Polyurethane Elastomer As a Possible Resilient Material for Denture Protheses: A Microbiological Evaluation

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Polyurethane Elastomer As a Possible Resilient Material for Denture Protheses: A Microbiological Evaluation

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A polyurethane elastomer was microbiologically evaluated in vitro for its potential use in resilient denture liners. Specimens were immersed in suspensions of ten selected oral microorganisms; this was followed by viable cell counts at intervals during a 16-week period. Results indicated that the polyurethane neither supported the growth of the organisms nor was it degraded.

The preservation of residual oral structures, the maintenance of oral health, and the ability of patients to wear denture prostheses comfortably are among the prime objectives in rehabilitation procedures.

Patients with chronic soreness and irritation from dentures present an extremely difficult problem for prosthodontic treatment. This condition can arise from bruxism, atrophic or unfavorable alveolar ridges, or as a result of an adverse health condition.[1–3] To remedy this condition, soft lining materials have been used with varying degrees of success in certain selected clinical situations. The reason for using resilient liners is to replace the missing resilient tissue layers covering the residual ridge with a similar layer on the denture base. This reduces the direct shock of mastication on the atrophied area of support and also results in a more uniform distribution of occlusal pressure over the ridge.

Resilient materials that have been used generally can be grouped into natural rubber, plasticized polyvinyl resin, methyl methacrylate copolymer, and more recent silicone products. However, none of these materials meet all the essential requirements that an ideal material should have.[4–7] One of the serious problems associated with a widely used silicone product, such as Silastic 390,[a] is the growth of *Candida* organisms on and within the material when it is used in some patients.[8–12] The fungal growth destroys the surface quality of the material and causes irritation of the oral tissues. Additional reported problems associated with the use of resilient materials are lack of abrasive resistance, poor adhesion to the hard denture base, discoloration, lack of permanent resiliency, and difficulty in adjustment and repair.[11,13,14]

Research on polyurethane[15,16] for its potential use in dentistry is an area yet to be explored. Its dental application, a recent event, originated in the search for a better material for extraoral prostheses. At the Mayo Clinic and Mayo Foundation, Rochester, Minn, a polyurethane elastomer[b] has been successfully used in the fabrication of various facial prostheses.[17] One of its greatest assets probably lies in the fact that it can be processed with varying degrees of resiliency to suit a particular clinical requirement. Although polyurethane has never been used intraorally, results from preliminary tests of its physical properties suggested its feasibility as a resilient lining material for denture prostheses.[18]

The purpose of the present in vitro study was to perform a microbiological evaluation of a polyurethane elastomer for potential use as resilient denture liner. The study consisted of two parts: (1) to determine if the polyurethane, in the absence of extrinsic nutrients, could support the growth of ten selected isolates of oral microorganisms; these microorganisms consisted of four *Candida* species from invasion sites of Silastic 390 denture liners[19] and two common oral bacteria, *Staphylococcus epidermidis* and *Neisseria pharyngitis* (although the literature does not report bacterial invasion of previous resilient materials); and (2) to determine if the polyurethane, in a nutrient me-

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[a] Dow Corning Corp., Midland, Mich.

[b] Dermathane 100, Medical Industrial Polymer Sciences, Inc., Brookfield, Wis.

dium, could be invaded by the organisms, and if it had any effect on microbial growth.

Materials and Methods

The following materials were used: polyurethane elastomer (66 each of nonpigmented and pigmented specimens); cultures of *Candida albicans* (isolates 1 to 5), *C krusei, C tropicalis, C paropsilosis, S epidermidis, N pharyngitis;* and culture media: phosphate-buffered, 0.85% sodium chloride solution, with a pH of 7.2,[c] phosphate-buffered distilled water, with a pH of 7.2,[c] Sabouraud's dextrose broth and agar plates,[d] and Trypticase soy broth and agar plates with 5% sheep blood.[e]

EXPERIMENT 1.—The polyurethane specimens were processed according to the manufacturer's instructions[20] to yield a Shore A hardness value of 25 to 30. An aluminum mold was used to ensure surface smoothness and to control the dimensions of the specimens (Fig 1). The processed specimens were washed with soap and tap water and autoclaved at 121 C for 25 minutes.

For each *Candida* isolate, a series of nine culture tubes, each containing 14.85 ml of sterile sodium chloride solution, was set up. Similar series were set up for the bacteria, except that each tube contained 14.5 ml of sterile distilled water. Three nonpigmented and three pigmented polyurethane specimens were placed in the first six tubes, while the remaining three tubes served as controls. The *Candida* cells from Sabouraud's dextrose agar plates and the bacteria from Trypticase soy agar were transferred and washed three times in saline and water, respectively, to prevent inclusion of medium. The *Candida* was standardized in a spectrophotometer[f] to yield a suspension of 1×10^7 cells/ml, and 0.15 ml of this suspension was pipetted into each culture tube to give a final concentration of 1×10^5 cells/ml. The tubes were loosely capped and incubated at an optimal temperature of 30 C. The bacteria were standardized with a MacFarland spectrophotometric standard[g] to yield a suspension of approximately 3×10^8 cells/ml, and 0.5 ml of this suspension was pipetted into each culture tube to yield a final concentration of 1×10^7 cells/ml. The tubes were incubated at 37 C.

The effect of polyurethane on the viability of the organisms was determined by quantitation of the number of viable cells in those tubes containing the specimens and in

[c] Prepared in laboratory.
[d] Lab-Tek, Division of Miles Laboratory, Inc., Naperville, Ill.
[e] Baltimore Biological Laboratories, Cockeysville, Md.
[f] Bausch and Lomb, Spectronic 20, Rochester, NY.
[g] Kostner and Company, W. Ger.

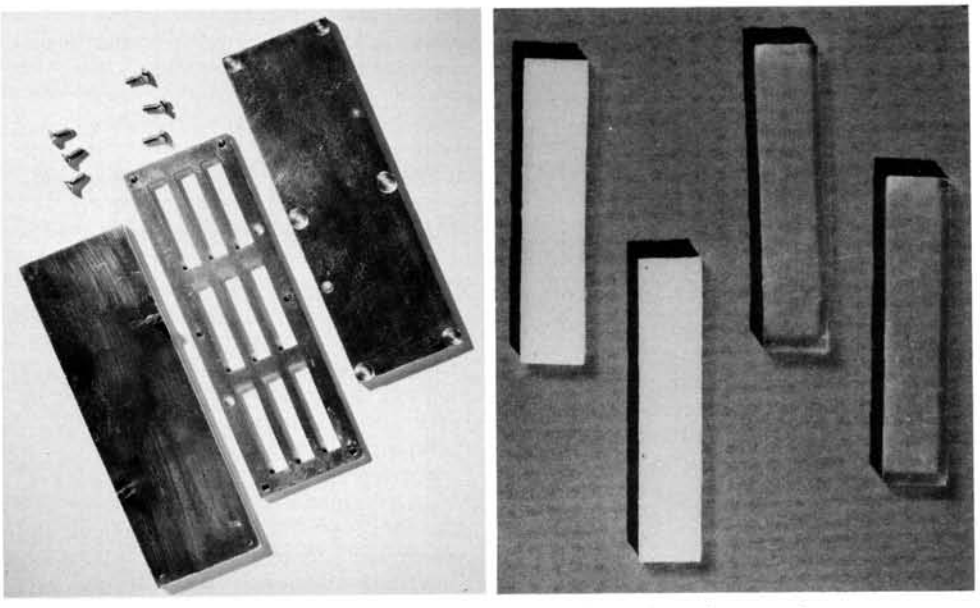

FIG 1.—*Left,* three-piece design aluminum mold in which polyurethane specimens were processed. *Right,* each finished specimen measured $3.81 \times 1.27 \times 0.32$ cm.

the control tubes at specific intervals. The number of viable *Candida* cells in each tube was determined at intervals of 1, 2, 3, 5, 7, 10, 13, and 16 weeks, and the bacteria at 6, 12, 24, 48, and 72 hours, by a method of serial dilution (Fig 2).

EXPERIMENT 2.—The method was basically similar to that in experiment 1 and the same organisms were used; however, Sabouraud's dextrose and Trypticase soy broth were used in place of saline and water. Both the *Candida* and bacteria organisms were standardized to a final concentration of 1×10^4 cells/ml. The number of viable *Candida* cells in each culture tube was determined on day 2 and at intervals of 1, 2, 3, 6, 9, 12, and 16 weeks, and the bacteria at days 1 and 3, and 1, 2, 5, 8, 12, and 16 weeks, by a method of serial dilutions.

At the end of the 16 weeks, the polyurethane specimens were removed from the tubes, rinsed with water, and allowed to air-dry. They were then placed in 10% Formalin for 36 hours, and air-dried again. The specimens were stained with 0.5% crystal violet for two minutes, rinsed with water, and air-dried. The surface quality of the specimens was examined both visually and microscopically by viewing thin sections made freehand with a single-edge razor blade.

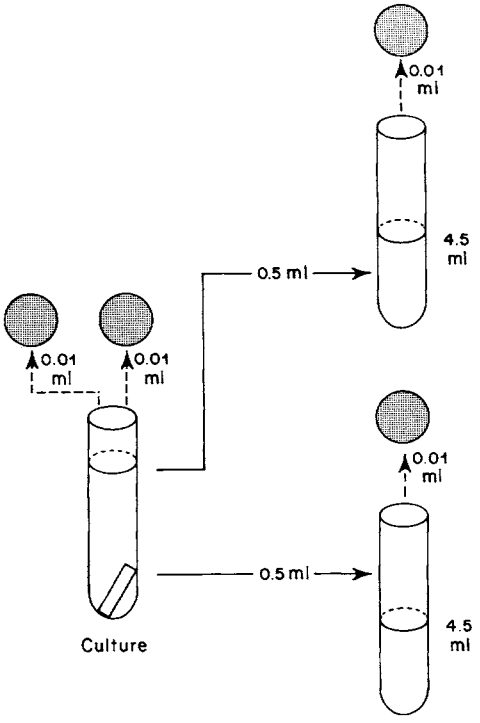

FIG 2.—Schema for colony count of yeast cultures in saline medium. From culture, 0.01 ml was transferred to each of two Sabouraud dextrose agar plates; 0.5 ml from culture was transferred to each of two culture tubes containing 4.5 ml of sterile saline. Again, 0.01 ml from these diluted suspensions was transferred onto another two agar plates. Plates were incubated at 30 C and number of colonies was determined by colony counter.

Results

The effects of polyurethane on the viability of the microorganisms were expressed graphically by plotting the logarithmic number of the viable cells per milliliter determined at the selected intervals against the time.

Figures 3 to 5 are graphic representations of the mean viable cell counts of *C albicans* (isolates 1 and 5) and *S epidermidis,* in a nonnutrient medium (saline and water) containing the polyurethane specimens, compared with the mean control counts. Little deviation was observed in the pattern of counts obtained in the presence of the polyurethane and those of the control. This was true with all the test organisms. Therefore, in a medium essentially free of nutrients, the polyurethane apparently did not support the growth of the organisms. A one-

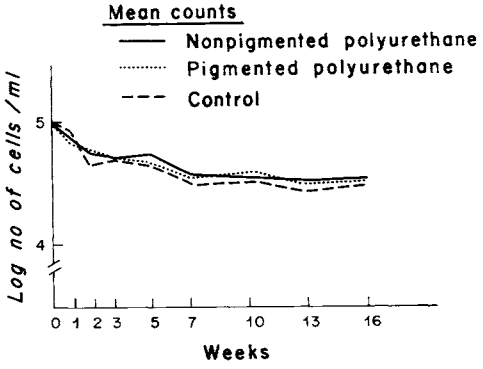

FIG 3.—Viable cell counts of *C albicans* (isolate 1) in saline medium. From initial inoculum of 1×10^5 (log 5.00) cells/ml, cell counts exhibited gradual decrease during 16-week period. No significant deviation was noted between counts in presence of polyurethane and those of control.

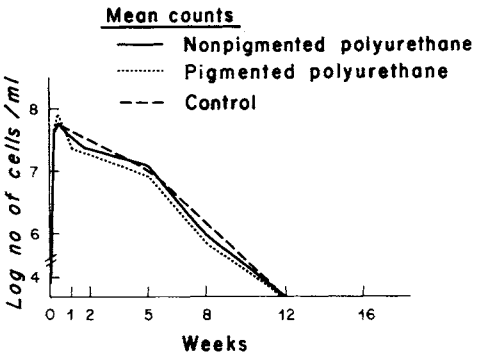

FIG 4.—Viable cell counts of *C albicans* (isolate 5) in saline medium. More rapid decrease in counts was noted; however, difference was insignificant in pattern of counts between those in presence of polyurethane and those of control.

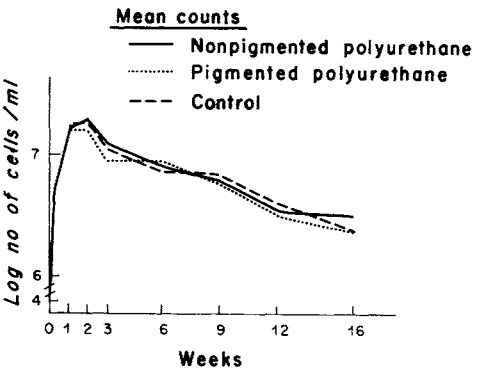

FIG 6.—Viable cell counts of *C albicans* (isolate 3) in Sabouraud's dextrose broth. From initial inoculum of 1×10^4 (log 4.00) cells/ml, rapid initial proliferation of cells was followed by gradual decrease in counts. Slight difference was noted between counts in presence of polyurethane and those of control.

way analysis of the variance of mean colony-forming units did not show any significant difference in mean counts in the nonpigmented or the pigmented polyurethane and the control. The only exception in which a statistical difference ($P < 0.01$) was found was between the *Staphylococcus* mean counts in the presence of pigmented polyurethane and that of the control at the 48-hour interval. This was probably the result of an experimental error since the following 72-hour interval did not show any viable cells with either the pigmented polyurethane or the control.

Figures 6 to 8 are graphic representations of the mean colony-forming units of *C albicans* (isolate 3), *C paropsilosis*, and *N pharyngitis* in a broth medium. Again, slight if any deviation in the pattern of counts was noted between those in polyurethane and the control. This was true with all of the test organisms. A one-way analysis of the variance did not show a significant difference in the mean counts between the polyurethane and the control. Visual and microscopic examinations of polyurethane sections were without evidence of invasion by the organisms nor was there any apparent effect on the specimens by cell metabolites or by the media alone. The surface remained smooth and intact (Figs 9–11).

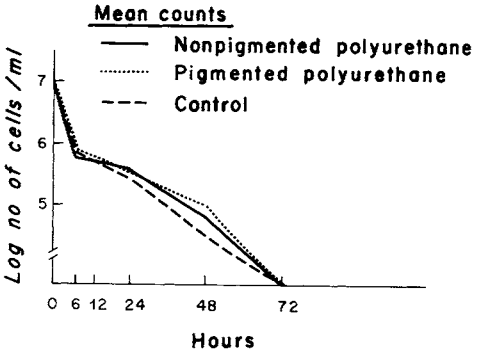

FIG 5.—Viable cell counts of *S epidermidis* in distilled water. From initial inoculum of 1×10^7 (log 7.00) cells/ml, there was abrupt fall in counts. No viable counts were registered at 72-hour interval.

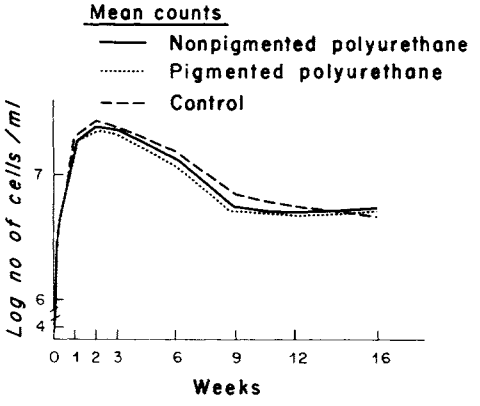

FIG 7.—Viable cell counts of *C paropsilosis* in Sabouraud's dextrose broth. Insignificant difference was noted in pattern of counts.

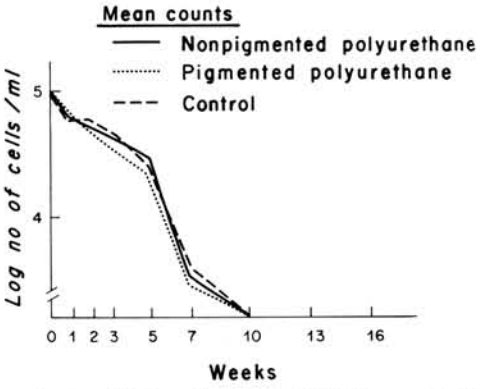

FIG 8.—Viable cell counts of *N pharyngitis* in Trypticase soy broth. Again, no significant difference was noted in pattern of counts.

Discussion

Studies on the subject of resilient lining materials for denture prostheses have been numerous. One of the problems associated with the use of the material is the growth of *Candida* organisms on and within the material, resulting in its degradation and subsequent irritation to oral tissues (Fig 12).

The literature contains only three reports on attempts made to determine if denture lining material can, in fact, be a nutrient source for the propagation of certain oral microorganisms. Love,[21] searching for visual growth of colonies of *C albicans* on Silastic, reported that both Silastic 390 and Silastic 616 supported the growth of *C albicans* without additional nutrients. This finding, however, was not verified by Masella.[19] Using a

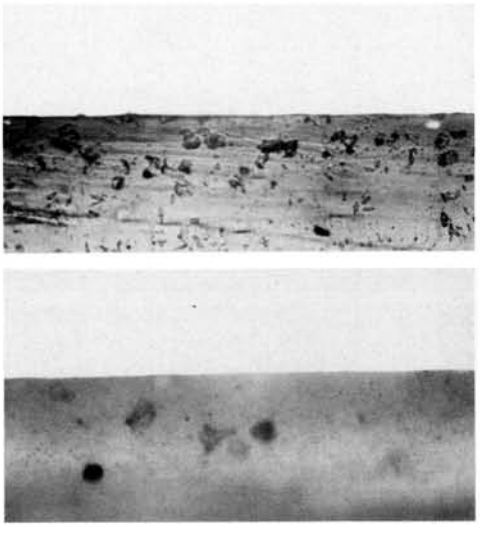

FIG 10.—No invasion of nonpigmented (*top*) and pigmented (*bottom*) polyurethane by *S epidermidis* is evident (0.5% crystal violet; mag ×210).

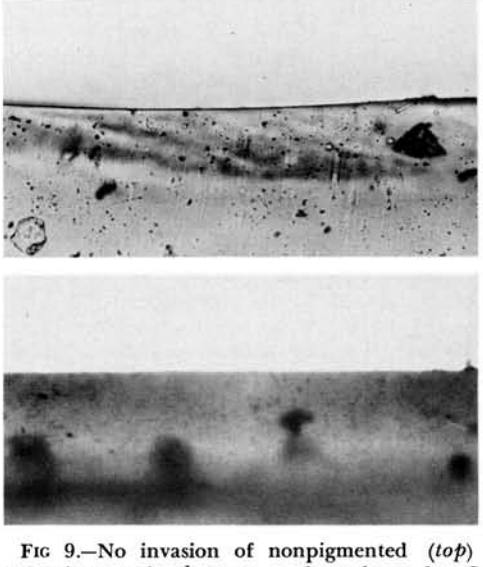

FIG 9.—No invasion of nonpigmented (*top*) and pigmented (*bottom*) polyurethane by *C albicans* (isolate 1) is evident (0.5% crystal violet; mag ×210).

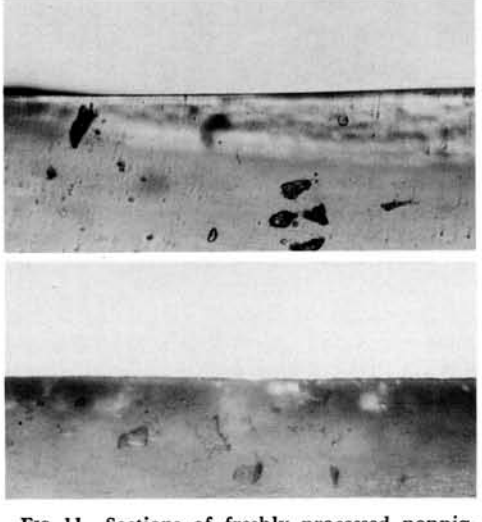

FIG 11.—Sections of freshly processed nonpigmented (*top*) and pigmented (*bottom*) polyurethane. No significant difference was found in overall surface quality between these control specimens and that of test specimens.

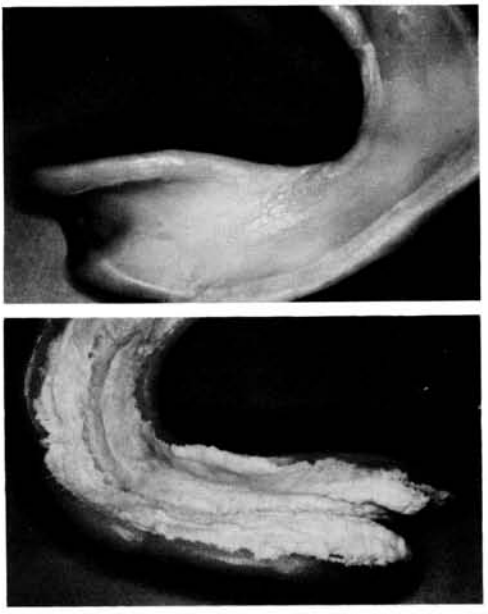

FIG 12.—*Top,* Freshly processed Silastic 390 denture liner. *Bottom,* Resilient liner that has been worn for some time. There is significant loss of surface integrity of liner.

different method of inoculation, he reported that Silastic 390 did not support the growth of *C albicans.* Nevertheless, both authors agreed that Silastic was susceptible to invasion by *C albicans* in the presence of nutrients, and this resulted in its deterioration. Williamson,[22] using a method of surface viable counts, indicated that, during a five-day period, silicone materials tested would not support the growth of *C albicans.* He indicated that Molloplast B[g] might even have an inhibitory effect on the organisms in saline medium, although this effect was negated later when a small amount of saliva was introduced.

As stated previously, we attempted to answer two basic questions: (1) Will polyurethane support the growth of the ten selected oral microorganisms? and (2) In the presence of nutrients, will polyurethane be affected by the organisms, resulting in its degradation?

An analysis of the results in experiment 1 indicated that, since essentially no cell growth was observed in the presence of polyurethane, it seemed unlikely that the polyurethane possessed any nutritional value to support the growth of the organisms. No significant difference was noted in the results obtained with nonpigmented and with pigmented polyurethane. This can be accounted for by the fact that the basic composition of these substances is the same and that the addition of pigments did not alter their behavior. Although they both showed similar resistance to fungal and bacterial invasion, similarity in their physical properties and clinical behavior still remains to be shown. The use of buffered media prevented any significant variations in the pH of the media. This eliminated the effect that such variation might have on the stimulation or inhibition of the growth of the organisms.

In experiment 2, in which nutrient media were used, results did not show significant differences between the mean colony-forming units from the media containing the polyurethane and the control. No inhibitory or other effect of the polyurethane was evident, thus further suggesting the inertness of the material. The organisms in both the tubes containing the specimens and the control exhibited a rapid initial proliferation until maximal colony-forming units were reached. Subsequent counts showed a gradual decrease in the number of colony-forming units, a change that apparently occurred as the available nutrients diminished, the toxic waste accumulated, and the pH in the media changed. The difference in the results between the nonpigmented and pigmented polyurethane was not significant.

Visual and microscopic examination of polyurethane sections did not show evidence of invasion by the organisms and there was no apparent effect on the polyurethane by cell metabolites or the media alone. In all, the surface remained smooth and intact. This is in direct contrast to the results reported by Love[21] and Masella[19] when they used Silastic. Both reported in vitro invasion of the Silastic by *C albicans* when extrinsic nutrients were available, and this finding is significant. The apparent inertness and the ability of polyurethane to retain surface smoothness represent distinct advantages of this material over Silastic. It is also a step closer to solving the problem of tissue irritation induced by the disruption of the surface integrity of the lining material by oral microorganisms.

As there are many organisms other than *Candida* present in the mouth, the next logical step in the evaluation of the polyurethane is the clinical assessment of the mate-

rial in the oral environment followed by periodic examinations of the material and a study of its physical properties and behavior. The present study, nevertheless, showed the possibility of the polyurethane as a potential resilient denture liner.

Conclusions

A microbiological study was done to evaluate, in vitro, the suitability of a polyurethane elastomer as a resilient denture liner. The study consisted of two parts. One was to determine if the nonpigmented or pigmented polyurethane, in the absence of extrinsic nutrients, would support the growth of ten selected oral microorganisms (eight *Candida* isolates from invasion sites of Silastic denture liners and two bacteria, *S epidermidis* and *N pharyngitis*). The other was designed to determine if (1) the polyurethane, in a nutrient environment, could be invaded by the organisms, and (2) the polyurethane had any effects on microbial growth. The method involved quantitatively evaluating the effect of polyurethane on suspension of yeasts and bacteria and subsequent microscopic examination of sections of polyurethane for evidence of deterioration or invasion.

We concluded that the polyurethane elastomer did not support the growth of the ten selected oral microorganisms. In a nutrient environment, no inhibitory or other effects of the polyurethane on the colony-forming units were observed. Fungal or bacterial invasion of the polyurethane specimens was not evident. There was no apparent effect on the polyurethane by cell metabolites or the media alone, and the surface of the specimens, whether nonpigmented or pigmented, remained smooth and intact. These findings justify a further in-depth evaluation of the physical properties and a clinical assessment of the polyurethane elastomer to determine its suitability as a resilient denture liner.

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