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SURVIVAL OF SOME BACTERIAL SPECIES NO METAL AND CARTON SURFACES STORED AT ROOM TEMPERATURE FOR DIFFERENT PERIODS OF TIME.

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SUMMARY

A total of 14 different bacterial species were monitored for their capabilities in survival on each of metal (coins) and carton surfaces. Species used were: B. subtilis, Br. abortus, Coryn. pyogenes, E. coli Kl. pneumoniae, L. monocytogenes, Past. multocida, Pr. vulgaris, Ps. aeruginosa, Sal. typhi, Staph. aureus, Staph. epidermidis, Strept. equi and strept. zooepidemicus. Results showed that all species were recovered from carton surfaces after 7 days of storage at room temperature (18-24 °c), and only B. subtilis was detected from the same surface after 14 days of storage at the same temperature. But from metal surfaces only Staph. epidermidis was recovered after 7 days of storage.

INTRODUCTION

The adhesion of bacteria to inanimate surfaces ddpends on attractive forces between the tow surfaces[1]. At the same time, repulsive forces can occur which may offset the attractive interaction or even inhibit adhesion. Most bacteria have a net negative charge, so do most of solid surfaces, hence, electrostatic repulsion between such surfaces is likely to occur[2].

Several studies have been made concerning the survival of bacteria on solid surfaces. Dyer & Maxcy[3] studied the fate of E. coli & Sal. typhimurium in a food film on stainless steel at 5°C. They found that after 24hr., 99.9% of the cells died during the test period and

and 90% of the survivors were injured. The microenvironment and time for incubation deterimine. whether bacteria grow, sustain injury or die [4,5].

A common approach to studies of factors affecting survival of bacteria on inanimate surfaces has been to incubate contaminated surfaces at room temperatuer in away to simulate imperfect cleaning of food processing or storage equipment, and the interval of storage between use. Accordingly, this work has been undertaken using pathogenic bacterial species to determine the differences in patterns of their survival no matal and carton surfaces.

MATERIALS & METAHODS

A- Surfaces used: Metal (in the form of popular 25 fils coins) and carton peices (cut from boxes used for transport of meat chops), were used; 28 peices from each type of surface were sterilized in the autoclave at 121 °c for 15 min., and left to dry for 15 min. All the surfaces were then stored separately in sterile screw-capped containers until used.

B- Contamination of surfaces: Each type of surface was contaminated individually with one species of the following bacteria: B. subtilis, Br. abortus, Cory. pyogenes, E. coli, Kl. pneumoniae, L. monocytogenes, past. multocida, Pr. vulgaris, Ps. aeruginosa, Staph. aureus, Staph. epidermidis, Sal. typhi, Strept. equi and Sterpt. zooepidemicus. Every species was grown overnight at 37°C on 5% sheep blood agar under aerobic conditions, with the exception of Br. abortus which was grown in a candle jar for 3 days before use.

Few colonies from the growth of every species were transferred to individual ~10ml amounts of tryptic soy broths and incubated at 37°C prior to contamination of the surfaces. Bacterial species used were identified according to the criteria described by Cowan[6]. Ten-fold dilutions from every grown

species were made in buffered peptone water, and the counts were done according to Miles et al. method[7]. 100ul drops from the broth of every specie were layed in duplicate onto each metal (25 fils coin) and carton ($2x2cm^2$) surfaces. Contaminated pieces were stored individually in wells in Falcon flasks, then covered and stored in the dark.

C- Sampling procedures: After 7 and 14 days of storage, contaminated surfaces (from each type) representing every bacterial specie used were droped individually in 10 ul tryptic soy broths, and incubated overnight at 37°C or until turbid, followed by straking the appropriate agar medium and incubated as above (in B).

RESULTS

After 7 days of storage, all the 14 bacterial species were recovered from carton pieces, but from coins, only Staph. epidermidis was recovered (Table 1). However, after 14 days of storage, only B. subtilis was recovered from carton pieces and not from coins. The rest of the bacterial species used were not recovered at all from any of the surfaces used at this period of storage.

Staph. epidermidis did not ferment mannitol, was catalase (+ve) and coagulase (-ve). B. subtilis was motile, hydrolyzed starch and liquefied gelatin, showing oval terminal, subterminal and central spores (Cowan 1977).

Table 1: Survival of different bacterial species on metal and carton surfaces.

Bacterial species*	7 days storage		14 days storage	
	Carton	Metal	Carton	Metal
1. Bacillus subtilis	+	–	+	–
2. Brucella abortus	+	–	–	–
3. Corynebacterium equi	+	–	–	–
4. Escherichia coli	+	–	–	–
5. Klebsiella pneumoniae	+	–	–	–
6. Listeria monocytogenes	+	–	–	–
7. Pasteurella multocida	+	–	–	–
8. Proteus vulgaris	+	–	–	–
9. Pseudomonas aeruginosa	+	–	–	–
10. Salmonella typhi	+	–	–	–
11. Staphylococcus aureus	+	–	–	–
12. Staph. epidermidis	+	+	–	–
13. Streptoacoccus equi	+	–	–	–
14. Strept. zooepidemicus	+	–	–	–

* Contamination dose for every species ranged between 1.6 and 2.0 x 10^{9} cfu per 100ul drops.

DISCUSSION

In orecious experiments, it had been shown that E. coli was able to survive for 10 days on glass and polystyrene surfaces stored at either 4°C or at room temperature (18-20°C). and also on wooden surfaces for 40 days, but showed a positive survival on metal surfaces for only 3 days of storage at room temperature[8]. This could also be the case in this experiment where E. coli was able to survive on carton and not on metal surface after 7 days of storage at room temperature. It is worth to mention here that the presence of certain elements in metal consistency, like zinc or nickel or iron etc., that could be toxic to such bacteria[8].

An exception was Staph. epidermidis which showed a successful survival on metal up to 7 days of storage, and that could be attributed most probably to the mode of living of such organisms, being common commensals of external surfaces, like skin etc.[9]

With regard to survival for 14 days or probably more, only B. subtilis did so, but only on carton surfaces, Experience with such organisms reveals that the structure of its cell wall might enable it to withstand adverse conditions, like surviving on a solid surface with depletion from nutrients[10]

Carton and metal surfaces are routinely used for many purposes related to public health practice, hence this study and other forthcoming ones shall add much information to our knowledge regarding better hygienic measures, because it should always be remembered that injured bacterial cells may regain normally if offerred a favorable environment[3].

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بقاء بعض انواع من البكتريا حيه على سطوح معدنية وكارتونية مخزونة في درجة حرارة الغرفة لفترات زمنية مختلفة

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الخلاصة

جرت دراسة قابلية ١٤ نوعا من البكتريا المرضية على البقاء حية على كل من سطحي المعدن (بهيئة نقود) والكارتون. وقد شملت البكتريا المستخدمة في التجربة : B. subtilis Kl. pneumoniae, E. coli, Coryn. pyogenes, Br. abortus Pr. vulgaris, Past. multocida, List. monocytogenes, Staph.epidermidis, Staph.aureus, Sal.typhi, Ps.aeruginosa, Strept. zooepidemicus, Strept. equi.

اظهرت النتائج بأن كل الانواع عزلت من سطوح الكارتون بعد ٧ أيام من الخزن بدرجة حرارة الغرفة (١٨-٢٠م) وبعد نفس الفترة ايضا عزلت الـ Staph epidermids فقط من المعدن.

أما بعد ١٤ يوماً من الخزن، فقد عزلت الـ B. subtilis فقط من الكارتون ولم تعزل اي من الانواع الاخرى من أي سطح.