

The Effect of Sodium Saccharin on the Growth of Escherichia coli, Proteus, Pseudomonas aeruginosa, Staphylococcus epidermidis, Staphylococcus aureus and Enterococcus faecalis.

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ÖZET

Sodyum Sakkarin'nin Escherichia coli, Proteus spp, Pseudomonas aeruginosa, Staphylococcus epidermidis, Staphylococcus aureus ve Enterococcus faecalis'in üremesi üzerine etkisi.

Sodyum sakkarin, Escherichia coli, Proteus spp, Pseudomonas aeruginosa, Staphylococcus epidermidis, Staphylococcus aureus, Enterococcus faecalis'in üremesi üzerine etkisi.

E.coli, Proteus spp, P.aeruginosa, S.epidermidis, S.aureus ve E.faecalis'in üremesi 20-320 mg/ml Na-saccharin varlığında incelenmiştir. Üremeyi ölçmek için spektrofotometrik yöntemler kullanılmıştır. Na-saccharin'in düşük konsantrasyonlarında P.aeruginosa, S.epidermidis, S.aureus ve E.faecalis'in üremesi üzerine etki saptanamazken, E.coli ve Proteus spp üremeleri inhibe olmuştur. Yüksek konsantrasyonlarda P.aeruginosa, S.epidermidis, S.aureus ve E.faecalis dahil tüm bakteriler inhibe olmuştur.

Anahtar Kelimeler: Sodyum sakkarin, E.coli, Proteus spp, P.aeruginosa, S.epidermidis, S.aureus, E.faecalis.

SUMMARY

The Effect of Sodium Saccharin on The Growth of Escherichia coli, Proteus, Pseudomonas aeruginosa, Staphylococcus epidermidis, Staphylococcus aureus and Enterococcus faecalis.

Growth of Escherichia coli, Proteus spp, P.aeruginosa, Staphylococcus epidermidis, Staphylococcus aureus and Enterococcus faecalis were studied in the presence of 20 to 320 mg/ml Na-saccharin. Spectrophotometric methods were used as a measure for growth. At low concentrations Na-saccharin had an inhibitory effect on the growth of E.coli and Proteus spp but had no effect on the growth of P.aeruginosa, S.epidermidis, S.aureus and E.faecalis. On the other hand, at higher concentrations Na saccharin inhibited the growth of all bacterial species including P.aeruginosa, S.epidermidis, S.aureus and E.faecalis.

Key Words : Sodium Saccarin, E.coli Proteus spp, P.aeruginosa, S.epidermidis, S.aureus, E.faecalis.

INTRODUCTION

Saccharin has been used as a non-nutritive sweetener agent in food. Some authors suggested that it has some antibacterial properties and this activity was first shown in vitro by using serotype of Steptococcus mutants (1). Then in a further study, Linke determined the inhibitory effect of saccharin on the growth of Gram-positive and Gram-negative rods and cocci from the human oral cavity (2).

In this study, we aimed to detect the inhibitory effect

of Na-saccharin on the growth of Escherichia coli, Proteus spp, Pseudomonas aeruginosa, Staphylococcus epidermidis, Staphylococcus aureus and Enterococcus faecalis by using various concentrations of Na-saccharin.

MATERIALS AND METHODS

Bacterial strains

All bacterial strains (E.coli ATCC 11229, Proteus spp ATCC 14153, P.aeruginosa ATCC 25619-27853, S.epidermidis ATCC 12228, S.aureus ATCC 25923, E.faecalis ATCC 29212) were obtained from Department of Clinical Microbiology, Medical Faculty, Marmara University.

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Growth of Bacteria

All bacterial strains were maintained in a complex medium of Mueller Hinton Broth (MHB) (Oxoid, CM405) at pH 7,2.

Na-saccharin inhibition studies

A stock solution containing 9,6 g Na-saccharin (Sigma, 5-1002) was prepared in 15 ml MHB and sterilized by passing through the membrane filter (Millipore, Millex-GS, 0,45 μ m). The following dilutions 320, 160, 80, 40 and 20 mg ml⁻¹ were prepared by using the broth according to the NCCLS M7-A3 methods (3-5). For each Na-saccharin concentration, 1 ml of bacterial suspension with turbidity of 0.5 mefarland was added to the 1 ml MHB at pH 7,2 including Na-saccharin. All the tubes were incubated at 37 °C in a aerobic incubator. After 24 hours, the absorbance of each tubes as a measure for growth was determined at 530 nm by using Basuch & Lomb, Spectronic 20.

RESULTS

Effect of Na-saccharin on bacterial growth

As indicated in Figure 1, *E.coli* and *Proteus* spp were inhibited by Na-saccharin. A more significant inhibition was seen with the higher Na-saccharin concentration. At low Na-saccharin concentrations no inhibitory effect was observed on the growth of *P.aeruginosa* (Figure 1), *S.epidermidis*, *S.aureus* and *E.faecalis* (Figure 2). However at low Na-saccharin concentrations, growth of these strains was inhibited. At low Na-saccharin concentrations, an obvious inhibitory effect on growth of *P.aeruginosa*, *S.epidermidis*, *S.aureus* and *E.faecalis* was significantly increased.

The absorbance values of all bacterial strains slightly decreased according to the Na-saccharin concentrations after 24 h of incubation at 37°C.

DISCUSSION

On one hand, saccharin serves as an artificial

sweetener and on the other hand it exhibits strong antibacterial properties (1). Linke determined the inhibitory effect of saccharin on growth of Gram-positive cocci, especially for oral streptococci which were strongly inhibited by saccharin (2). Some authors in different studies showed that all the Gram-positive rods, i.e. *Actinomyces viscosus*, *Lactobacillus acidophilus*, *Bacillus subtilis* and *Corynebacterium diphtheriae*, and the Gram-positive cocci, i.e. *Streptococcus* spp, *S.aureus* and *Micrococcus lutene* were significantly inhibited by saccharin, especially at higher concentrations. While the Gram-negative cocci, i.e. *Veillonella* spp and *Neisseria sicca* were strongly inhibited by all the test saccharin concentrations, the Gram-negative bacilli exhibited little inhibition (6).

The mechanism of the inhibitory effect of saccharin on Gram-positive cocci is only partially known. In cell free extracts of *Streptococcus* mutants, saccharin inhibited glycolytic enzymes. In the presence of 10⁻² M Na-saccharin, the glycolytic enzymes like hexokinase, glyceraldehyde-3-phosphate dehydrogenase, phosphoglycerate mutase and pyruvate kinase were inhibited (7).

Another study indicated that saccharin was found to inhibit the glucose-6-phosphate phosphohydrolase and glucase-1-phosphate phosphotransferase

Figure 1 : Effect of Na-saccharin on growth of Gram negative bacteria (mean values were obtained from 3 experiments)

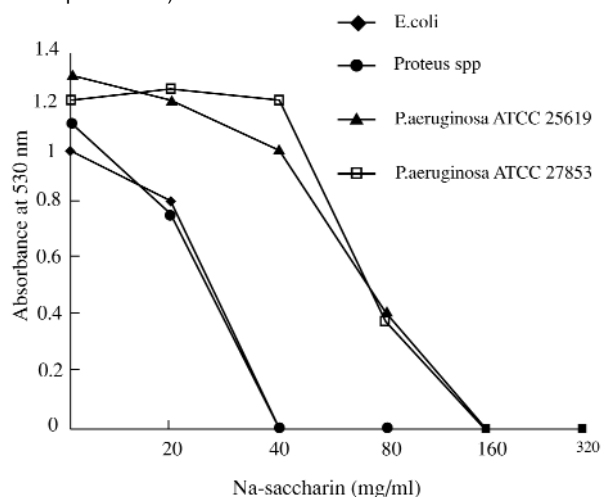
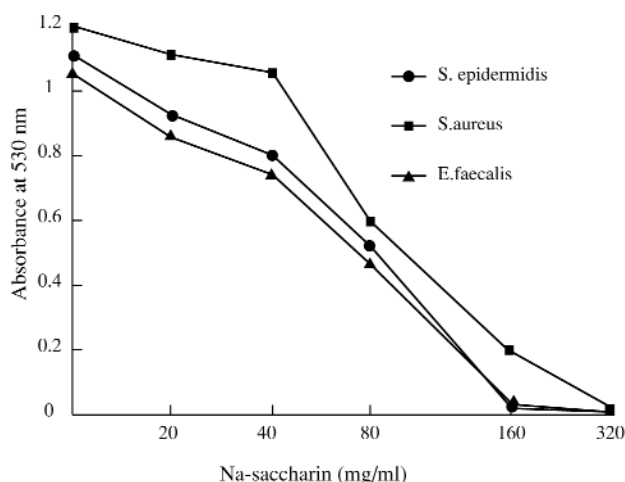


Figure 2 : Effect of Na-saccharin on growth of Gram (positive) bacteria (mean values were obtained from 3 experiments)



activities of beef liver microsomal glucose-6-phosphatase (8).

In our study, we detected the inhibitory effect of Na-saccharin on growth of *E. coli*, *Proteus* spp, *P. aeruginosa*, *S. epidermidis*, *S. aureus* and *E. faecalis* by using various concentrations of Na-saccharin. But further research is needed to clarify this inhibitory mechanism and also how to use the inhibitory effect of saccharin for food hygiene, especially in sugar-supplemented foods, as an antibacterial agent.

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