
Introduction

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For more than a century, there has been a strong tradition for studies of microorganisms as planktonic, liquid cultures. This has imprinted the view that bacteria live as unicellular organisms. Although test tube laboratory studies have led to fundamental insights into basic life processes and have unraveled complex intracellular regulatory networks, it is now clear that microbial activity in nature is mainly associated with surfaces and scientists therefore need to turn their attention to studies of the sessile mode of life (Marshall, 1992). Indeed it may be argued that the ability to form surface-associated, structured and cooperative consortia (referred to as biofilms) is one of the most remarkable and ubiquitous characteristics of bacteria (Costerton *et al.*, 1987). From an evolutionary perspective the biofilm lifestyle makes great sense. Sessile consortia of bacteria exhibit increased resistance and elegant adaptive responses to a range of factors that would otherwise negatively impact on their activity.

Studies of microbial biofilms have a long history. Antonie van Leeuwenhoek, in the late eighteenth century, marveled at the diversity of the small life forms he observed in his microscope of biofilm scrapings from teeth surfaces. Early observations on surface related phenomena also included those of swarming by *Proteus* spp. on agar plates used for the detection of pathogenic bacteria (Hauser, 1885). It was seen that the swarming phenomenon allowed the bacteria to effectively colonize the entire surface of the agar plate. In the twentieth century, the development of slimes, or biofilms, on surfaces subjected to water flow was recognized and explored in a variety of habitats and applications. In particular, the effects of biofilms were regularly reported by engineers concerned by alterations in the efficiencies of high-speed vessels, of heat-transfer systems and of flow rates in water reticulation and hydroelectric pipelines. Furthermore, the importance of an increased surface area of microbial populations in the treatment of sewage and industrial wastes was realized at that time. While most studies of biofilms in this regard were carried out by engineers, microbiologists were also conscious of the effects of surfaces on microbial growth in natural habitats, such as soils (Cholodny, 1930) and waters, where the so-called "bottle effect," with water samples in bottles of varying sizes or with added glass beads, resulted in a rapid increase in microbial numbers because of the increased surface area (Henrici, 1933; Heukelekian and Heller, 1940; Stark *et al.*, 1938; ZoBell, 1937, 1943; ZoBell and Anderson, 1936). The importance and impacts of biofilms in natural ecosystems were also increasingly understood in the latter half of the last century. For example, the prevalence

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of bacterial and algal biofilms on surfaces in natural streams and the predominant effect of such mixed community biofilms on the otherwise pristine water ecosystem were unraveled by Geesey *et al.* (1977, 1978). Also, biofilm-forming marine bacteria were shown to be the initial colonizers with significant effects on the subsequent development of complex biofouling communities (Wilson, 1955).

What were the early observations on mechanisms of attachment and biofilm formation? For a biofilm to develop, micro-organisms must first attain the proximity of the surface in question and then attach to that surface. Micro-organisms may be transported to surfaces in aquatic environments by several means (Marshall, 1986), including sedimentation, fluid dynamic forces, motility guided by chemotaxis and more passive mechanisms such as Brownian motion and cell surface hydrophobicity (Marshall and Cruickshank, 1973).

The first definitive microscopic studies of bacteria in the vicinity of a solid surface was undertaken by ZoBell (1943), who noted that attachment was probably a two-phase process consisting of a primary, but reversible, attraction to the surface followed by a later firm, irreversible, adhesion. ZoBell (1943) also speculated on the possibility of polymer production by the bacteria being involved in the firm adhesion phase. Later, Marshall *et al.* (1971) published a seminal paper detailing the behavior of marine motile bacteria at solid surfaces and laid the foundation for a mechanistic physico-chemical explanation of the initial stages of reversible and irreversible attachment and hence colonization of surfaces by bacteria.

Marshall and co-workers addressed why bacteria which possess a net negative charge are not repelled from like-charged surfaces in natural habitats. As a bacterium approaches a solid surface, the energy of interaction that occurs between the interacting electrical double layers around such surfaces was explained in terms of the Derjaguin-Landau and Verwey-Overbeek (or DLVO) theory (Marshall, 1976). At low electrolyte concentrations the thickness of the double layers of counter ions increases resulting in a total repulsion between the interaction surfaces, whereas at high electrolyte concentrations the double layers are compressed exposing a secondary attraction zone, resulting from London-van der Waals attraction forces, at some distance from the surface. It is this zone that accounts for the reversible attraction of bacteria to a surface. Marshall *et al.* (1971) also demonstrated the need for the production by the bacterium of very fine extracellular polymeric fibrils to bridge the repulsion barrier and, hence, ensure irreversible adhesion to the surface, at a primary attraction zone in close proximity to the surface, observations subsequently confirmed by several investigators (e.g. Marshall and Cruickshank, 1973, and Fletcher and Floodgate, 1973). Further details and our current understanding of the orientation, motility and the shift from reversible to irreversible attachment by surface localized bacterial cells are provided in the chapter by MacEachran and O'Toole in this volume.

While other factors, such as cell-surface hydrophobicity (Marshall and Cruickshank, 1973), the nature of the conditioning films on surfaces (Baier, 1980) surface active components for altered viscosity, temporary adhesion and translational motion (Humphrey *et al.*, 1979), and extracellular matrix components (Pamp *et al.*, this volume) contribute to bacterial adhesion processes in natural habitats, the discovery of the mechanisms of reversible and irreversible adhesion as the key steps in early bacterial colonization, laid the foundation for modern molecular biofilm research, as detailed in this monograph, for a range of organisms and habitats. Also, genetic analysis substantiated the important role of

surface appendages, such as fimbriae and flagella, in specific adhesin–receptor mediated irreversible binding (Marshall, 1984).

The current volume presents recent progress in our understanding of specific mechanisms that underpin the biofilm mode of life. This includes, in addition to areas highlighted above, studies on interactions among biofilm cells at a variety of levels; metabolic interactions, cell to cell communication and signal transmission across cells and species (see chapters by Yarwood, Wood and Bentley, Atkinson *et al.*, Kolenbrander *et al.*, Dow *et al.*, and Eberl *et al.*, this volume). Moreover, interactions based on competition and predation across species and domains have enjoyed strong interest in recent years as described in the chapters by Hogan, Matz and Eberl *et al.* Modern molecular techniques in combination with confocal laser scanning microscopy have also contributed greatly to our understanding of biofilm community structure and composition (see chapters by Kjelleberg and Givskov, and Kolenbrander *et al.*) as well as mechanisms of biofilm dissolution (see Chapter 9). Genomic based studies of such biofilm specific traits are rapidly unraveling much novel information on both monospecies and mixed species communities. Interestingly modern molecular microbiology increasingly adopts approaches developed in microbial ecology to better understand the sessile life style of bacteria in particular microbial habitats, including those of the human body.

The recent surge in studies of biofilms is driven not only by our desire to explore the basic mechanisms that mediate bacterial colonization on surfaces, but also the realization that bacterial evolution very effectively allows for resistance to develop and hence makes almost any control measure we develop ineffective. Hence, like a century ago we still face bacterial supremacy, a fact that urges us to focus on more applied aspects of the discipline with new ideas and approaches generated from our basic knowledge about “life on surfaces” to win the battle against unwanted bacteria. The updated aspects of biofilm research reported in this volume should encourage interested researchers to integrate these new concepts in attempts to develop newer innovative approaches in studies of the behavior and functions of sessile bacteria.

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