

SURVIVAL OF DIFFERENT BACTERIAL SPECIES ON DIFFERENT
INANIMATE SURFACES STORED AT DIFFERENT CONDITIONS:

(1) SURVIVAL OF Escherichia coli ON VARIOUS SOLID
SURFACES STORED AT TWO TEMPERATURES.

A.S.R. AL-Obaidi

Department of Microbiology, College of Veterinary
Medicine, University of Baghdad.

SUMMARY

Survival of E. coli (0114:K90) on each of glass, wood, polystyrene and metal surfaces was studied. Artificially contaminated surfaces were stored at both 4°C and room temperature (18-20°C), and samples from each surface were collected approximately after 1, 3, 10 and 40 days of storage, and cultured for bacterial growth. E. coli was recovered from glass and polystyrene surfaces after 1, 3 and 10 days of storage at both temperatures; from wood up till 40 days of storage at both temperatures; but the only exception was metal surface, where the organism was recovered only after 1 and 3 days of storage at room temperature, but was not isolated from the same surface stored at 4°C.

INTRODUCTION

The sorption of bacteria to surfaces is a general phenomenon encountered in natural environments with important ecological implications(1). In addition, bacterial adhesion to a surface is known to play an important role in many activities, like infection of different tissues (2), ship fouling (3), etc.; but still little is known about such adherence mechanism. Extra-cellular polymer production is to accompany attachment to polar substrate, since the polymers are mostly polysaccharides (1, 4). On the other hand, certain bacteria are known to attach rapidly and firmly to nonpolar substrate, suggesting that time-dependent physiological processes, like polymer secretion, are not

essential to initial attachment, and that physiochemical forces predominate.

Bearing in mind the probable consequent effects of bacteria on public health, either directly or indirectly, when they become attached or adherent to surfaces, this study was undertaken to explore the duration of survival, the role of temperature of storage, the type surface and the abilities of different media in recovering the bacteria from the surfaces being studied.

MATERIALS & METHODS

- A. Surfaces used: The surfaces used in this experiment were: glass (microscope glass slides); metal (small coins); polystyrene (weighing boats, 4 x 4 cm), and wood (small pieces of palm-tree stems). Sixteen of each of the glass, metal and wood were sterilized by autoclaving at 121°C for 15 min., and the polystyrene by immersion in 70% ethanol for 5 min., then left to dry before use. All sterile surfaces were kept separately in sterile screw-capped containers until being used.
- B. Contamination of surfaces: *E. coli* serotype O114:K90, recovered from a case of infantile diarrhoea (at Sadam's Central Hospital For Children) was cultured onto MacConkey agar plates and incubated overnight at 37°C, then transferred to 50 ml. buffered peptone water kept in 250 ml flask, and mixed by a magnetic stirrer at 50 rounds per minute for 4-5 hours. This was termed: "The original broth". Viable bacterial count was then done, using the technique of Miles *et al.* (5), with buffered peptone water as the diluent and both MacConkey and Brain heart-infusion agars as the counting media. The initial contamination dose was found to be 1.8×10^8 colony-forming-units (cfu)/200 microliter (ul).

Each surface was contaminated with 200 ul dose taken from the original broth. The surfaces were

then left to dry for few minutes, and stored afterwards in covered tissue culture multiwell plates, at both room (18-20°C) and 4°C temperatures.

- C. Bacteriological methods: The strain of E. coli used in the experiment was checked for its purity using appropriate biochemical tests (6). The sensitivity to different antibiotics was also done before and after each sampling period.

At each storage period, every surface was dropped into brain heart-infusion and also MacConkey broths, incubated overnight at 37°C and then subcultured onto both MacConkey and tryptic soy agars, and incubated at 37°C for 18-24 hr. Results were recorded as positive bacterial growth, followed by biochemical identification, done according to Cowan (6).

RESULTS

Table 1 shows that E. coli was recovered from wooden surfaces after 40 days of storage at both 4°C and room temperature, compared to polystyrene and glass surfaces, where E. coli could not be detected after 10 days of storage at both temperatures.

On the other hand, E. coli was not recovered at all from metal surfaces stored at 4°C, but it was recovered after 1 day and 3 days of storage at room temperature. Results showed also that there were no significant differences between any of the media used for recovery or for enrichment of the organisms.

Table 1: Results of the recovery of E. coli from artificially contaminated surfaces stored at two temperatures.

Surface used"	Storage at 4°C		Storage at room temperature (RT)					
	1day	3days	10days	40days	1day	3days	10days	40days
Glass (G)	+	+	+	-	+	+	+	-
Metal (M)	-	-	-	-	+	+	-	-
Polystyrene (P)	+	+	+	-	+	+	+	-
Wood (W)	+	+	+	+	+	+	+	+

" Original contamination dose of E. coli (0114:K90) was 1.8×10^5 colony-forming-units (cfu) per 200 microliter (ul).

' was 18-20°C

Positive growth was scored on both MacConkey and BHI agar media.

DISCUSSION

The results (Table 1) showed clearly that E. coli tendency live on wooden surfaces was comparatively better than that on of the other surfaces used in this experiment. That might be mostly due to the higher content of starch in wood, in addition to humidity absorption and cellulose contents, and that is very readily utilized by bacteria like E. coli (7).

However, polystyrene and glass surfaces were nevertheless also good for survival of E. coli (Table 1), if compared to metal surface, where the organisms did not survive at all at 4°C storage temperature, but survival was demonstrated for only 1 and 3 days at room temperature (18-20°C). This phenomenon needs further investigation, for the purpose of checking whether wooden surfaces are prone to bacterial contamination in a way rendering them as sources of infection for long periods and threaten public health, when compared to metal surfaces. It might be that the attachment of E. coli, in particular, is much approved to a better performance on wooden surfaces than that on metallic surfaces. Other, equally-important organisms might or might not share the same properties as E. coli used in this experiment; or they may even survive longer depending on the temperature used. That needs further investigations.

On the other hand, metal surfaces used in this experiment contain some elements in their structure, like nickel, mercury, zinc etc. which might exert some toxic effect on the bacteria in close contact with them (7).

Many workers discussed the phenomenon of bacterial attachment to surfaces, and conclusively, some postulates have emerged concerning sorption, i.e. a reversible phase, during which the bacterium is weakly attached to the surface, followed by a time-dependent irreversible phase, in which the bacterium is firmly attached (1).

The pH (8), and electrolyte concentration (1) of the medium can affect adhesion, and increase in temperature can alter the quantity or characteristics of the adhesive polysaccharide produced (4).

Absolom and his co-workers (9) studied the surface thermodynamics of bacterial adhesion of Staph. aureus, Staph. epidermidis, E. coli, and List. monocytogenes to various polymeric surfaces. They found that the number of bacteria adhering per unit surface area correlated well with the thermodynamic predictions. They suggested that their data could be used to determine the surface tension of different bacterial species. They didn't study the duration of survival of the organisms they used, nor did they check the phenotypic changes expected on those organisms in relation to adhesion versus duration.

Our work showed a clear effect of temperature upon survival of E. coli on metal surface (Table 1). Such effect need further investigation, since factors playing a role in such phenomenon are at the mean time beyond the scope of our knowledge; hence, a series of experiments shall follow in the future taking many different factors into consideration, regarding the pattern of survival of different bacterial species on various inanimate surfaces.

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بقاء أنواع جرثومية متنوعة حية على سطوح
صلبة مختلفة مخزونة في ظروف مختلفة
(I) بقاء جرثومة الـ E. coli حية على سطوح صلبة
مختلفة مخزونة في درجتي حرارة مختلفتين

عامر سليم رحيم العبيدي
فرع الاحياء المجهرية، كلية الطب البيطري، جامعة بغداد

الخلاصة

تمت دراسة قابلية جرثومة الـ (E. coli) على البقاء حية على سطوح مختلفة شملت الزجاج والخشب والبولي ستايرين والمعدن، حيث جرى خزن السطوح الملوثة في درجتي حرارة 4 مئوية وفي الغرفة (18-20 م) وأخذت نماذج (باستخدام مسحات معقمة) من كل سطح بعد يوم واحد وثلاثة وعشرة ايام وكذلك بعد اربعون يوما من الخزن، وزرعت على اوساط زرعية مختلفة للتأكد من نمو الجراثيم، وقد اظهرت الجراثيم نموا ايجابيا من المسحات المأخوذة من سطوح الزجاج والبولي ستايرين المخزونة لمدة يوم واحد وكذلك 3 و 10 ايام في كلا درجتي الحرارة، وكذلك عزلت من الخشب في كافة فترات الخزن أي لغاية 40 يوما وفي كلا الدرجتين. أما من المعدن فقد عزلت الجراثيم بعد يوم واحد و 3 ايام من السطوح المخزونة في درجة حرارة الغرفة، ولم تعزل نهائيا من السطوح المخزونة في 4 م.