

International Journal of Pharmaceutics 149 (1997) 251-254

Modification of biomaterial surface characteristics by body fluids in vitro

J.G. McGovern, C.P. Garvin, D.S. Jones, A.D. Woolfson, S.P. Gorman *

Pharmaceutical Devices Group, School of Pharmacy, Medical Biology Centre, The Queen's University of Belfast, 97 Lisburn Road, Belfast BT9 7BL, U.K.

Received 5 December 1996; accepted 16 January 1997

Abstract

In practice, polyvinyl chloride endotracheal tubes and polyurethane urinary catheters are located in areas where they are exposed to the conditioning fluids saliva and urine, respectively. Samples of both biomaterials were incubated in these conditioning fluids and, following treatment, dynamic contact angle measurement and surface roughness assessment by atomic force microscopy were used to analyse surface characteristics. Over a 24 h period of contact with the conditioning fluids, the surface of both materials became significantly more hydrophilic (p < 0.05) and non-uniform changes in surface roughness were observed. This study has shown that in vitro treatment of polyvinyl chloride and polyurethane with conditioning body fluids alters the surface characteristics of the biomaterials and this effect is progressive with time as determined by surface advancing contact angle measurement. © 1997 Elsevier Science B.V.

Keywords: Contact angle; Polyurethane; Polyvinyl chloride; Saliva; Surface roughness; Urine

Body fluids such as plasma and serum can interact with polymers with a subsequent inhibition of microbial adhesion (Carballo et al., 1991). In the case of polyvinyl chloride (PVC) endotracheal (ET) tubes, the device is exposed to a salivary liquid medium. The transient bathing of the constituent polymer by a body fluid may result in unexpected and undesirable sequelae such as colonisation by potentially pathogenic microorganisms and subsequent infection (Gorman et al., 1993a). Polyurethane (PU) catheters reside in the urinary tract and are in regular contact with urine conditioning the device surface, with encrustation and microbial colonisation being reported (Keane et al., 1994). It remains unclear whether such conditioning is instantaneous or is progressive with time. This investigation endeavours to address the topic by examining surface roughness and hydrophobicity, through dynamic contact an-

^{*} Corresponding author.

^{0378-5173/97/\$17.00 © 1997} Elsevier Science B.V. All rights reserved. *PII* S0378-5173(97)04883-7

Mean (\pm S.D.) PU advancing contact angle, receding contact angle and surface roughness following incubation in conditioning fluids for 1 min, 4 h and 24 h

Condition- ing fluid	Advancing contact angle (°)			Receding contact angle (°)			Surface roughness (nm)		
	1 min	4 h	24 h	1 min	4 h	24 h	1 h	4 h	24 h
Volunteer urine 1	95.4 <u>+</u> 0.4	57.4 ± 3.3	62.0 ± 0.4	58.5 ± 0.3	44.4 ± 0.1	62.0 ± 0.1	59.5 <u>+</u> 11.0	49.3 ± 9.2	183.9 ± 20.5
Volunteer urine 2	99.9 ± 0.5	49 .2 ± 1.5	74.4 <u>+</u> 4.3	67.1 ± 0.6	48.1 ± 0.6	63.0 ± 0.1	61.1 ± 10.7	78.4 ± 9.3	22.5 ± 6.7
Volunteer urine 3	101.9 ± 0.3	47.7 ± 2.2	59.1 <u>+</u> 1.0	69.1 ± 1.8	51.2 ± 0.1	58.4 ± 0.1	35.1 ± 6.4	86.9 ± 3.4	36.9 ± 3.6
Volunteer urine 4	97.7 <u>+</u> 0.9	51.8 ± 2.7	71.5 ± 1.7	58.8 ± 0.5	47.9 ± 0.1	61.8 ± 0.2	56.4 ± 8.8	56.6 <u>+</u> 10.1	43.0 ± 9.6
Patient urine 1	98.4 ± 0.8	95.1 ± 0.3	96.7 ± 1.1	74.03 ± 0.6	58.2 ± 1.5	63.5 ± 0.4	47.1 ± 9.7	62.2 ± 11.7	34.0 ± 7.0
Patient urine 2	96.0 ± 0.4	97.2 ± 0.7	69.0 ± 0.9	64.6 ± 1.7	57.4 ± 1.7	46.2 ± 0.6	40.7 ± 11.3	36.6 ± 11.2	39.2 ± 8.5
Patient	97.9 ± 0.4	87.7 ± 5.9	86.3 ± 2.7	74.9 <u>+</u> 1.6	60.5 ± 1.4	52.1 ± 0.5	31.7 ± 7.8	35.8 ± 7.9	38.4 ± 10.5
Patient urine 4	96.5 ± 0.4	94.0 ± 1.1	65.6 ± 3.4	74.9 ± 1.1	61.8 ± 1.5	52.0 ± 0.3	41.1 ± 10.8	48.4 ± 12.1	48.3 ± 13.0
Artificial urine	51.6 ± 0.7	48.5 ± 0.2	59.4 ± 0.2	49.9 ± 0.2	48.4 ± 0.2	60.3 ± 0.1	56.2 ± 10.0	70.9 ± 9.0	63.2 ± 8.2
PBS	98.7 <u>+</u> 1.0	70.2 ± 3.8	91.9 <u>+</u> 0.3	61.5 ± 1.1	46.2 ± 1.1	58.5 <u>+</u> 0.3	20.2 ± 4.3	56.6 ± 8.2	16.9 <u>+</u> 3.6

gle measurement, of PU and PVC following incubation in urine and saliva, respectively.

Saliva and urine samples were collected from four healthy adult volunteers and additional urine samples obtained from donors diagnosed as having renal calculi; artificial saliva (Zampatti et al., 1994) and artificial urine (Tunney et al., 1996) were also prepared. Sections of PVC were submerged in volunteer saliva, artificial saliva and PBS and incubated at 37°C for periods of 1, 5, 10, 15, 30, 45 and 60 min and 2, 4, 8 and 24 h before removal from fluid to determine surface roughness and dynamic contact angle. Tabulated data shows results only for 1 min, 4 h and 24 h. These parameters were measured as described previously (Jones et al., 1997). The experimental procedure was repeated for PU using volunteer, patient and artificial urine and PBS. Results were analysed using two-way analysis of variance (p < 0.05 denoting significance).

Advancing contact angles for both biomaterials generally exhibited a significant decrease (p < 0.05) between 1 min and 24 h (Tables 1 and 2); decreases observed in receding contact angle were less in magnitude and in some cases an increase was recorded.

In general, PVC incubated with human saliva showed a significantly lower (p < 0.05) advancing contact angle relative to treatment with PBS or artificial saliva. Incubation of PVC with human saliva produced no identifiable pattern in advancing contact angle initially. However, advancing contact angle later decreased between 4 and 24 h.

Each of the volunteer urine samples had little effect on the advancing contact angle of PU initially. A significant decrease (p < 0.05) between 1 min and 4 h was consistent with each sample and measurements significantly increased (p < 0.05) by 24 h. With patient urine, the general path followed by advancing contact angle of PU was non-specific in the first 4 h; by 24 h, a significant

Table 2

Mean (\pm S.D) PVC advancing contact angle, receding contact angle and surface roughness following incubation in conditioning fluids for 1 min, 4 h and 24 h

Condi- tioning fluid	Advancing contact angle (°)			Receding contact angle (°)			Surface roughness (nm)		
	l min	4 h	24 h	1 min	4 h	24 h	1 h	4 h	24 h
Volunteer saliva 1	84.7 <u>+</u> 1.8	89.6 <u>+</u> 1.3	55.3 ± 2.3	56.0 ± 0.1	59.9 ± 0.1	59.8 ± 0.1	114.7 ± 12.9	117.9 ± 18.0	110.6 ± 14.2
Volunteer saliva 2	86.7 ± 0.3	63.7 ± 4.3	52.4 ± 2.4	60.9 ± 0.5	55.9 ± 0.2	59.1 ± 0.0	216.7 ± 25.3	164.7 ± 9.0	101.4 ± 8.9
Volunteer saliva 3	89.0 ± 0.2	71.2 ± 2.1	66.8 ± 2.8	58.1 ± 0.3	53.0 ± 0.1	54.4 ± 0.3	65.3 ± 8.1	132.1 ± 12.1	145.3 ± 11.5
Volunteer saliva 4	85.6 ± 1.4	85.6 ± 0.6	51.6 ± 1.2	58.3 ± 0.1	60.4 ± 0.3	59.9 <u>±</u> 0.4	64.9 <u>+</u> 7.9	40.6 ± 4.2	84.0 ± 7.1
Artificial saliva	97.0 ± 1.0	90.7 ± 0.3	64.7 ± 1.8	64.6 ± 1.2	62.8 ± 0.2	56.0 ± 0.1	101.5 ± 13.5	109.4 <u>+</u> 9.6	63.7 ± 3.8
PBS	94.9 ± 1.7	73.5 ± 2.1	68.3 <u>+</u> 0.7	69.9 ± 0.2	59.5 ± 0.2	58.0 ± 0.1	38.4 ± 4.4	15.0 <u>+</u> 1.6	49.8 ± 5.2

decrease (p < 0.05) in advancing contact angle was observed with three of the patient urine samples.

Biomaterial surface roughness was measured after 1, 4 and 24 h. Following incubation with controls and saliva or healthy urine, surface roughness values lacked consistency and no overall trends were identified during the 24 h period. However, measurements of microrugosity were more consistent for PU which had been incubated in patient urine and, generally, significant differences in surface roughness were not observed between incubation times.

Irregularities in the surface of PVC appear in unused ET tubes and these areas are more prone to accumulation of microbial biofilm (Gorman et al., 1993a). Where tubes are colonised, microorganisms preferentially adhere to a biological film of human origin rather than to the constituent polymer itself (Poisson et al., 1991). In previous work, treatment of PVC with saliva reduced the adherence of saliva-treated respiratory isolates and decreased PVC contact angle and surface microrugosity (Jones et al., 1997). In this study it was observed that incubation of PVC for 24 h with PBS, artificial saliva or human saliva led to a significant decrease in contact angle. However, initial measurement of contact angle revealed patterns which were non-uniform and a definite decrease as a result of saliva treatment was not observed until after 4 h. Changes in contact angle of PVC were not reflected in surface microrugosity.

When PU was incubated with volunteer urine, an earlier onset in decreasing advancing contact angle and a tendency to an increasing contact angle after longer periods of incubation in urine were observed. These findings signify the complexity of the interaction between the body fluid components and the biomaterial surface. Treatment of catheter materials with body fluids such as plasma and serum albumin significantly reduced bacterial adherence (Carballo et al., 1991), an action attributed to the adsorption of components of the body fluids on to the surface of the biomaterials. A significant decrease (p < 0.05) in contact angle was observed between 1 min and 24 h with each of the volunteer samples, indicating the change to a more hydrophilic biomaterial surface in practice.

After exposure to volunteer urine, PU showed considerable variation in surface roughness and no firm conclusions could be drawn from these data. However, PU which was incubated with patient urine displayed a more consistent microrugosity. Surface roughness irregularities observed on peritoneal catheter materials can be inherent as a result of the manufacturing process and can also be induced through long term use in the patient. Such changes to the catheter surface allows increased bacterial adherence and hence catheter-related infection (Gorman et al., 1993b). Interestingly, this study showed that treatment of the material with spent peritoneal dialysate significantly reduced microbial adherence.

In conclusion, the in vitro development of conditioning films on polyvinyl chloride and polyurethane by saliva and urine, respectively, has been shown to progress with time. Contact angle measurement of surface characteristics appears to be a useful method of assessing changes arising in biomaterial surfaces in contact with conditioning fluids but further studies are required to determine the nature and influence of 'conditioned' surfaces in respect of medical device use in clinical practice.

References

Carballo, J., Ferreiros, C.M. and Criado, M.T., Importance of

experimental design in the evaluation of the influence of proteins in bacterial adherence to polymers. *Med. Microbiol. Immunol.*, 180 (1991) 149-155.

- Gorman, S.P., Adair, C., O'Neill, F., Goldsmith, C. and Webb, H., Influence of selective decontamination of the digestive tract on microbial biofilm formation on endotracheal tubes from artificially ventilated patients. *Eur. J. Clin. Microbiol. Infect. Dis.*, 12 (1993a) 9-17
- Gorman, S.P., Mawhinney, W.M., Adair, C.G. and Issouckis, M., Confocal laser scanning microscopy of peritoneal catheter surfaces. J. Med. Microbiol., 38 (1993b) 411-417.
- Jones, D.S., McGovern, J.G., Woolfson, A.D. and Gorman, S.P., The role of physiological conditions in the oropharynx on the adherence of respiratory isolates to endotracheal tube polyvinyl chloride. *Biomaterials*, 1997 (in press)
- Keane, P.F., Bonner, M.C., Johnston, S.R., Zafar, A. and Gorman, S.P., Characterisation of biofilm and encrustation ureteric stents in vivo. *Br. J. Urol.*, 73 (1994) 687–691.
- Poisson, D.M., Arbeille, B. and Laugier, J., Electron microscopic studies of endotracheal tubes in neonates: do microbes adhere to the polymer? *Res. Microbiol.*, 142 (1991) 1019-1027.
- Tunney, M.M., Bonner, M.C., Keane, P.F. and Gorman, S.P., Development of a model for assessment of biomaterial encrustation in the upper urinary tract. *Biomaterials*, 17 (1996) 1025-1029.
- Zampatti, O., Roques, Ch., Michel, G., An in vitro mouth model to test antiplaque agents: preliminary studies using a toothpaste containing chlorhexidine. *Caries Res.*, 28 (1994) 35-42.