Life Sciences Research Report 31

Microbial Adhesion and Aggregation

K. C. Marshall, Editor

Dahlem Konferenzen

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Report of the Dahlem Workshop on
Microbial Adhesion and Aggregation
Berlin 1984, January 15 – 20

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Dahlem Workshop Reports
Life Sciences Research Report 31
Microbial Adhesion and Aggregation

The goal of this Dahlem Workshop is:
to explore the mechanisms and consequences
of microbial adhesion and aggregation

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The Dahlem Konferenzen

Founders

Recognizing the need for more effective communication between scientists, especially in the natural sciences, the Stifterverband für die Deutsche Wissenschaft*, in cooperation with the Deutsche Forschungsgemeinschaft**, founded Dahlem Konferenzen in 1974. The project is financed by the founders and the Senate of the City of Berlin.

Name

Dahlem Konferenzen was named after the district of Berlin called "Dahlem", which has a long-standing tradition and reputation in the arts and sciences.

Aim

The task of Dahlem Konferenzen is to promote international, interdisciplinary exchange of scientific information and ideas, to stimulate international cooperation in research, and to develop and test new models conducive to more effective communication between scientists.

Dahlem Workshop Model

Dahlem Konferenzen organizes four workshops per year, each with a limited number of participants. Since no type of scientific meeting proved effective enough, Dahlem Konferenzen had to create its own concept. This concept has been tested and varied over the years, and has evolved into its present form which is known as the *Dahlem Workshop Model*. This model provides the framework for the utmost possible interdisciplinary communication and cooperation between scientists in a given time period.

**The Donors Association for the Promotion of Sciences and Humanities*

***German Science Foundation*

The main work of the Dahlem Workshops is done in four interdisciplinary discussion groups. Lectures are not given. Instead, selected participants write background papers providing a review of the field rather than a report on individual work. These are circulated to all participants before the meeting to provide a basis for discussion. During the workshop, the members of the four groups prepare reports reflecting their discussions and providing suggestions for future research needs.

Topics
The topics are chosen from the fields of the Life Sciences and the Physical, Chemical, and Earth Sciences. They are of contemporary international interest, interdisciplinary in nature, and problem-oriented. Once a year, topic suggestions are submitted to a scientific board for approval.

Participants
For each workshop participants are selected exclusively by special Program Advisory Committees. Selection is based on international scientific reputation alone, although a balance between European and American scientists is attempted. Exception is made for younger German scientists.

Publication
The results of the workshops are the Dahlem Workshop Reports, reviewed by selected participants and carefully edited by the editor of each volume. The reports are multidisciplinary surveys by the most internationally distinguished scientists and are based on discussions of new data, experiments, advanced new concepts, techniques, and models. Each report also reviews areas of priority interest and indicates directions for future research on a given topic.

The Dahlem Workshop Reports are published in two series:
1) Life Sciences Research Reports (LS), and
2) Physical, Chemical, and Earth Sciences Research Reports (PC).

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Microbial Adhesion and Aggregation, ed. K.C. Marshall, pp. 1-3. Dahlem Konferenzen 1984. Berlin, Heidelberg, New York, Tokyo: Springer-Verlag.

Introduction

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Microbial adhesion and aggregation can mean many things to many people. Why is this so? These processes of adhesion and aggregation are manifest in many forms and are studied by researchers from a wide variety of disciplines. Adhesion and/or aggregation of microorganisms are involved in certain diseases of humans and animals, in dental plaque formation, in industrial processes, in fouling of man-made surfaces, in syntrophic and other community interactions between microorganisms, and in the activity and survival of microorganisms in natural habitats. Different approaches to the study of these phenomena have developed depending on whether the work has been carried out by microbiologists, biotechnologists, physical chemists, or engineers.

The goal of this workshop was to explore the mechanisms and consequences of microbial adhesion and aggregation. Both of these processes involve the interaction between microorganisms and some type of surface. These surfaces may be inert materials, the exterior of other, usually larger, organisms, or other microorganisms of the same or different species (aggregation). Several symposia and a number of books have been devoted to the subject of adhesion and aggregation in recent years, but they have not provided an adequate forum for the exchange of ideas and concepts from the different interested disciplines. This workshop involved the bringing together of microbial physiologists, ecologists and geneticists, industrial microbiologists, physical and polymer chemists, and bioengineers in an attempt to assess the current status of the field and, hopefully, to develop new approaches and concepts as a result of the interdisciplinary discussions.

The first really serious study on microbial adhesion was that of ZoBell (2), who recognized the possibility of macromolecular conditioning films modifying surfaces prior to microbial adhesion and suggested that adhesion was a biphasic phenomenon, with an initial reversible phase being followed by a firm, irreversible binding of microorganisms to surfaces. Progress in all aspects of adhesion and aggregation processes was very slow, until a reawakening of interest in the early 1970s. At that time, research workers in various fields became conscious of the almost universal association of microorganisms with surfaces or with each other (1). Research on microbial adhesion and aggregation has now reached almost explosive proportions and, consequently, it seemed timely to stand back and reassess the directions in which this research might move in the future. Because of the major contributions made to research on adhesion and aggregation by microbiologists, chemists, and engineers, it was deemed essential that all of these fields be represented at the workshop in order that the subject be considered from a very broad viewpoint.

The workshop was planned to consider four major aspects of the main topic, namely: a) the mechanisms whereby microorganisms adhere to surfaces, b) the development of biofilms and the consequences of such films, c) the activities of microorganisms on surfaces, and d) the process and consequences of microbial aggregation. Each aspect was considered by a group consisting of representatives of different disciplines, and each group aimed to examine old concepts and attempted to evolve new concepts based on the context of its discussions. Of course, one of the ultimate aims in studying microbial adhesion and aggregation is to evolve the means for manipulating these processes to our own advantage. This may be to control the fouling of ship hull or heat exchanger surfaces, to control dental plaque or periodontal disease, to control cholera and other intestinal diseases, to enhance aggregation in activated sludge systems, to enhance biofilm development in fixed film fermenters, or to modify microbial associations in nature to establish more effective consortia.

Finally, some brief statements regarding certain confusing terminology that has developed in this field. Adhesion is a process that can, in essence, be explained on a physicochemical basis. Consequently, the word adhesion should be used in place of the term adherence (which is best used in the sense of, say, adherence to a principle). There is a need to clarify the usage of the terms substrate and substratum. A substrate (pl.: substrates) is a material utilized by microorganisms, generally as a source of energy. A substratum (pl.: substrata) is a solid surface to which a microorganism may attach.

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(1) Marshall, K.C. 1976. Interfaces in Microbial Ecology. Cambridge, MA: Harvard University Press.

(2) ZoBell, C.E. 1943. The effect of solid surfaces upon bacterial activity. J. Bacteriol. 46: 39-56.

Standing, left to right:
Paul Rutter, Rolf Freter, Mike Silverman, Ian Robb, Hinrich Mrozek, Frank Dazzo, David Gingell

Seated, left to right:
Garth Jones, Stanislawa Tylewska, Staffan Kjelleberg, Ellen Rades-Rohkohl, Kevin Marshall

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Mechanisms of Adhesion
Group Report

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INTRODUCTION
The subject, Mechanisms of Microbial Attachment, has proved to be a fertile source of argument between both microbiologists and physical chemists. In order to find common ground for discussion, the group endeavored to come to a consensus regarding a number of definitions pertinent to the subject. Some of these are described in the text and others will be found at the end of this report.

The group restricted its discussions to the interactions between microbial cells and substrata in close proximity. The general topic of deposition was not covered in any detail. It was noted at the outset that the adhesion of microbial cells to animate and inanimate surfaces and the mechanism by which they adhere can be influenced directly by changes in environmental conditions. Many examples of this will be found in Rutter and Vincent, Kjelleberg, Jones, Dazzo, and Silverman et al. (all this volume).

WHAT IS ADHESION?
It was agreed that microbial adhesion can be defined unambiguously only in terms of the energy involved in the formation of the adhesive

junction. Thus, the strength with which a microbial cell may be said to have adhered to a substratum can be quantified as the work required to remove the cell to its original isolated state. It is important that removal of the cell from the substratum does not leave fragments of the cell wall or cell surface polymers remaining on the substratum. If this is so, then both adhesive and cohesive failure will have occurred, giving smaller energies of adhesion than the true value. All other definitions can lead to confusion, although they might be useful to researchers working with particular, well characterized systems. In addition to the definition of adhesion given above, it was felt that a more functional description of adhesion was also required. This prompted the use of the term "association" which is defined at the end of this report.

MEASUREMENT METHODS

Having agreed on what was meant by adhesion and association, a number of methods of measuring these two phenomena were discussed. The experimental details of a number of assays designed to quantify microbial association in particular systems are described in the papers by Rutter and Vincent, Kjelleberg, Jones, and Dazzo (all this volume).

The Direct Measurement of Adhesion by Methods Involving the Separation of Cell and Substratum

Physicochemical techniques are now available for measuring the force of adhesion between molecularly smooth surfaces. It may be possible to measure the adhesion of bacteria grown as monolayer cultures on well-defined surfaces by a similar technique (4). Other techniques involving micromanipulation or well-defined shear conditions might also be devised.

Deposition Studies under Controlled Hydrodynamic Conditions

These methods quantify the association of microbial cells with substrata. It is possible to devise experimental systems where a suspension of microbial cells undergoes laminar flow over a surface. Under these circumstances it is often possible to calculate the number of collisions between the cells and substratum from the bulk properties of the system, e.g., cell concentration, viscosity, temperature, flow velocity, etc. Comparison between the observed cell deposition rate and calculated collision rate enables the deduction of a collision efficiency (deposition rate/collision rate) which may be used to quantify the association under study. The advantages of this type of technique are that many of the often poorly defined quantities in experiments designed to compare the interactions of microbial cells with different surfaces, e.g., cell concentration, settling under gravity, etc., are encompassed by the collision rate calculations (2).

Techniques Involving Contact Angles

Good correlations have been obtained between certain association phenomena and interaction energies calculated from deduced surface energies of the apposing surfaces. These techniques do not measure adhesion but calculate the energy involved from contact angle measurements and compare this with the observed interaction of cells with different surfaces, including those of phagocytes (1). The striking fact about these studies is the good correlation observed. However, it was felt that there were a number of problems arising from the measurement of microbial cell surface contact angles, particularly with respect to the effect of the diagnostic liquids on the cell surface components and the effect of removing the cells from their original environment, especially if drying is involved.

Mammalian Cell Techniques

It was pointed out that techniques involving the deformation of mammalian cell aggregates under gravity and the response to electrolytes of red blood cells held in secondary minimum contact with a substratum have been used to deduce adhesive forces. It may be possible to adapt these techniques to the study of certain types of microbial adhesion, although doubts were expressed because of the apparent rigidity of bacterial cell walls.

Measurement of the Association of Microbial Populations with Surfaces

It was recognized that the proportion of bacteria within a population which remain attached to a surface in a particular experimental environment is a useful and convenient measure of association (see, for example, Dazzo, Jones, and Kjelleberg, all this volume).

SPECIFIC AND NONSPECIFIC ADHESION

Discussion of the terms "specific" and "nonspecific" occupied a considerable amount of the group's time but eventually led to definitions which the group considered to be helpful.

Before arriving at a definition, it was necessary to come to some agreement as to what constituted a microbial cell, host cell, and inanimate substratum surface. Animate cell surfaces are invariably coated with complex macromolecules extending in various ways into the surrounding environment. It was also the experience and opinion of the group that