J Stoma 2016; 69, 4: 443-447
DOI: 10.5604/00114553.1225382

ORIGINAL ARTICLE

Influence of different types of plant extracts on the *Streptococcus sobrinus* culture

Wpływ różnych wyciągów pochodzenia roślinnego na hodowlę *Streptococcus sorbinus*

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Abstract

Introduction. *Microbiological status of children and adolescents with the establishment of the prevalence of certain kinds of microorganisms is the etiological factor of occurrence and progression of various dental diseases.* ***Aim of the study.*** *To determine the effect of different types of plant extracts on the culture of Streptococcus sobrinus.* ***Materials and methods.*** *The study examined the effect of plant extracts on the culture of Streptococcus sobrinus, cultured in the oral cavity fluid in 146 children aged 3-8 years of different ethnic groups with varying degree of caries who resided permanently in biogeochemical conditions with shortage of fluoride and iodine.* ***Results.*** *Absence of growth of Str. sobrinus when exposed to cranberry, red currant, cherry and jostaberry extracts was demonstrated. Several dilutions of extracts were studied. No microorganism growth when exposed to the plum extract in dilutions of 1:1 and 1:2, 1:8, 1:16 was observed; in dilutions of 1:4 and 1:6 significant growth inhibition was reported when compared to controls ($1\pm0.15\times10^2$, $1\pm0.1\times10^2$; 0; $p<0.05$).* ***Conclusions.*** *The most effective plant extracts against Str. Sobrinus are extracts of cranberry, redcurrant, cherry and jostaberry in that they can be recommended in your diet in comprehensive prevention of dental caries in children as no effective means of endogenous drug prevention in preschool and primary school age has so far been introduced.*

Streszczenie

Wstęp. *Mikrobiologiczny status dzieci i młodzieży, u których określono występowanie niektórych gatunków mikroorganizmów, jest czynnikiem etiologicznym występowania i rozwoju różnych chorób zębów.* ***Cel pracy.*** *Ustalenie wpływu różnych rodzajów ekstraktów roślinnych na hodowlę Streptococcus sobrinus.* ***Materiał i metody.*** *Zbadano wpływ różnych rodzajów ekstraktów roślinnych na hodowlę Streptococcus sobrinus w płynie pobranym z jamy ustnej 146 dzieci w wieku 3-8 lat zróżnicowanych etnicznie z różnym poziomem próchnicy, na stałe zamieszkujących rejony ubogie w fluorki i jod.* ***Wyniki.*** *Wykazano brak wzrostu Str. sobrinus po ekspozycji na ekstrakty żurawiny, czerwonej porzeczki, czereśni i porzeczkoagrestu. Zbadano reakcję na różne rozcieńczenia ekstraktów. Nie odnotowano wzrostu mikroorganizmów przy ekspozycji na ekstrakt śliwki w rozcieńczeniu 1:1 i 1:2, 1:8, 1:16; w rozcieńczeniu 1:4 i 1:6 zaobserwowano znaczne zahamowanie wzrostu w porównaniu z grupą kontrolną ($1\pm0,15\times10^2$, $1\pm0,1\times10^2$; 0; $p<0,05$).* ***Wnioski.*** *Ekstrakty żurawiny, czerwonej porzeczki, wiśni i porzeczkoagrestu okazały się najbardziej skuteczne i mogą być zalecane w ogólnej profilaktyce próchnicy u dzieci, gdyż jak dotąd nie zaproponowano żadnego endogennego sposobu zapobiegania próchnicy u dzieci w wieku przedszkolnym i szkoły podstawowej.*

KEYWORDS:
children dental caries, hygiene habits, nutrition habits, Antibacterial activity, *Streptococcus sobrinus*

HASŁA INDEKSOWE:
próchnica zębów u dzieci, nawyki higieniczne, nawyki żywieniowe, działania przeciwbakteryjne, *Sobrinus Streptococcus*

Introduction

In the available sources of scientific literature much attention is paid to the study of microbiological status of children and adolescents with the establishment of prevalence of certain species of microorganisms as etiological factors of occurrence and progression of various dental diseases.[1-7] Epidemiological studies among children of the Transcarpathian region revealed that it is essential microbial oral coenosis as a factor in the origin and progression of the disease, which differs significantly among children of different ethnic groups.[1-3,8-11] It was established that the presence of oral *Str. sobrinus* in number 1h104KUO, dysbiotic changes in the mouth, accompanied by a decrease in the concentration of Bifidobacterium less than ± 2×103 1×102 CFU, reduction or complete disappearance of *Lactobacillus salivarius*, appearance of pathogenic *Escherichia coli* are all recognized factors in the formation of cavities. The progression of tooth decay, leading to the development of sub- and decompensated form of caries is characterized by the following parameters: the elevated count of *Str. sobrinus* 1h104KUO, increasing the total number of microbial mouth to 1h109 and as well as a large number of pathogens, including *Pseudomonadas aeruginosa, Escherichia coli, S. aureus, Proteus vulgaris*.[12]

The evaluation of oral microbial environment showed that in 100% of children of mixed ethnic groups *Str. sobrinus* is the only acid-forming microorganism, while in all the children of the Romany ethnic group *Streptococcus mutans* was identified. This *Str. sobrinus* culture is resistant to almost all standard antibiotics, except for antibiotics reserve; culture of *Streptococcus mutans* that is sowing in children of Romany ethnic group is sensitive to 25 of 34 antibiotics. The obtained data indicate significant aggressiveness of *Str. sobrinus* as evidenced by the increasing activity of caries in children are sowing in this type of *Streptococcus*.[12] This prompted the search for new methods of inhibiting the growth of acid-producing *streptococci* and the results are presented in this paper.

The aim of the study is to determine the influence of different types of plant extracts on the culture of *Streptococcus sobrinus*.

Materials and Methods

A clinical study to establish the reliability of this assumption was conducted. Oral and immunological status of 146 children aged 3-8 years of different ethnic groups with varying degrees of caries activity residing permanently in conditions biogeochemically deficit in fluoride and iodine. Of the 146 children surveyed, 37 – Romany ethnic group accounted for 25.3%, 109 of other ethnicities – 74.7%. The children were divided into groups depending on the activity of caries 35 – compensated with caries (24.0%), 26 – with subcompensated (17.8%), 35 – decompensated caries (24.0%) and 50 healthy children, 25 Roma ethnic groups and 25 other ethnic groups that constituted the control group.

To establish the level of microbial environment oral fluids and microbial soft plaque in the cervical region were collected. Hung conducted on the nutritional substrate: - meat peptonnyy selective agar, semi-selective medium, semi-liquid environment, environment Endo, bismuth-sulfite agar, enterococcus agar, laktobak agar, bifidum, Bifidobacterium Agar for cultivation.

Results and Discussion

In the study screening of extracts of edible plants and drinks that have a strong antibacterial effect was conducted. For the study, extracts of coffee, tea, cranberry, pomegranate peel, plum, plums, red currants, blueberries, cherries and jostaberry were selected.

The protocol of the study:

1. Preparation of the bacteria suspension concentration 1.5h108kl/ml.
2. Preparing the spot with the studied extracts – (it is necessary to prepare a series of dilutions of the tested substances in 96-well dilution spot is studied extracts of 1: 1, 1: 2, 1: 4, 1: 6, 1: 8 and 1:16.
3. Preparation of tea extract, dry powdered plant material extracted with distilled water. Extraction time 24 hours. The extract was filtered using sterile membrane filters.

Table 1. The results of the effect of different types of plant extracts on *Streptococcus sobrinus* culture

Cultivation extract	1:1	1:2	1:4	1:6	1:8	1:16
Cranberry	no growth	no growth	no growth	no growth	no growth	no growth
Red currant	no growth	no growth	no growth	no growth	no growth	no growth
Cherry	no growth	no growth	no growth	no growth	no growth	no growth
Jostaberry	no growth	no growth	no growth	no growth	no growth	no growth
Cherry plum	no growth	no growth	$(1\pm0.15)*10^2$	$(1\pm0.1)*10^2$	no growth	no growth
Pomegranate rind	no growth	no growth	$(1\pm0.3)*10^2$	$(1\pm0.2)*10^2$	no growth	no growth
Coffee	$(1.5\pm0.1)*10^8$	$(1.5\pm0.2)*10^8$	$(1.5\pm0.1)*10^8$	$(1.5\pm0.3)*10^8$	$(1.5\pm0.4)*10^8$	$(1.5\pm0.4)*10^8$
Tea	$(1\pm0.3)*10^2$	$(1\pm0.25)*10^3$	$(1\pm0.22)*10^5$	$(1\pm0.15)*10^5$	$(1\pm0.1)*10^7$	$(1\pm0.15)*10^7$
Plum	$(1\pm0.3)*10^2$	$(1\pm0.4)*10^2$	$(1\pm0.3)*10^2$	$(1\pm0.25)*10^2$	$(1\pm0.2)*10^2$	$(1\pm0.2)*10^2$
Blueberry	$(1\pm0.2)*10^2$	$(1\pm0.5)*10^2$	$(1\pm0.4)*10^4$	$(1\pm0.3)*10^4$	$(1\pm0.3)*10^2$	$(1\pm0.1)*10^2$

4. Preparation of coffee extract: fine ground coffee with added boiling water and salt. Salt is used to extract caffeine from coffee, which is a biologically active substance. Extraction time 24 hours. The extract was filtered using sterile membrane filters.
5. Preparation of pomegranate peel extract: powdered pomegranate skin soaked in water, left for 6 hours. After 6 hours defending this extract is boiled in a water bath. The extract was filtered using sterile membrane filters. When making cranberry extract, plant material is crushed and filtered using sterile membrane filters.
6. Preparation of extracts of plum, plums, red currants, blueberries, cherries, jostaberry, was carried out by methanol extraction. To get rid of methanol, which would have affected the results of the study, extracts were subjected to vacuum vidpartsi. Add cells to each well of the test extracts and incubated at 37°C in a thermostat on the floating surface for 15 minutes. After incubation, each extract was titrated for analysis, so that the concentration of m/v decreased from 108 to 102 and once in reverse order (from 102 to 108) were grown in 5 ml culture medium in petri dishes. After 24 hours the cultivation of Petri dishes sown with droplets of bacteria 370S take into account the number of colonies that grew and dilution are depressed CFU.Serial dilutions method. Preparation of the bacterial suspension 0.5 mF. In break-back trap conduct research investigated dilution of the extract in distilled water 1: 1 1: 2; 1: 4; 1: 6; 1: 8; 1:16. In each well diluted extract was studied by adding 0.01 ml bacterial suspension of *Str. sobrinus*.

The results of the effect of different types of plant extracts on *Str. sobrinus* culture are shown in Table 1. the impact of different types of plant extracts was assessed. There was no growth of *Str. sorbinus* culture when exposed to cranberry, red currant, cherry and jostaberry dilutions. No microorganism growth was observed when exposed to plum extract in dilutions of 1:1 and 1:2, 1:8, 1:16, and in dilutions of 1:4 and 1:6 there is a significant growth inhibition compared to controls ($1 \pm 0.15 \times 10^2$, $1 \pm 0.1 \times 10^2$; 0; $p < 0.05$).

In the study of pomegranate peel extract, no growth of *Str. sobrinus* was found in dilutions of 1:1, 1:2, 1:8, 1:16, and in dilutions of 1:4 and 1:6 inhibition of growth was negligible when compared with the controls ($1 \pm 0.3 \times 10^2$, $1 \pm 0.2 \times 10^2$; 0; $p < 0.05$).

Extracts of such foodstuffs as coffee, tea, blueberries and words in all tested dilutions do not demonstrate 100% inhibition of *Str. Sorbinus* growth. However, significant inhibition of bacterial growth was noted by the action of bilberry extract in dilutions of 1:1, 1:2, 1:8, 1:16 ($1 \pm 0.3 \times 10^2$; $1 \pm 0.2 \times 10^2$; $1 \pm 0.3 \times 10^2$, $1 \pm 0.1 \times 10^2$; 0; $p<0.05$); plum extract in all dilutions ($1 \pm 0.3 \times 10^2$; $1 \pm 0.4 \times 10^2$; $1 \pm 0.3 \times 10^2$, $1 \pm 0.25 \times 10^2$, $1 \pm 0.2 \times 10^2$, $1 \pm 0.2 \times 10^2$; 0; $p<0.05$). Slight growth inhibition was observed in the extract of tea than in dilution of 1:1, which revealed significant inhibition ($1 \pm 0.3 \times 10^2$; 0; $p<0.05$); no coffee extract showed antibacterial effects on *Str. Sobrinus* growth.

Conclusions

In assessing the impact of different types of plant extracts on Str. sobrinus culture no growth was found when exposed to cranberry, red currant, cherry and jostaberry extracts in all dilutions. No microorganism growth when exposed to plum extract in dilutions of 1: 1 and 1: 2, 1: 8, 1:16 was noted, and in dilutions of 1:4 and 1:6 there was a significant growth inhibition compared to controls ($1 \pm 0.15 \times 10^2$, $1 \pm 0.1 \times 10^2$; 0; $p<0.05$).

In the study of pomegranate peel extract there was no growth of *Str. sobrinus* in dilutions of 1:1, 1:2, 1:8, 1:16, and in dilutions of 1:4 and 1:6 the inhibition of growth was negligible compared to the controls ($1 \pm 0.3 \times 10^2$, $1 \pm 0.2 \times 10^2$; 0; $p<0.05$).

Extracts of foodstuffs such as coffee, tea, blueberries and words in all tested dilutions did not block the growth of *Str. sobrinus* in 100%, but significant inhibition of bacterial growth was noted by the action of the bilberry extract in dilutions of 1:1, 1:2, 1:8, 1:16 ($1 \pm 0.3 \times 10^2$; $1 \pm 0.2 \times 10^2$; $1 \pm 0.3 \times 10^2$; $1 \pm 0.1 \times 10^2$; 0; $p<0.05$); plum extract in all dilutions ($1 \pm 0.3 \times 10^2$; $1 \pm 0.4 \times 10^2$; $1 \pm 0.3 \times 10^2$, $1 \pm 0.25 \times 10^2$, $1 \pm 0.2 \times 10^2$, $1 \pm 0.2 \times 10^2$; 0; $p<0.05$); a slight growth inhibition was observed for tea extract whereas dilution of 1:1 demonstrated significant inhibition ($1 \pm 0.3 \times 10^2$; 0; $p<0.05$). Coffee extract showed no antibacterial action on the growth of *Str. sobrinus.*

Thus, the most effective plant extracts against *Str. sobrinus* are extracts of cranberry, red currant, cherry and jostaberry, allowing recommendation of these plants in the overall prevention of caries in children serving as an effective preventive endogenous tool in preschool and early school age.

Acknowledgements

The study is part of research projects of the department of pediatric dentistry dental faculty SIHE (State Institution of High Education) "Uzhhorod National University" "Improving the provision of dental care to children who live in conditions biogeochemically deficit in fluoride and iodine" (state registration number 0114U004123), which was performed on the basis of agreements on scientific and technical cooperation with the University of Presov (Slovakia, 2006), the Institute of Dentistry NMAPE named *P.L.Shupyk* (2009), with the Clinic of Maxillofacial Surgery of *P.J. Safarik* University (Slovakia, 2010), University of Debrecen (Hungary, 2010).

A survey of children was carried out after the parents or guardians gave their consent.Microbiological and immunological studies were conducted in microbiological laboratories of the Transcarpathian Branch of Microbiologists of Ukraine named after SM Winogradsky (Chairman of the Company, Dr., Professor *N. Boyko*).

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Received: 27th June 2016
Accepted: 23rd Ocober 2016