$. Oxygen-free nitrogen (OFN) sparging, at a flow rate of $900\ cm^3\ min^{-1}$, was used to remove dissolved oxygen. The bacterial suspension ($3\ cm^3$ of 3×10^4 *C. perfringens* spores cm^{-3}) was placed in the reactor and gas-sparged in the dark for 15 min. A 10 μl sample was removed at $t = 0$ min, the electrode was irradiated and samples were removed every 15 min thereafter for a period of 120 min. Disinfection experiments were carried out in triplicate.

2.2. Electrochemically assisted photocatalysis

To investigate the effect of EAP the quartz-jacketed reactor was used as a one-compartment photo-electrochemical cell. The TiO_2 electrode acted as the working electrode (WE), the counter electrode (CE) was a platinum rod (diameter 5 mm, length 30 mm) and the reference electrode, a miniature saturated calomel electrode (SCE) (ThermoRussell). Potentiometric control was achieved using an electrochemical workstation (Sycopel AEW 2) with PC control. Photocurrent was recorded against time for the duration of the experiments. Experiments were carried out using the *Degussa*-Ti foil and *Aldrich*-Ti foil electrodes under open circuit, short circuit and under external bias at +1.0 V (SCE). Disinfection experiments were carried out in triplicate.

2.3. C. perfringens growth, sporulation and detection

C. perfringens (NCTC 8239) was cultured overnight in cooked meat media (Oxoid) at 37 °C. The *C. perfringens* cells were precultured in fluid thioglycolate media (Difco), incubated overnight at 37 °C, and a 5% inoculum transferred to freshly prepared Sacks and Thompson sporulation media [25]. The sporulation media was modified by a threefold increase in the concentration of dextrin and the addition of $50\ \mu g\ cm^{-3}$ of theophylline [25]. Incubation for 56 h at 37 °C produced a ratio of 95% spores to cells, determined by phase contrast microscopy. The spores were harvested by centrifugation (10,000 × *g*, 4 °C, 15 min), washed three times and stored in sterile distilled water at 4 °C. The spores showed no reduction in number, or ratio of cells to spores, over a period of 3 months. Appropriate dilutions were prepared and stored as above.

C. perfringens spores were detected using membrane *C. perfringens* (mCP) agar [26], as modified by Armon and Payment [27]. mCP agar is listed by the European Union as the agar of choice for the detection of *C. perfringens* spores in water. A spread plate technique was used rather than membrane filtration

due to the methods simplicity. Samples taken from the reactor were diluted appropriately in 1/4 strength Ringer's solution, plated in triplicate and incubated under anaerobic conditions at 44 °C for 18–24 h. Colony-forming units (CFUs) were visually identified and reported as average CFU cm^{-3}. The limit of detection for this method was calculated to be 100 CFU cm^{-3}.

2.4. E. coli growth and detection

E. coli K12 was aerobically subcultured overnight from stock suspension in Luria Bertani (LB) medium as previously described [8]. The cells were harvested by centrifugation, washed three times in 1/4 strength Ringer's solution and resuspended in distilled water at an initial cell concentration of $\sim 7.5 \times 10^6$ cells cm^{-3}. Samples removed from the photocatalytic reactor were diluted appropriately in Ringer's solution and plated in triplicate onto LB agar, incubated for 24 ± 4 h at 37 °C. Colony-forming units (CFUs) were visually identified and reported as average CFU cm^{-3}. The limit of detection for this method was calculated to be 100 CFU cm^{-3}.

2.5. Data analysis

Data points represent the average number of colonies counted on triplicate agar plates. The variance between repeat experiments was determined by standard error calculation, represented by error bars in the figures. The significance of the inactivation trends between experiments (i.e. between non-treatment control and treatment experiments) was assessed using the Mann–Whitney *U*-test.

3. Results and discussion

3.1. Photocatalytic inactivation

Photocatalytic inactivation of *C. perfringens* spores was observed using *Degussa P25* TiO_2 immobilised on Ti alloy, Ti foil and ITO-coated glass, and *Aldrich* TiO_2 immobilised onto Ti foil (Fig. 1). The *Degussa*-Ti alloy electrode showed the greatest percentage inactivation with 99.7% of the initial spore count inactivated with 120 min irradiation. No significant difference was observed between front-face and back-face irradiation of the *Degussa*-ITO glass film ($p > 0.1$) or between *Degussa*-Ti foil and *Aldrich*-Ti foil electrodes ($p > 0.1$).

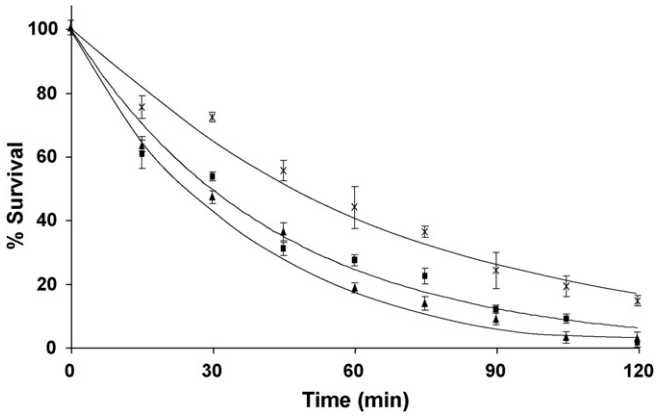

Fig. 1. *C. perfringens* spores survival vs. photocatalytic treatment time. *Aldrich*-Ti foil (×), *Degussa*-ITO (back-face) (■), and *Degussa*-Ti alloy (▲.)

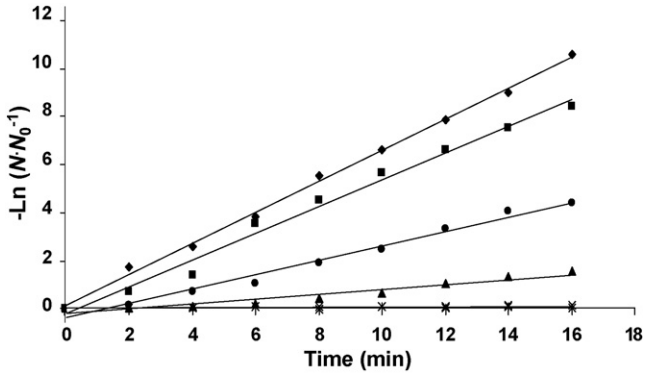

Fig. 2. First order rate plot for the photocatalytic disinfection of *E. coli*. *Degussa*-ITO (front-face) (♦), *Degussa*-ITO (back-face) (■), UV + air sparging (●), TiO_2 electrode only (▲), TiO_2 electrode only (×), and no treatment (*).

The rate of photocatalytic inactivation of *C. perfringens* spores was calculated using Chick's law, with $-\mathrm{Ln}(N \cdot N_0^{-1})$ plotted as a function of time; where N is the number of bacterial cells at a given time and N_0 is the initial bacterial cell count. The first order rate constant, k, was calculated per unit area of TiO_2 with *Degussa P25* showing higher efficiency compared to *Aldrich* (anatase) (Table 1).

The photocatalytic inactivation of *E. coli* was also investigated under identical conditions to those used for *C. perfringens* disinfection using a *Degussa*-ITO glass electrode irradiated in back-face and front-face configurations (Fig. 2). No significant difference was observed ($p > 0.1$) between front-face and back-

Table 1
Disinfection rate constants for the photocatalytic disinfection of *C. perfringens* spores and *E. coli.*

Electrode	*C. perfringens* inactivation rate, $k \times 10^2$ (min^{-1} cm^{-2})	*E. coli* inactivation rate, $k \times 10^1$ (min^{-1} cm^{-2})
Degussa-Ti alloy	2.12	[a]
Degussa-ITO glass (front-face)	1.95	3.87
Degussa-ITO glass (back-face)	1.62	3.40
Degussa-Ti foil	1.32	[a]
Aldrich-Ti foil	1.07	[a]

Experimental conditions: UV intensity 5.8×10^{-8} Einstein s^{-1} cm^{-2}, air sparging at 900 cm^3 min^{-1}, *C. perfringens* spore loading 3×10^4 spores cm^{-3}, *E. coli* cell loading 7.5×10^6 cells cm^{-3}.

[a] Experiment not undertaken.

face irradiation (99.9% and 99.6% *E. coli* inactivation within 10 min irradiation respectively) and the rate constants calculated by Chick's law are given in Table 1. The inactivation rate for *E. coli* was approximately one order of magnitude greater than that observed for *C. perfringens* spores.

E. coli has been described as an "easy target" for assessment of new/novel disinfection systems [28,29] however, the disinfection of chlorine resistant bacterial spores provides a more robust test model. The photocatalytic inactivation of *Bacillus pumilus*, *Bacillus stearothermophilus* spores [30–32] and *C. perfringens* [33] have been reported. However, in later study the authors failed to report a sporulation stage, which is required to induce the production of the disinfection resistant spores [34].

The mechanism of photocatalytic bacterial inactivation is thought to proceed mainly via hydroxyl radical attack [4,10]. The hydroxyl radical is a powerful indiscriminate oxidising agent and has been shown to cause damage to biological structures and systems [35]. Blake et al. reported that the direct attack by hydroxyl radicals is only partly responsible for the disinfection of the bacterial spores. Superoxide radical anion, hydroperoxyl radical and hydrogen peroxide, formed by the reduction of dissolved oxygen, can also feed into the photocatalytic disinfection mechanism [10]. Hydrogen peroxide, although less reactive than the hydroxyl radical, can enter the bacterial structure and be activated by ferrous iron via the Fenton reaction. Fe^{2+} is incorporated into the outer coating of bacterial spores and cells for use in superoxide dismutases and protein structures. Therefore, H_2O_2 produced by the photocatalytic reaction may feed into an *in vivo* Fenton reaction resulting in OH radical formation within the target organism. When both hydrogen peroxide and superoxide radical anion are present, the iron-catalysed Haber–Weiss reaction (Eq. (1)) could provide a secondary pathway to form additional hydroxyl radicals [36].

$$H_2O_2 + O_2^- \rightarrow O_2 + OH^- + OH^\bullet \quad (1)$$

The pseudo-first order inactivation rate constants determined, confirm that the bacterial spores are more resistant to photocatalytic disinfection than bacterial cells. *C. perfringens* spores have a greater ability to endure oxidative attack due to a dipicolinic acid–calcium–peptidoglycan complex (~10–20% of core dry weight) within the outer spore coating [37]. This complex is associated with resistance to chemical and physical disinfectants through a lowering of the core water content and protection of the spore DNA from UV and radical damage [38].

Other photocatalytic disinfection studies have reported the use of TiO_2 immobilised onto glass substrates, e.g. soda glass plates coated with a sol–gel film have been used for the photocatalytic disinfection of *E. coli* cells and endotoxin in water [39,40], the disinfection of *Lactobacillus casei* phage PL-1 [11] and the photocatalytic disinfection of air [41]. Belhacova et al. [42] studied the inactivation of *E. coli* and the bacteriophage λNM1149 using front-face illumination of a *Degussa P25*-coated glass plate. Disinfection of 8×10^4 CFU cm^{-3} was achieved following 6 h illumination however, due to the flow rate bacterial exposure was limited to a total of 9 min. It is difficult to compare results with this study due to differences in reactor geometries, incident light intensities and UV source spectral emission.

No significant difference was observed between disinfection experiments using the ITO glass electrode in back-face or front-face irradiation. The back-face configuration has a number of advantages in that it allows the direct irradiation of the TiO_2 film preventing photon loss due to absorption or scattering by species present in the water. However, under back-face irradiation, controlling the film thickness is critical. With low light intensities and thick TiO_2 films, only the catalyst close to the support is activated and diffusion of the pollutant through the TiO_2 film may be rate limiting. The porous nature of the film, produced using electrophoretic immobilisation of TiO_2 powder, provides a large surface area to geometric area ratio however, this is only effective where small molecules are able to diffuse into the pores to reach the photo-activated surface [43]. Bacteria and spores, with dimensions in the micron scale, will not be able to penetrate into a meso or micro porous film, leaving only the surface layers of the TiO_2 available as the active sites for organism inactivation. ROS generated within the porous film would be required to diffuse out to the surface before reacting with the target organisms. The later would lead to a lowering of quantum efficiency for disinfection due to surface recombination reactions.

3.2. Control experiments for *C. perfringens*

Control experiments were undertaken to assess the contribution of photolytic disinfection, the effect of dark adsorption and air sparging (Fig. 3). The inactivation of *C. perfringens* spores in distilled water was not observed under dark conditions. The effects of UVA irradiation, adsorption of spores onto the TiO_2 electrode and air sparging resulted in a small, but not a significant decrease in the number of colony-forming units. The synergistic effect of UV irradiation and air sparging was responsible for ~60% spore inactivation in 120 min. The rate of inactivation under UVA with air sparging was at least an order of magnitude

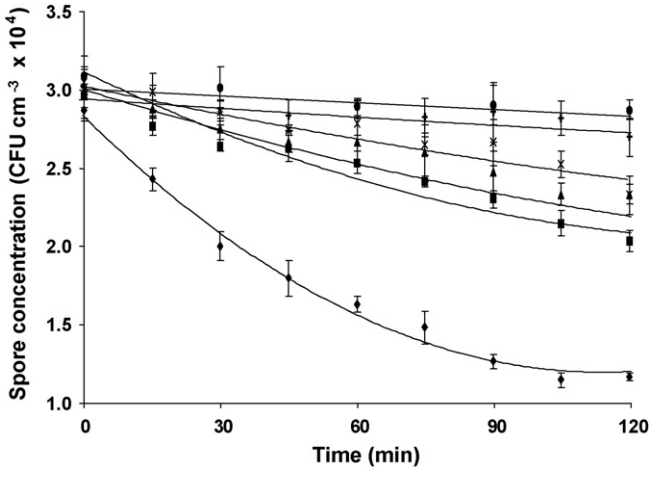

Fig. 3. *C. perfringens* spore count vs. time for control experiments. No treatment (●), TiO_2 electrode only (+), UV only (×), air sparging only (▲), TiO_2 + air sparging (■), UV + air sparging (♦).

Table 2
Disinfection rate constants for the control experiments during photocatalytic disinfection of *C. perfringens* spores and *E. coli*.

Condition	*C. perfringens* inactivation rate, $k \times 10^3$ (min^{-1} cm^{-2})	*E. coli* inactivation rate, $k \times 10^1$ (min^{-1} cm^{-2})
UV + air sparging	7.44	2.96
TiO_2 + air sparging	3.01	0.79
Air sparging only	2.46	[a]
TiO_2 electrode only	0.90	0.06
UV only	1.79	[a]
No treatment	0.50	0.01

Experimental conditions, where appropriate: UV intensity 5.8×10^{-8} Einstein s^{-1} cm^{-2}, air sparging at 900 cm^3 min^{-1}; TiO_2 electrode: *C. perfringens Degussa*-Ti foil; *E. coli Degussa*-ITO glass, *C. perfringens* spore loading 3×10^4 spores cm^{-3}; *E. coli* cell loading 7.5×10^6 cells cm^{-3}.
[a] Experiment not undertaken.

lower than that observed for the photocatalytic inactivation, i.e. UVA with TiO_2 (Table 2).

The control experiments show that photolytic disinfection is effective against *C. perfringens* spores, but slower than photocatalytic disinfection, under the conditions studied. Solar irradiation of water is known to have a bactericidal effect, with bacterial spores showing a greater resistance to treatment than vegetative bacterial cells [44]. Dark controls showed no significant reduction in the spore concentration after 2 h confirming that dark adsorption did not play a significant role in the reduction of colony-forming units.

3.3. The effect of initial spore loading

Photocatalytic experiments were carried out using a *Degussa*-Ti alloy electrode and an initial spore concentration in the range of 1×10^4 to 2×10^5 spores cm^{-3}. As expected the photocatalytic treatment time required for complete spore inactivation increased with higher initial spore loadings, i.e. pseudo-first order kinetics were observed (Fig. 4).

Although it is difficult to directly compare data obtained from various photocatalytic reactors, a (pseudo) first order kinetic model is generally observed in respect of increased initial bacterial loading in slurry reactors [29,42,45]. First order disinfection kinetics using immobilised TiO_2 films under EAP conditions have also been previously reported [8,46].

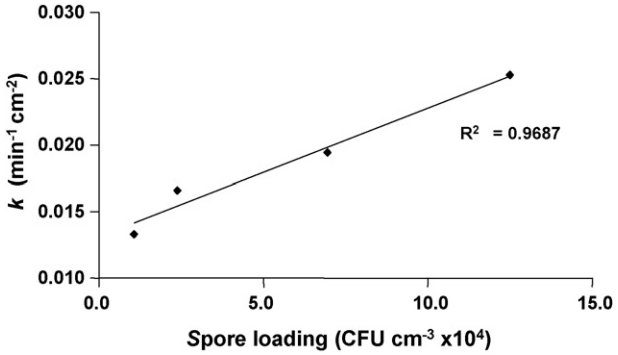

Fig. 4. Plot of pseudo-first order rate constants vs. spore loading.

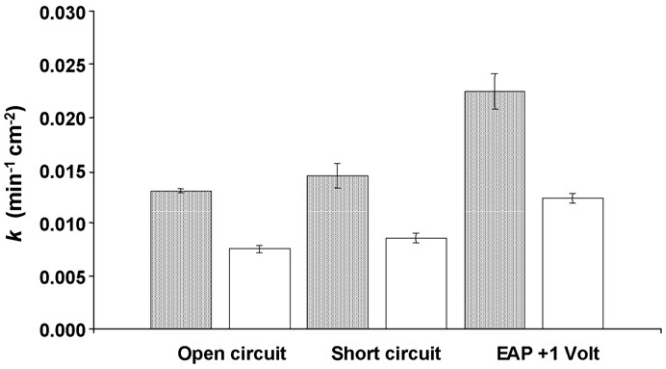

Fig. 5. Rate constant for *C. perfringens* spore inactivation for photocatalysis and EAP treatment. *Degussa*-Ti foil (▨), *Aldrich*-Ti foil (□).

3.4. Electrochemically assisted photocatalytic (EAP) disinfection

EAP increased the rate of inactivation in comparison to open circuit conditions (WE and CE not connected) and short circuit conditions (WE connected to CE without electrical bias) (Fig. 5). The rate of EAP disinfection increased by 72% using *Degussa* electrodes and by 56% using *Aldrich* electrodes.

The application of a positive bias to the TiO_2 electrode lowers the *Fermi* level of the contact electrode therefore increasing the rate of electron transfer from the particulate film to the contact electrode. Transfer of electrons to the counter electrode reduces electron–hole recombination by physically separating the charge carrier redox sites [47]. Additionally EAP increases mass transfer by electro-migration of negatively charged bacteria towards the TiO_2 electrode. EAP has previously been shown to increase the degradation rate of organic compounds [48,49] and microbial pollutants [8,22,33] in comparison to photocatalytic treatment.

The difference in EAP disinfection rate using Aldrich (100% anatase) and Degussa P25 films (20% anatase, 80% rutile) is thought to result from differing spatial separation of electrons and holes in mixed phase films and the subsequent increase in the charge carrier lifetime and or recombination within the thin films. Electron paramagnetic resonance studies of Degussa P25 have shown that the particles contain a small rutile core surrounded by anatase crystallites. Given this structure, the high photocatalytic efficiency has been ascribed to catalytic "hot spots" at the intersection of the two phases due to the formation of unique surface chemistry [50]. Further work is required to elucidate the importance of these characteristics under electrochemical bias.

3.5. Photocurrent response of Ti foil electrodes

The steady-state photocurrent for both *Degussa* and *Aldrich* electrodes was measured during the disinfection experiments. Short circuit photocurrent was higher for *Degussa* electrodes than *Aldrich* electrodes, 11.0 and 3.0 μA cm^{-2}, respectively and photocurrent increased to 83.0 and 28.0 μA cm^{-2}, respectively during EAP.

The relatively small photocurrents observed are due to photocurrent quenching by dissolved oxygen and surface recom-

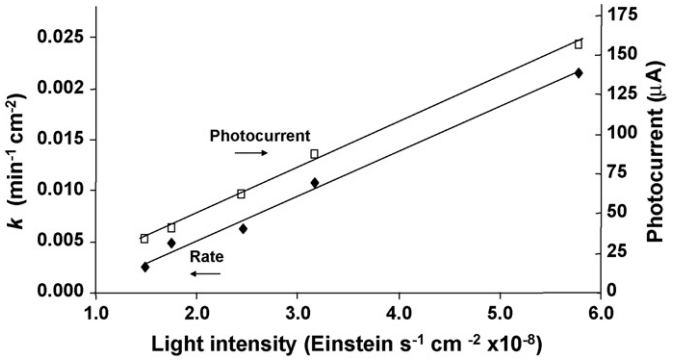

Fig. 6. Rate constant for *C. perfringens* spore inactivation and photocurrent as a function of light intensity. Photocurrent (□) and inactivation rate (♦).

bination reactions. Previous studies using electrophoretically coated electrodes have shown similar photocurrent densities in 0.5 M KCl with the absence of effective hole scavengers [21].

3.6. The effect of incident light intensity during EAP

Both the disinfection rate constant (k) and the photocurrent density were found to be directly proportional to the incident light intensity (Fig. 6). We have previously reported a similar trend during the PC and EAP disinfection of *E. coli* using immobilised *Degussa P25* electrodes [8]. A log plot of k as a function of incident light intensity (I) confirmed the commonly observed trend where rate $\propto I^1$ at low light intensities [51].

The concurrent and parallel increase in the photocurrent response and disinfection rate suggests that a measure of photocurrent may be used as an indirect method to predict the disinfection rate, under strictly controlled conditions. Previously, the photocatalytic degradation rate of formic acid was reported to be directly proportional to the photocurrent under air-sparged conditions [49]. A decrease in the photocurrent response was correlated with a decrease in the concentration of pollutant as a function of treatment time. As concentration of the hole-acceptor became the rate-limiting step, charge carrier recombination increased and the photocurrent reduced. In EAP disinfection experiments a decrease in photocurrent response would indicate mineralisation of the bacterial pollutant. This was not observed during the time-scale of these experiments.

With respect to practical application of photocatalytic technology using solar irradiation, Sichel et al. show that a minimum solar dose is required for the inactivation of *E. coli*, *Fusarium solani* and *Fusarium anthophilum*. Once this dose has been received, the photocatalytic disinfection efficacy is not enhanced by a further increase in solar flux. Solar disinfection (no photocatalyst) was shown to be more susceptible to changes in solar irradiation with disinfection only observed at higher irradiation intensities [52].

4. Conclusion

The photocatalytic disinfection of water containing *C. perfringens* spores was investigated using *Degussa* and *Aldrich* TiO_2 films. 99.7% inactivation of 3×10^4 spores cm^{-3} was achieved in 2 h using a *Degussa*-Ti alloy electrode. Analysis of pseudo-first order rate constants showed higher photocatalytic activity for *Degussa* TiO_2 immobilised onto Ti alloy in comparison Ti foil and ITO glass as the supporting substrates. *Degussa P25* is well known to be an excellent photocatalyst, with the high photocatalytic efficiency ascribed to "hot spots" at the intersection of the anatase–rutile phases present within the P25 particles.

No significant difference was observed ($p > 0.1$) for *E. coli* disinfection using front-face and back-face irradiation of a *Degussa*-ITO glass electrode (99.9% and 99.6% inactivation within 10 min irradiation, respectively).

The photocatalytic treatment time required for complete spore inactivation increased with higher initial spore loadings, i.e. pseudo-first order kinetics. The rate of inactivation was approximately one order of magnitude greater for *E. coli*.

Electrochemically assisted photocatalysis (WE biased at +1.0 V (SCE)) significantly increased the rate of disinfection of *Degussa* and *Aldrich* TiO_2 films on Ti foil (72% and 56% respectively, as compared to open circuit conditions).

The inactivation of the *C. perfringens* spores was found to be linearly proportional to the incident light intensity (within the range investigated).

EAP is an energy efficient, chemical-free and inexpensive method which gives rise to enhanced photocatalytic disinfection on immobilised photocatalysts. Electrochemical processes are scalable and as a result are commonly used in industry. We are currently investigating larger volume EAP reactors for the disinfection of water.

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