ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN — POLONIA
VOL. LVII, N 2, 140 SECTIO D 2002

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# *The activity of dental caries in students of the Faculty of Stomatology examined by using microbiological and biochemical tests – Dentocult LB*

One of the major factors conditioning the development of dental caries is the presence of cariogenic oral flora. An essential role in this process is attributed to *Streptococcus mutans* and *Lactobacillus acidofilus* (3). The former is responsible for the onset of dental decay, the latter for its further development and secondary caries (10, 11, 12). The development of dental caries or its inhibition depends on the effects of pathological and protective factors on the tooth tissue (11). It is believed that with the progress of decay, the bacterial aerobic flora changes into the relatively anaerobic one. Evaluating the patient's exposure to dental caries, the numbers of *Lactobacillus acidofilus* should be taken into account (4, 10). An analysis of the saliva level of these bacteria may be performed using the Dentocult LB test.

The aim of the study is to examine the relation between the numbers of *Lactobacillus acidofilus* and the dental state expressed by the numbers of DMF, $DMF_s$ and $D_s$ among the dentistry students.

## MATERIAL AND METHODS

The examined group included 66 students of the Faculty of Stomatology aged 20-25 years (30 men and 36 women). The patients subjected to the tests did not take any antibiotics during the month preceding our examinations and did not rinse the oral cavities with an antiseptic mouthwash. The production of saliva was stimulated by chewing paraffin tablets and the saliva samples were collected to disposable test-tubes. A ready-made Dentocult LB medium was covered with saliva on both sides and placed in the thermostat for 4 days at 37°C to culture the colonies of *Lactobacillus*. After incubation, the bacterio-

logic data were determined by evaluating the colony density on agar plates and by comparing them with the standard results provided by the producer. The values of the Dentocult LB tests were presented in 5 classes (4, 5, 7): Class 0:0 CFU/ml, Class 1: $10^3$ CFU/ml, Class 2:$10^4$ CFU/ml, Class 3:$10^5$ CFU/ml, Class 4:$10^6$ CFU/ml.

The clinical examinations were carried out using the mirror and the probe in artificial light. The findings were expressed as the numbers of DMF, $DMF_s$ and $D_s$. Then the individual elements were analysed, particularly $D_s$ (the number of surfaces with active caries). The results were statistically analysed to examine the relation between Dentocult LB and DMF, $DMF_s$, $D_s$. The DMF values were divided into two categories: DMF≤14 and DMF>14; the $DMF_s$ values – into $DMF_s$≤ 25 and >25 while $D_s$ – into $D_s$≤ 3 and >3. The statistical analysis was performed using the chi-square test. The risk of opinion error was 5%. The results were compiled in the tables and illustrated graphically.

Table 1. Correlation between *Lactobacillus acidofilus* class and DMF of the students

| LB class | Number of examined people | DMF | | Total |
|---|---|---|---|---|
| | | < = 14 | > 14 | |
| 1 | Number of examined people | 13 | 5 | 18 |
| | % of examined people | 72.2 | 27.8 | 100.0 |
| | % of whole examined group | 19.7 | 7.6 | 27.3 |
| 2 | Number of examined people | 9 | 10 | 19 |
| | % of examined people | 47.4 | 52.6 | 100.0 |
| | % of whole examined group | 13.6 | 15.2 | 28.8 |
| 3 | Number of examined people | 13 | 9 | 22 |
| | % of examined people | 59.1 | 40.9 | 100.0 |
| | % of whole examined group | 19.7 | 13.6 | 33.3 |
| 4 | Number of examined people | 3 | 4 | 7 |
| | % of examined people | 42.9 | 57.1 | 100.0 |
| | % of whole examined group | 4.5 | 6.1 | 10.6 |
| Total | Number of examined people | 38 | 28 | 66 |
| | % of examined people | 57.6 | 42.4 | 100.0 |
| | % of whole examined group | 57.6 | 42.4 | 100.0 |

Value of Dentocult LB test.
Class 1: $10^3$ CFU/ml; class 2: $10^4$ CFU/ml; class 3: $10^5$ CFU/ml; class 4: $10^6$ CFU/ml.
$Chi^2 = 3.033$; $p = 0.387$

Table 2. Correlation between *Lactobacillus acidofilus* class and $DMF_s$

| LB class | Number of examined people | $DMF_s$ | | Total |
|---|---|---|---|---|
| | | <= 25 | > 25 | |
| 1 | Number of examined people | 13 | 5 | 18 |
| | % of examined people | 72.2 | 27.8 | 100.0 |
| | % of whole examined group | 19.7 | 7.6 | 27.3 |
| 2 | Number of examined people | 9 | 10 | 19 |
| | % of examined people | 47.4 | 52.6 | 100.0 |
| | % of whole examined group | 13.6 | 15.2 | 28.8 |
| 3 | Number of examined people | 13 | 9 | 22 |
| | % of examined people | 59.1 | 40.9 | 100.0 |
| | % of whole examined group | 19.7 | 13.6 | 33.3 |
| 4 | Number of examined people | 4 | 3 | 7 |
| | % of examined people | 57.1 | 42.9 | 100.0 |
| | % of whole examined group | 6.1 | 4.5 | 10.6 |
| Total | Number of examined people | 39 | 27 | 66 |
| | % of examined people | 59.1 | 40.9 | 100.0 |
| | % of whole examined group | 59.1 | 40.9 | 100.0 |

Value of Dentocult LB test.

Class 1: $10^3$ CFU/ml, class 2: $10^4$ CFU/ml; class 3: $10^5$ CFU/ml; class 4: $10^6$ CFU/ml. $Chi^2 = 2.375$; $p = 0.498$.

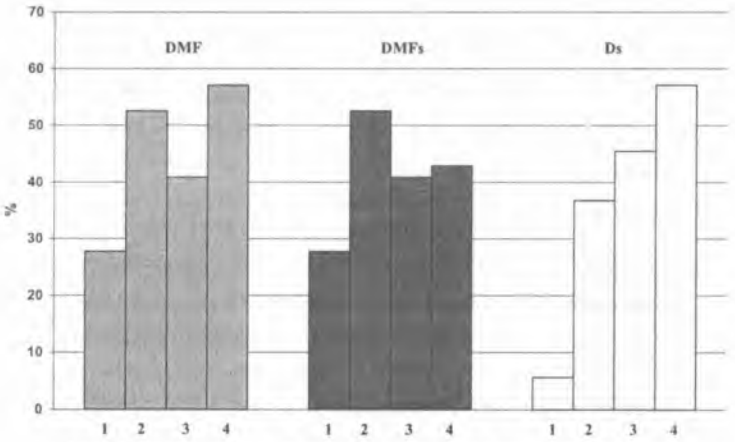


Fig. 1. Relationship between caries activity and LB class

Table 3. Correlation between *Lactobacillus acidofilus* and Ds

| LB class | Number of examined people | Ds | | Total |
|---|---|---|---|---|
| | | <= 3 | > 3 | |
| 1 | Number of examined people | 17 | 1 | 18 |
| | % of examined people | 94.4 | 5.6 | 100.0 |
| | % of whole examined group | 25.8 | 1.5 | 27.3 |
| 2 | Number of examined people | 12 | 7 | 19 |
| | % of examined people | 63.2 | 36.8 | 100.0 |
| | % of whole examined group | 18.2 | 10.6 | 28.8 |
| 3 | Number of examined people | 12 | 10 | 22 |
| | % of examined people | 54.5 | 45.5 | 100.0 |
| | % of whole examined group | 18.2 | 15.2 | 33.3 |
| 4 | Number of examined people | 3 | 4 | 7 |
| | % of examined people | 42.9 | 57.1 | 100.0 |
| | % of whole examined group | 4.5 | 6.1 | 10.6 |
| Total | Number of examined people | 44 | 22 | 66 |
| | % of examined people | 66.7 | 33.3 | 100.0 |
| | % of whole examined group | 66.7 | 33.3 | 100.0 |

Value of Dentocult LB test

Class 1: $10^3$ CFU/ml, class 2: $10^4$ CFU/ml; class 3: $10^5$ CFU/ml; class 4: $10^6$ CFU/ml. $Chi^2 = 9.569$; $p = 0.022$

## RESULTS AND DISCUSSION

The analysis of the results showed a statistical correlation between $D_s$ and LB (Table 3; $p=0.022$); i.e. $D_s>3$ was observed in 5.6% of the patients with LB $10^3$ (class 1), in 36.8% with LB $10^4$ (class 2), in 45.5% with LB $10^5$ (class 3) and in 57.1% with LB $10^6$ (class 4). These findings reveal that the higher the LB values the higher the $D_s$ values.

No significant correlation was found among DMF (Table 1; $p=0.387$), $DMF_s$ (Table 2; $p=0.241$) and LB. However, LB affects DMF and $DMF_s$.

The microbiological tests are relevant accessory tools to define individual caries risk. It is thought that high caries risk occurs when the amount of *Lactobacillus* is $10^6$ CFU/1 ml of saliva (7). Barańska-Gachowska et al. in their studies among 12-year-old children also found a statistically significant correlation between the amount of saliva LB and $D_s$. The authors report that the high amount of saliva LB was observed in 52.6% of the

children with low caries intensity (the average number of $D_s$=0.79, ranges from 0 to 4), in 90.9% of those with medium caries intensity (the average number of $D_s$=5.0, ranges from 2 to 12) and in all the patients with high caries intensity (the average number of $D_s$=16.75, ranges from 10 to 40) (1). Similarly to our findings, these results show a strong relation between LB and $D_s$. Llena-Puy et al. examining 167 children aged 12-13 by using the same tests (Dentocult LB) found a statistical relation between the index of caries and the number of bacteria (9). This relation between the intensity of caries and the number of LB was further confirmed by Barańska-Gachowska et al. who examined the effects of sanation of the oral cavity (removing the active caries foci) on the amount of saliva SM and LB. They noted that high numbers of *Lactobacillus* in some patients after sanation was likely to indicate the presence of clinically undetectable active caries cavities on the contact surfaces and therefore full sanation should be performed only after X-ray examinations (2). Radlińska et al. had similar results confirming a significant correlation between the intensity of caries and LB. The average numbers of $D_s$ were statistically significantly higher in children with high *Lactobacillus* numbers compared to those with low LB numbers (12). The number of *Lactobacillus* in the Dentocult LB test indirectly indicates the amount of carbohydrates ingested. If there is a discrepancy between the clinical state and diet data reported by patients, "the truth" may be easily found out thanks to this test; more than 10,000CFU of *Lactobacillus* in 1 ml of saliva (classes 3 and 4) prove high carbohydrate consumption. High LB numbers are also found when the retentive places develop (cavities, marginal defects in fillings, orthodontic devices) (4, 5, 7, 13). However, dental caries will occur only when the number of saliva *Streptococcus mutans* is high. The results of the studies concerning the relation between dental caries, pH and bacterial flora of the dental plaque performed by Lingstrom et al. indicated that increased activity of caries was related to elevated amount of dental plaque with acidogenic and acidophilic bacteria (8). Van Ruyven et al. found out that the plaque covering the foci of acute caries ("white spot caries") consisted mainly of the "low-pH" bacteria (min. pH<4.4) (13). However, the effects of other factors conditioning caries, such as the composition of the dental plaque, quality of enamel, saliva parameters (lysozyme, immunoglobulins, peroxidase, lactoferrin, number and activity of granulocytes) cannot be neglected (1, 6, 12, 13).

## CONCLUSIONS

1. A statistically significant correlation between LB and $D_s$ was found in the 20-25 age group of patients.

2. The influence of LB on DMF and $DMF_s$ values was observed in the examined group.

3. The microbiological LB test may be useful to determine individual caries risks.

4. *Lactobacillus acidofilus* was detected in all the patients, which shows that this bacterium is commonly present in human saliva.

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2002.05.08

SUMMARY

The aim of the study was to examine the relation between the value of Dentocult LB and numbers of DMF, $DMF_s$ and $D_s$ in the 20-25 age group. Sixty-six randomly chosen students of the Faculty of Stomatology, Medical University of Lublin were included in the study. In all of them, their dental state was assessed by using average numbers of DMF, $DMF_s$ and $D_s$. In the microbiological examinations, a ready-made medium from the Dentocult LB set was used. Statistically significant, directly proportional dependence of the number of surfaces with $D_s$ active caries and the LB number was found.

Aktywność próchnicy zębów u studentów stomatologii, badana za pomocą testów mikrobiologicznych i biochemicznych Dentocult LB

Celem badań jest zbadanie związku pomiędzy wartością Dentocult LB a liczbami PUW, $PUW_p$ i $P_p$ w grupie wiekowej 20 – 25 lat. Materiał badawczy stanowiło 66 studentów oddziału stomatologii Akademii Medycznej w Lublinie. U wszystkich badanych dokonano oceny stanu uzębienia za pomocą średniej liczby PUW i $PUW_p$ oraz $P_p$. Do badań mikrobiologicznych wykorzystano gotowe podłoże wchodzące w skład zestawu Dentocult LB. Stwierdzono istotną wprost proporcjonalną zależność między ilością powierzchni z próchnicą aktywną $P_p$ a liczbą LB.