that the surface charge density of the bacterial cells is on the order of 0.3 C.m⁻². This value is much larger than expected from the zeta potential of the pure bacterial suspension (8.10⁻³C.m⁻²). We conclude that the chains adsorb through a very thin surficial layer close to the surface, within the lipopolysaccharidic brush, mainly within the Stern layer, as the charged groups on the chains replace the cations condensed on the surface. This mechanism increases the entropy of the system.

The polymer layer of adsorption at saturation may be thought of as a network of polymeric rods crossing each other at distances on the order of 1 nm. Below the plateau of adsorption, the distance between crossing points increases.

Flocculation of the suspension

a) Optimum of flocculation: the polymer was added to the bacterial suspension, as mentioned above. After the stirring period, sedimentation of the samples was allowed for one to two hours. The optical density of the supernatant was then measured at 450 nm (optical path: 1 cm). A low optical density is the sign of a strong flocculation, since aggregated bacteria are removed from the suspension during the sedimentation period.

Results: three regimes are observed, as expected by previous studies on abiotic colloids. At low polymer dosages, a partial flocculation of the suspension occurs, increasing with the quantity of polymer added. At intermediate dosages, the flocculation is optimal; in our case, almost all bacterial cells aggregate, and the value of the optical density is close to zero. At large polymer dosages, the cells are redispersed by the polymer.

Interpretation: we believe that our results can be explained by electrostatic arguments. The bridging mechanism does not play a role here, since the chains are stuck on the surface and no loop can extend towards the solution. When the quantity of adsorbed polymer increases, the overall electrostatic repulsion between the cells decreases, vanishes at the isoelectric point, and then increases again. The redispersion regime can thus be explained by an electrostatic restabilization of the suspension. The flocculation of the cells can be explained by a "patch" mechanism, as proposed by other authors for abiotic colloids. The positively charged surface on one cell would attract the negatively charged regions on another cell.

b) Size and structure of the aggregates: the distribution of size and the structure of the aggregates was investigated with a granulometer: a laser bearn is diffused by the suspension. The intensity diffused is recorded as a function of the wave vector.

Results: we find that the size polydispersity of the aggregates is very large at low polymer dosages and small at the optimum of flocculation. In this regime, a plateau in the slope of the scattering curve suggests that the aggregates have a self-similar structure with a fractal dimension of 1.9.

Interpretation: these results suggest that aggregates formed in the regime of the optimum of flocculation correspond to a diffusion-limited-aggregation process, while those formed at low polymer dosages correspond to a reaction-limited aggregation process. At the optimum of flocculation, bacterial cells stick to each other easily; while at low polymer dosages, they aggregate only where the area of contact is covered by some polymer.

Conclusions

We have shown that methods and concepts of modern polymer and colloid physics can be successfully applied to bacterial surfaces in order to describe quantitatively their interfacial properties and the properties of the aggregates that they form. Extension of our work to weakly charged and to negatively charged polymers could lead to some new insights on biofilm formation and biofilm structures. Further studies with mixed suspensions of bacteria and mineral particles, may also lead to a better quantitative description of soil microstructures. Further details and references can be found in our detailed paper, submitted April 2000, to the journal "Langmuir".

References

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