Linking discolouration modelling and biofilm behaviour within drinking water distribution systems

S. Husband, K. E. Fish, I. Douterelo and J. Boxall

ABSTRACT

High quality drinking water exits modern treatment works, yet water quality degradation such as discolouration continues to occur within drinking water distribution systems (DWDS). Discolouration is observed globally, suggesting a common process despite variations in source, treatment, disinfection and network configurations. The primary cause of discolouration has been identified as mobilisation of particulate material from pipe walls and the verified Prediction of Discolouration in Distribution Systems (PODDS) model uses measurable network hydraulics to simulate this response. In this paper the cohesive properties of discolouration material are explored and it is hypothesised that in simulating the turbidity response, the PODDS model is actually describing the development and cohesive strength behaviour of biofilms. Applying this concept can therefore facilitate a rapid and simple assessment of DWDS biofilm activity. A review of the findings from PODDS studies conducted internationally is presented, focussing on the macro or observable aspects of discolouration. These are compared and contrasted with associated biofilm studies which consider discolouration material at the micro-scale. Combining the results from these (past) studies to improve the understanding of interactions between microbial ecology and discolouration are discussed with a view to DWDS operational strategies that safeguard and optimise drinking water supply.

Key words | biofilms, discolouration, mobilisation

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INTRODUCTION

Extensive research and development combined with stringent regulations has ensured that consistently high quality water exits modern treatment works. Yet water quality failures, such as discolouration, remain a worldwide phenomenon (Vreeburg & Boxall 2007) and a primary cause of customer dissatisfaction regarding water quality (Prince *et al.* 2003). This water quality degradation must arise within the high-surface area, complex physical, chemical and biological reactors that are drinking water distribution systems (DWDS). Discolouration occurs across networks despite variations in source water, treatment processes, disinfection residuals and distribution configurations (materials, diameter, ages, complexity of layout, etc.). This suggests a common governing process. In the UK, water companies are now accurately simulating and predicting the hydraulically mediated discolouration response of DWDS pipelines using the empirically validated Prediction of Discolouration in Distribution Systems (PODDS) model (Husband & Boxall 2010). PODDS is based upon a 'cohesive layer theory', which proposes that layers of particulate material with a defined profile of shear strength properties accumulate continuously on pipe walls and are conditioned by hydraulic shear stress forces. Although a constant low-level background concentration of particulate material exists in the bulk water (Verberk *et al.* 2006), elevated concentrations that are visible to consumers are considered to be a result of rapid material mobilisation from the accumulated

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layers occurring when the system shear stress exceeds the conditioned state.

This paper explores the hypothesis that biofilms govern the processes of accumulation, composition and ultimately cohesive-strength properties of discolouration-material and that during mobilisation events the PODDS model is effectively describing biofilm erosion. Support for this hypothesis is drawn from research conducted by the authors, including extensive fieldwork studies regarding the world-wide application of PODDS and laboratory studies using a full-scale test facility incorporating detailed and novel microbiological analysis of DWDS biofilms. By acknowledging that biofilms are integral to DWDS material accumulation at the micro-scale, and that the macro-behaviour of discolouration (i.e. turbidity observed throughout a DWDS) can be modelled, holistic network and treatment strategies can be considered and assessed to safeguard the long-term efficient supply of drinking water.

MODELLING DISCOLOURATION: PODDS AND ASSOCIATED FIELD WORK

Water companies have historically controlled/managed discolouration by replacing mains, re-lining pipes, invasive cleaning and/or flushing. All of these are typically expensive and logistically challenging. Figure 1(a) shows a section of an 800 mm trunk-main just 12 months after robust invasive cleaning. Although qualitative, this image shows the presence of material around the entire circumference and indicates a limited short-term benefit of stand-alone cleaning. Three years later, an investigation of the same network reported that discolouration risk was returning to the levels recorded prior to the £25 million cleaning program. This information supports the concept that material accumulation is an ongoing process. Periodic maintenance is therefore required, but optimal frequencies and strategies have yet to be established. By demonstrating that

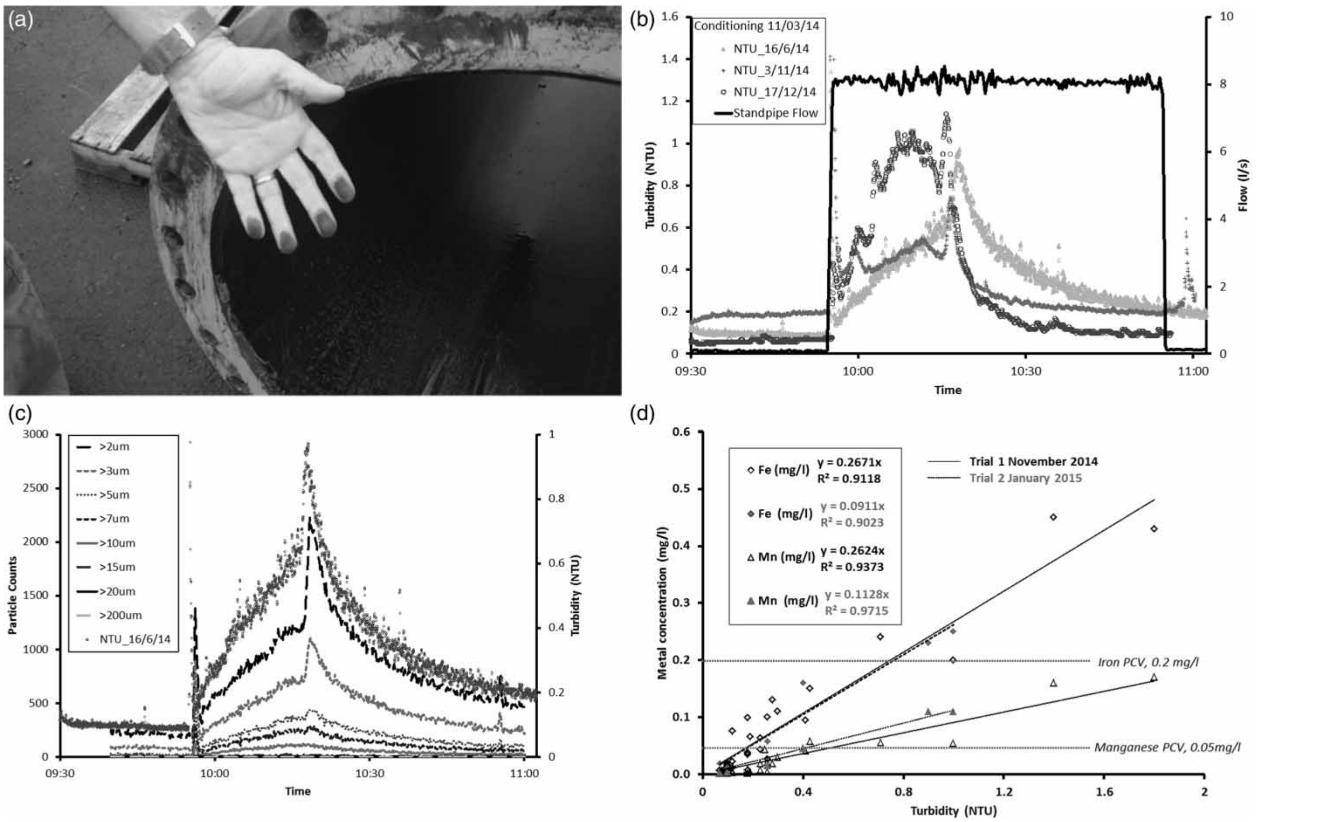

Figure 1 | Results from PODDS trials in trunk mains. (a) Evidence of material regeneration around full pipe circumference in 800 mm main 12 months after invasive cleaning. (b) Turbidity response from the three consecutive 8 l s^{-1} flushing-trials conducted in Study-site 1. (c) Particulate and turbidity response in the first flushing-trial of Study-site 1 – plotted in (b). (d) Turbidity and iron/manganese correlations for the consecutive flushing-trials conducted in Study-site 2.

discolouration is a continuous and ubiquitous issue and providing a tool to simulate and predict discolouration behaviour, PODDS can facilitate justifiable discolouration management strategies. Such strategies typically focus on incremental and low-level releases of accumulated material by controlled and monitored flow increases to raise the hydraulically conditioned state and hence resilience of targeted mains (Cook *et al.* 2015).

The PODDS model is coded as a user function into the one-dimensional hydraulic EPAnet modelling software (Rossman 2000) as a force-balance between cohesive material retention and system mobilising shear stress. Since its publication, over 1,000 field and laboratory tests have been conducted that have verified PODDS as valid to DWDS, primarily in the UK but also internationally including Australia, the Netherlands and Portugal. In brief, these tests generally involve increasing the flow rate above a background peak flow and measuring the turbidity response (metals, inorganics, particle sizes/counts and live/dead cell counts may also be measured). A discolouration response has been observed in every test and, regardless of variation in asset or environmental factors, the calibrated PODDS model parameters have demonstrated a consistency which suggests a process common to all networks in the accumulation and mobilisation of discolouration material.

Results from two previously unreported field studies are shown here to demonstrate the consistent macro-behaviour of discolouration that is observed. Study-site 1 involved three consecutive flushing-trials of a 600 m section of a polyethylene (PE) trunk-main pipe (total length 3 km, internal diameter 251 mm) that had been in operation for 5 years (2009–2014). Each of the three flushing-trials involved increasing flows above the typical peak flow-rate (based on 2014 data) of $15\,\mathrm{l\,s^{-1}}$ (equivalent shear stress of $0.24\,\mathrm{N\,m^{-2}}$) for two turnovers. Flushing flow rates were measured with a Langham hydrant flow meter using ABBs' electromagnetic AquaMaster (15 second logging) and ATi Nephnet portable turbidity monitor (1 second logging, 0.001 NTU resolution) was used to measure discolouration response. PODDS model simulations and an initial conditioning site visit had indicated an $8\,\mathrm{l\,s^{-1}}$ ($0.52\,\mathrm{N\,m^{-2}}$) step increase would remove material from the pipe wall targeting a non-visible 1 NTU turbidity response. The results are shown in Figure 1(b). Targeting a turbidity response below the regulatory 4 NTU (1 NTU was actually targeted to provide a high factor of safety) is necessary to allow the main to remain operational throughout the flushing-trial, a benefit of this non-invasive strategy. A steady rise in turbidity occurs from all trials at Study-site 1 with the peak turbidity at the end of the first pipe turnover (Figure 1(b)). This finding is consistent with observations from PODDS studies and can be explained by the PODDS concept; when the flow is increased, initial weaker layer material is eroded and transported away in the bulk flow, subsequently, material layers with greater shear strength (but still less than the imposed hydraulic force) properties are eroded. The steady rise in turbidity observed can be explained by mobilisation and transport of material distributed evenly along the entire pipe length and not as isolated sediment deposits that would result in significant turbidity fluctuations. With the flow rate known, individual features of the turbidity response can also be identified and assigned to specific pipe sections. In this instance, the turbidity spike 20 minutes after the flow increase was initiated can be associated with a connecting length of exposed iron main exiting the reservoir of Study-site 1. Cast iron mains have repeatedly been shown to have a greater turbidity response, and this is associated with additional accumulation of material due to localised corrosion (Husband & Boxall 2010).

Figure 1(c) presents an analysis of the particles (counts of particles of different sizes, measured using an ATi C10-77 particle counter) and the turbidity from the first of the three flushing-trials from Study-site 1. The data highlight that mobilised material is dominated by particle sizes $<10\,\mu\mathrm{m}$ and that the percentage distribution of particle size is consistent across the duration of the flush. Such consistent particulate size response has been observed irrespective of the applied shear stress; there is no evidence of greater particle size with greater mobilising force. This confirms that self-weight is not dominant in defining cohesive strength. Such small particle sizes result in the requirement for almost quiescent conditions for prolonged periods for the material to deposit, again suggesting self-weight is not a dominant process (Boxall *et al.* 2001). These observations indicate that gravitational (sedimentation) based discolouration models are not valid. Note that the short section of iron main also impacted the particle sizing with a significant increase in numbers of particles (matching the turbidity) supporting the addition of discolouration material resulting from corrosion processes, but not the percentage distribution of particle size (see the peak in Figure 1(c)).

Overall for this first example site, the increases in turbidity and particle counts are evidence that a discolouration risk exists even on a relatively new (<5 years in service) trunk-main pipe. Furthermore, a discolouration response was observed for each of the three repeat flushing-trials, suggesting that material generation between each trial was rapid, occurring even within just a few weeks (Figure 1(c)). In this study, the $8\,l\,s^{-1}$ ($0.52\,N\,m^{-2}$) increase represents a 53% flow (116% force) increase and for increases in force of this magnitude a turbidity response would be expected, even allowing for a short generation period. The reliability in response to successive flushing events indicates that a consistent generation (and mobilisation) process exists. Of note is that for the third flushing trial the background flow was higher than previous trials due to network changes, hence increased mobilisation (NTU) and a quicker turnover time were observed when the same additional demand was added. This change in site conditions is an example of the lack of control and hence variable study conditions typically experienced when investigating operational DWDS. One way to overcome this is to use laboratory studies to investigate discolouration processes. Combining field and laboratory experiments helped confirm the PODDS shear stress and material layer concept (Husband *et al.* 2008).

Sample analysis during flow trials has consistently shown a site-specific correlation of mobilised turbidity and metal concentrations. Figure 1(d) shows the turbidity to iron and manganese concentrations from two consecutive flushing-trials at Study-site 2, a 5 km long, 200 mm diameter mixed unplasticised-PVC and asbestos cement (AC) trunk-main pipe. Regression analysis shows an accurate fit of the linear models (R^2 values >0.9), which indicate repeating and statistically correlating behaviour. It should be noted that Study-site 2 shows a high turbidity to metal association compared to other sites trialled, with 1 NTU exceeding UK regulatory iron and manganese concentrations (during the trials at this site the receiving reservoir was isolated to minimise any customer risk). An investigation into this high correlation is ongoing, with initial findings linking source water quality and a ferric coagulation treatment process. From this it is recognised that discolouration management is not solely the concern of DWDS operators and that treatment processes have a downstream impact. PODDS has increased the understanding of network responses to hydraulic changes and the modelling accuracy has led to cost-effective DWDS maintenance strategies being utilised by the water companies affiliated with the research. Example strategies include trunk-main flushing where water is disposed to waste (no strict flow control required) or conditioning, using controlled flow increases, as demonstrated in Study-site 1, to remove material whilst the main remains in operation. Both these approaches allow network managers to manipulate flows for resilience purposes or discolouration risk mitigation. The incurred costs of these strategies are typically one-off enabling works such that substantive cost benefits extend well into the mid- and long-term asset lifespan. Some example applications and cost savings are listed in Table 1 (from Husband *et al.*

Table 1 | PODDS facilitated operational savings for discolouration management strategies of various UK DWDS pipelines (from Husband *et al.* 2015)

DWDS pipe properties	Proposed strategy	Proposed cost[a]	PODDS strategy	PODDS cost	Savings
4 km 600 to 400 mm mixed	Swabbing	£490 K	Overnight flushing	£227 K	£263 K
7 km 450 mm AC	Swabbing	£530 K	Trunk main conditioning	£150 K	£380 K
6 km 350 mm unlined ductile iron	Main replacement	£2 M	Trunk main conditioning	£40 K	[c]£2 M
10 km 500 mm and a 18″ mixed	Flushing infrastructure	£1.3 M	Trunk main conditioning	£40 K	£1.3 M
4 km 800 mm ductile iron, concrete lined	Jetting	£300 K	Trunk main conditioning	[b]£5 K	£295 K

[a]K = thousand, M = million.
[b]£5 K is water cost, already planned pump refurbishment and control valve installation.
[c]Savings deferred, £40 k due to control valve turbidity monitor installation.

2015), whilst incorporation into 'business as usual' stratagems has prevented discolouration incidents and reduced associated remedial costs.

As an empirical model, PODDS does have deficiencies. Particularly, that it does not specify the material responsible, or the source of the force retaining this material, to the pipe wall. The current PODDS model has been shown to simulate material mobilisation, however trials all demonstrate an ongoing generation which is not captured. Long-term maintenance plans with associated costs and justification of optimal investment strategies cannot therefore be evaluated. Research developing the PODDS model to incorporate both the continual erosion and generation of discolouration material in DWDS is however progressing and being validated (Furnass *et al.* 2014).

DISCOLOURATION AND BIOFILMS

DWDS are now appreciated as being infinitely varied and complex systems with large surface areas that interact with the transported water. Customer perceived 'red' or 'black' water (iron or manganese particles, respectively) has typically led to discolouration being considered as an inorganic issue (Sly *et al.* 1990, Sarin *et al.* 2004). Yet these metals are not always prevalent and even if present, there is no obvious inorganic chemical bonding that could explain the ubiquitous cohesive behaviour identified as governing discolouration. Analysis of treated and distributed water has shown organic matter represents the greatest fraction of suspended solids (Gauthier *et al.* 2001), suggesting a microbial link between DWDS and discolouration.

Support of the force balance and conditioning behaviour demonstrated by the PODDS research is readily evident in microbial research, including shear stress related biofilm mobilisation. The development of biofilms throughout DWDS has been previously established even in the presence of disinfectant residuals (Block *et al.* 1995; Momba *et al.* 1998; Zhang & Lu 2006) and irrespective of surface material (Hallam *et al.* 2001). This can explain why discolouration material is observed ubiquitously. Shear forces, as used to define the PODDS model, have been shown to influence biofilm cohesive strength, with erosion associated with increasing shear stress (Rittmann 1982; Choi & Morgenroth 2003; Abe *et al.* 2012). Commensurate with discolouration particulate behaviour, biofilm detachment due to changes in shear stress has also been shown to be particulate, although sloughing of unstable layers can also occur (Telgmann *et al.* 2004).

Further support implicating biofilms in discolouration comes from laboratory trials by the authors within a temperature controlled, full-scale pipe test facility, with hydraulic controls and water quality monitoring equipment. In brief, the facility (Figure 2(a)) comprises three high-density PE pipe loops (each 200 m in length, 79 mm internal diameter) around which water from the local DWDS is re-circulated with a 24 hour trickle turnover. Specialised PWG coupons (Figure 2(b)) were designed for simultaneous molecular and microscopy-based analyses of biofilm (Dienes *et al.* 2010) and are inserted around the entire circumference of a straight section of the pipe in each loop (Figure 2(a)). After just 28 days of development, significant biofilms have been observed upon the coupon surface (Figure 2(c)–2(f)). Community fingerprinting of these biofilms has shown that the bacterial, fungal and archaeal communities are unaffected by the coupon position around the pipe circumference (Fish *et al.* 2015). In addition to microbial cells, biofilms comprise an extensive matrix of extracellular polymeric substances (EPS, primarily carbohydrates and proteins) which form the greatest part of the biofilm and are accredited with many functions, including structure and stability. The physical characteristics of the biofilms, i.e. the quantity and location of cells and EPS, were also unaffected by coupon position (Fish *et al.* 2015). This further evidences that material-accumulation is not being dominated by gravitational settling/sedimentation, which re-enforces the PODDS concept. Trials studying the bacterial community dynamics during the early stages of development (Douterelo *et al.* 2014c), showed shifts with time and an increase in cell coverage, richness and diversity (with species related to *Pseudomonas* spp. and *Janthinobacterium* spp. dominating initial attachment). Based on fluorescent microscopy fingerprinting results, hydraulic regimes were shown not to significantly affect biofilm bacteriological composition, but did influence mechanical stability. This laboratory work highlights the endemic and constantly generating biofilm community and suggests that controlled hydraulic mobilisation, employed beneficially in operational

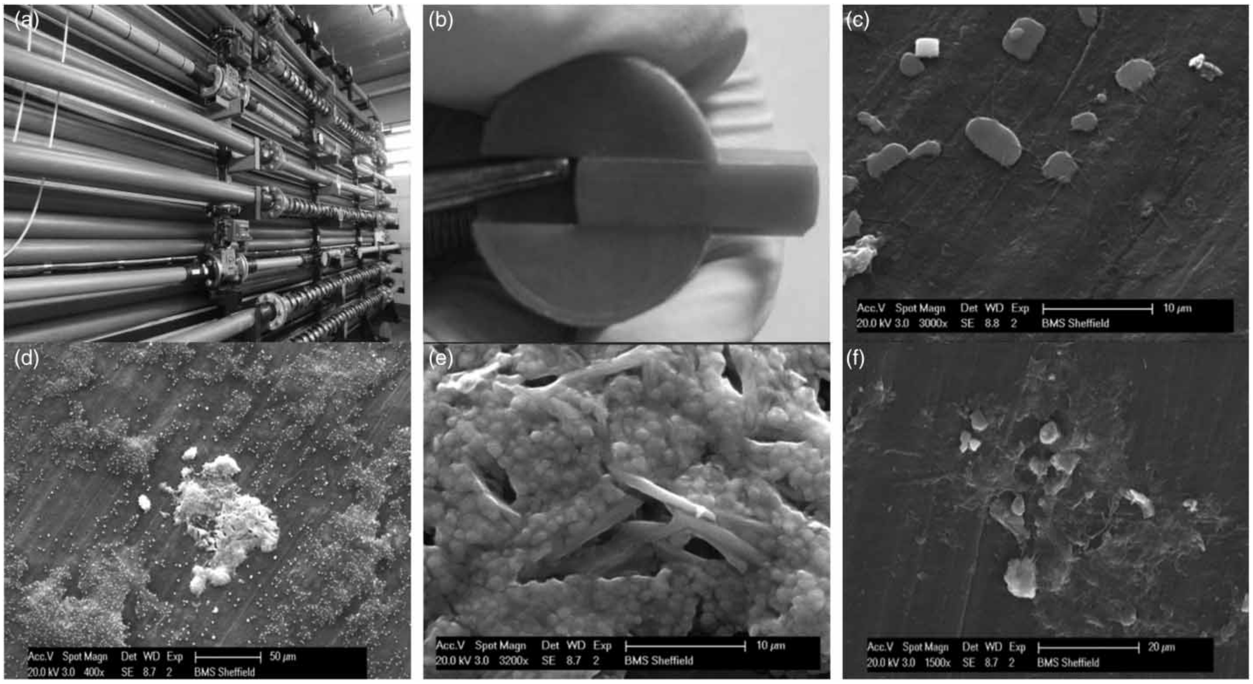

Figure 2 | PODDS laboratory pipe loop, coupon and scanning electron microscopy images of developing biofilm. Microscopy images courtesy of BMS Imaging Facility, University of Sheffield. Scale bars as indicated. (a) 600 m full scale temperature controlled pipe facility, University of Sheffield. (b) Coupon for simultaneous analysis of *in-situ* biofilm physical and community structure. (c) Primary attachment, potentially including inorganic material. (d)–(e) Biofilm after 28 days of development. (f) Qualitative evidence indicative of inorganic incorporation.

networks (see Table 1), is effectively harnessing the mechanical adaptability of biofilms.

Microbial analysis approaches used in the laboratory test facility have also been used to investigate the bacteriological composition of biomass recovered from operational DWDS. Bacterial 454 pyrosequencing has indicated that while source water may dictate overall levels of microbial activity, pipes of different material have distinctly different bacterial communities (Douterelo *et al.* 2014b). High relative abundances of *Alphaproteobacteria*, *Clostridia* and *Actinobacteria* were detected in the material removed from plastic pipes whilst sequences related to *Alphaproteobacteria*, *Bacilli* and *Gammaproteobacteria* were present in the samples obtained from cast iron pipes (Douterelo *et al.* 2014a). Both laboratory and field trials provide initial steps investigating microbial diversity and function, yet PODDS trials indicate a consistency in the erosion mechanics, highlighted by model parameters being transferable between sites. This suggests a common process within biofilms and therefore is perhaps not limited to specific species. Further work however remains to determine if these initial trials are representative of networks worldwide and to examine the dynamics of microbial ecology within these engineered ecosystems.

The PODDS model describes layers of material with the weakest cohesive-strength layer removed first. Stratification in the cohesion of biofilms, with the weakest layers on top, has also previously been measured (Derlon *et al.* 2008). On the micro-scale pipe surface, coverage is not homogeneous, as evident in Figure 2(d). Hence the mobilisation described by PODDS is not realistic at this level. The success of the PODDS model is therefore likely due to the large pipe surface area responding to hydraulic changes with the generalised (macro) behaviour that is observed.

The presence of biofilms also provides possible processes that can explain the accumulation of inorganics at the pipe wall. Biofilms are complex structures with function and integrity derived by EPS, which has been shown to have inorganics associated with it (Laspidou & Rittman 2004). The presence of EPS could also explain how microbially diverse communities can produce the consistent discolouration response observed to hydraulic changes. Hydraulic regimes condition material to have sufficient cohesive strength to resist detachment, and EPS has been shown to govern biofilm adhesive/cohesive strength (Neu & Lawrence 2009). Consequently, it is hypothesised here that the EPS entraps particles that are subjected to radial processes that bring them into contact with the biofilms on boundary surfaces (Van Thienen *et al.* 2011). This is qualitatively supported by Figure 2(f) which appears to show inorganic particles incorporated into the biofilm structure. Results from physical characterisation of the biofilm within the experimental pipe loop showed that the volume of EPS was nearly five-times greater than that of the cells within biofilms, with carbohydrates present as the dominant component (Fish *et al.* 2015). Additionally, the greatest proportion of EPS was located above that of the cells (Fish *et al.* 2015); thus EPS is ideally placed to entrap material from the bulk water and release it when detached, mirroring the discolouration behaviour described by PODDS and helping explain the metal and turbidity correlations shown in Figure 1(d). Chemical analysis also supports inorganic incorporation via EPS, as anionic groups are present that can cause a cation exchange potential, such that metal ions will bind to EPS (Flemming 1995). Furthermore, increased biofilm activity has been associated with increased iron and manganese deposition (Ginige *et al.* 2011). This work suggests that limiting EPS production, in addition to minimising inorganic loading, is therefore likely to be effective in controlling discolouration. Further work is required to investigate different methods to achieve this, for example disinfection to restrict microbial viability and hence EPS production, limiting essential growth nutrients, or other mechanisms that may perhaps limit biofilm development by restricting the proliferation of the key bacterial EPS producers.

FUTURE RESEARCH

With biofilms implicated as the key process governing material accumulation in DWDS, and their subsequent mobilisation resulting in water quality failures, network management strategies can be adapted to account for this behaviour. Given sufficient development time, biofilms may reach a hydraulically conditioned state when generation and mobilisation rates are in equilibrium. At this point, an effective maximum discolouration risk is attained that in UK DWDS has been shown to typically occur after 4 years, although this is dependent on water quality and in non-corroding pipes (Husband & Boxall 2011).

Although primarily an aesthetic risk, discolouration events indicate increased microbiological and metal concentrations that could lead to health concerns. Although there is currently little information, it has been concluded that a likely association exists between turbidity and intestinal diseases (Smith *et al.* 2006, Mann *et al.* 2007). However, establishing the viability of pathogens in DWDS biofilms and the impact of disinfection on microbial diversity and development requires further research. Cell cytometric analysis using recognised protocols (Gatza *et al.* 2013) during PODDS field trials has shown a 10x increase in total cells in an unchlorinated Dutch system compared to UK chlorinated systems, yet the supplied drinking water quality is regarded as amongst the best in the world. In the Netherlands most drinking water is distributed without disinfectant residuals and biofilm regrowth is limited by achieving biostable drinking water at the treatment stage (Van Der Kooij 1999). Improved understanding of the impact of treatment processes (and changes to) on DWDS microbial ecology is therefore needed if optimum service cost and quality is to be established. To achieve this, techniques are needed that can measure effectiveness of maintenance strategies. If it is accepted that PODDS is effectively modelling the cohesive strength behaviour of biofilms, this facilitates simple and easily applied non-disruptive methods by which DWDS biofilm activity can be assessed.

In the UK, discolouration complaints tend to follow annual temperature trends with fewer in the winter when water temperatures typically fall to $<6\,^{\circ}C$ (Cook *et al.* 2015). This suggests network maintenance could be planned to coincide with anticipated higher DWDS biofilm growth rates; in this case during the summer when water temperatures are higher. As part of this, research is required to identify key microbial species (and their international prevalence) in biofilm development. Additionally, function may be investigated thereby facilitating future engineering of these ecosystems. For example, could conditions be produced that favour slow growing or low EPS producing biofilms thereby restricting inorganic incorporation, or, conversely, could fast growing, rapid EPS production but potentially more easily mobilised biofilms be more advantageous.

CONCLUSIONS

Discolouration is a worldwide issue in DWDS. The mobilisation of particulate material from the pipe wall that causes discolouration may be simulated by the PODDS model through consideration of the force balance between the material layer properties and pipeline hydraulics. Research findings highlight biofilms, and specifically the EPS component, as a common feature governing the generation, composition and cohesive strength characteristics of the material layers on pipe walls that cause discolouration. PODDS modelling of the discolouration process is therefore effectively modelling the cohesive strength behaviour of biofilms. Applying PODDS techniques therefore facilitates rapid and simple assessment of DWDS biofilm activity. Combining the growing understanding of the impact of hydraulic conditions and continued research to elucidate microbial diversity and function will lead to improved understanding of biofilm behaviour. This can then help inform future network and treatment strategies to safeguard and optimise drinking water supply.

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