Does ultrapure dialysate prevent the development of biofilm in dialysis therapy?

Sir,
Biofilm, macroscopically visible as green/yellowish deposit of fluid pipes, is commonly present in dialysis modules and parts of the water treatment system. Biofilm consists of a fine fibrillar meshwork of bacterial origin, with trapped microorganisms [1]. The presence of biofilm in dialysis systems is a point of concern, first because biofilms continuously release bacterial components such as peptidoglycans and endotoxins. Moreover, bacteria present in biofilm are highly resistant against cleaning and disinfection procedures and therefore eradication, once a biofilm is present, is virtually impossible [1]. The continuous release of bacterial components into the dialysate may contribute to the activation of the immune system and, therefore, to a chronic inflammatory state in haemodialysis patients [2]. This suggests that prevention of the development of biofilm is relevant in haemodialysis therapy. As contamination of the dialysate may lead to the development of biofilm, the use of ultrapure dialysate is expected to result in a reduced development of biofilm; however, no data on this subject are as yet available.

In our hospital, a new water treatment system was installed in 1998, including continuous ultraviolet disinfection. Moreover, this installation is (except for the RO unit) treated
with ozone once a week in order to prevent bacterial growth. Since the installation of the new unit, no positive cultures were found in tap water, whereas endotoxin concentration (LAL test) was maintained at <0.03 IU/l.

The aim of this preliminary study was to investigate whether the use of ultrapure dialysate could prevent the development of biofilm. Therefore, we performed bacteriological analyses and investigated the development of biofilm in the tubing segment between the water tap and the dialysis module. This part was chosen because it is not included in the disinfection programme of either the water treatment system or dialysis module and is therefore prone to the development of biofilm.

Tubing samples of 5 cm length were taken on a weekly basis from the dialysis path of one dialysis machine over a period of 24 weeks (1034 treatment hours). The samples were taken under aseptic conditions. The whole inner surface of the tubes was sampled using a sterile cotton swab. The swab was put into 2 ml sterile NaCl 0.9% and vortexed thoroughly, and 100 μl of this solution was spread onto a Cled Agar Plate. After incubation for 48 h at 37°C the number of colony forming units (CFU) on the agar plate was counted and identified using the API-20 system (BioMerieux). All experiments were performed in duplicate.

In addition, samples obtained after 24 weeks were prepared for scanning electron microscopy (SEM). Air dried tubing was cut into pieces and plated with gold. The surface was studied by SEM (Philips XL 30, 10 kV, secondary electrons). Representative images are shown in Figures 1 and 2.

In the sample taken at week 1, 100 CFU/cm² Enterobacteriaceae were present. In the samples taken at weeks 4 and 6, 40 CFU/cm² Enterobacteriaceae and 700 CFU/cm² non-fermenters were found, respectively. Samples taken thereafter until the last sample in week 24 of the experiment showed no bacterial growth. As displayed in Figure 1, SEM of the samples taken after 4 weeks showed an absence of biofilm, which is in sharp contrast to the extensive biofilm found in a comparable tubing segment obtained from another dialysis unit (Figure 2). This is a preliminary observation, since only a single tube was studied and a control group is lacking. However, it is well known from the literature that biofilm is ubiquitously present in dialysis modules and water treatment systems [3,4].

In conclusion, in this preliminary study, the use of ultrapure dialysate was associated with absence of biofilm in the most vulnerable part of the dialysis module. The absence of biofilm development may have important consequences, as biofilm may have adverse effects in dialysis therapy as a residential source of contamination of the dialysis circuit. Further controlled studies are in progress to establish the presence or absence of biofilm in the tubing of dialysis equipment using either standard or ultrapure dialysate.

Fig. 1. Tubing segment, showing complete absence of biofilm, from a water treatment system delivering ultrapure water.
Fig. 2. Tubing segment, showing extensive biofilm formation, from a standard water treatment system.

1Department of Medical Microbiology
2Department of Internal Medicine
3Department of Electron Microscopy
University Hospital Maastricht Maastricht The Netherlands