Arch. Biol. Sci., Belgrade, 62 (3), 739-746, 2010 DOI:10.2298/ABS1003739G

ADDITION TO THE METHODOLOGY OF RESEARCH INTO PERMANENT TEETH HARDNESS

SANJA GNJATO

Health Care Center Banja Luka, 78000 Banja Luka, Republic of Srpska, Bosnia-Herzegovina

Abstract: This paper examines permanent teeth hardness (microhardness) using the Vickers method. An original methodology was developed and adopted for preparing the experimental material, i.e. the cross sections into four characteristic locations on the tooth: corona dentis, cervix dentis, pars medialis radicis dentis and apex radicis dentis. A new "hardness profile" was introduced, which connects hardness and location along the cross section of the tooth. Hardness was measured 664 times on experimental cross sections with total a average hardness of 73.17 HV according to Vickers and a standard deviation of 55.68 HV. The derived descriptive statistical indicators of tooth hardness were calculated for equivalent cross sections, individual teeth, teeth groups and tooth localizations. Two algorithms were developed for determining the rank of tooth hardness – one for estimating the rank of arithmetic mean of the hardness of the cross sections of the teeth, and the other for estimating the rank of hardness for individual teeth.

Keywords: Tooth hardness, enamel, dentin, cement, hardness profile

UDC 611.314:616.314

INTRODUCTION

Hardness is a property denoting the capability of a material to resist the penetration of other objects into its surface layers. Hardness is a surface characteristic. The extent of surface distortion depends on the material itself, but also on the way the surface is distorted. In this sense, it may prove useful to determine the amount of resistance to scraping, abrasion, dents from impact and indents. According to the type of surface distortion, corresponding hardness measurement methods were created. Even though other techniques are not insignificant, the most common hardness testing technique is indentation. There are several methods of indentation hardness testing: Rockwell's, Vickers', Brinell's and Knoop's. The principle of measurement in indentation-based methods is that the surface of the material being tested is put into contact with an indentor of specified shape and dimensions, which during a specified time interval applies pressure of a specified intensity of force, after which the hardness is calculated based on the dimensions of the indentor.

Tooth hardness – previous research

In the second half of the twentieth century, there were few papers that could have been used as reference in researching permanent teeth hardness. The papers that were available can be divided into three groups. The first is the group dealing directly with testing tooth hardness with measurement methods. The second is the group of papers that establish a connection between tooth hardness and tooth microstructure, and the papers in the third group try to connect tooth decay – caries – with tooth hardness.

All available papers from the aforementioned period either state that tooth hardness is not explored in a systematic or that it is their main goal. Most papers mention or explore tooth hardness either as a benchmark for the effect being explored (cause – tooth exposure, consequence – tooth decay), or as a bio-physical characteristic which bio-materials in prosthetics and implantology must be compatible with. Exceptions are the first paper on this subject by Saunsbury and Atkinson (1953)

and one of the relatively newer papers by Hata and Hata (2000) which makes the connection and calculates the regression curve between the frequency of the tactile sensor and microhardness index by Vickers. However, this paper is limited to surface measurements of the hardness of the enamel and dentin.

Research aim and hypotheses

The aim of the research was to examine permanent teeth hardness, both for individual teeth and teeth groups, on cross-sections (crown, neck and root), covering all tooth structures (enamel, dentin, cement). The significance of researching teeth hardness is in determining the difference in hardness between structures of various histological, morphological and chemical properties, which is directly correlated with the exposure of teeth to external (physical, chemical and biochemical) influences.

The practical significance of researching tooth hardness can be seen in all branches of stomatology, especially in prosthetics, conservative stomatology, oral surgery and others. The biophysical characteristics of teeth, especially teeth hardness, require that the material has appropriate physical and chemical properties, which means that the adequate therapy in tooth treatment is determined by tooth hardness, degree of damage, tooth class etc.

Tooth hardness, from the cross-section, declines from the exterior to the interior structures, i.e. from the enamel towards the cement, and from the crown to the tip of the root. It is assumed that hardness varies between different teeth groups, which is connected to their respective functional requirements.

MATERIALS AND METHODS

The material was gathered over several years in dentists' offices. The selection of the sample was made from a pool of 100 teeth, none of which had been recently extracted. The time range from the extraction to the selection was one to five years. From the available teeth, 52 human teeth were selected. The first selection criterion for the sample was tooth quality. Only teeth with no visible defects were selected, not taking into account any damage at the microstructural level. The second selection criterion was that the teeth belonged to permanent dentition. The third selection criterion for the sample was teeth diversity (belonging to various classes). The aim was to find approximately the same number of teeth from each class. However, considering the available number of teeth and the number of teeth in the sample, it was impossible to fully achieve this aim. But still, the sample contained at least one tooth from each class, so the sample can without a doubt be considered valid and the number of units in the sample, as well as its structure, is in no way a limiting factor for the research.

Preparation of the sample

Each tooth from the sample was cut transversely at the appropriate point. In order to minimize the influence of heat on the crystalline structure of the teeth, and consequently on the hardness of the teeth, during the cutting process the material and the tools were cooled with water. This produced 52 experimental dental plates. The reference for cutting was from the perpendicular to the longitudinal axis of the tooth. The longitudinal axis of the tooth is defined as an imaginary line connecting the two furthest points of the tooth. The experimental plates, the 2-3 mm thick cross sections, were highly polished. This produced the experimental material, 52 experimental tooth plates from 15 characteristic tooth points. The further analysis shows data only for four equivalent cross-sections.

The Vickers hardness test

The Vickers hardness testing method consists of indenting a small diamond square-based pyramid into the material. Because of the brittleness of the diamonds, the indentation must be performed very carefully. The point of the pyramid is cut under a 136° angle. The indentation force is arbitrary, from 1 to 120 daN, depending on the material being tested. The indentation is then enlarged on a

blurred plate, the diagonal is measured by fine measuring tools with 1/1000 mm accuracy, and then the surface of the imprint in mm^2 and the Vickers hardness degree are calculated following the following formula:

$$HV = \frac{2F\sin\frac{136^0}{2}}{d^2} \cong 1{,}854\frac{F}{d^2}$$

where HV – denotes the Vickers hardness degree, F – denotes the indentation force [daN], and d – denotes the pyramid diagonal [mm^2].

During this research, tooth hardness was measured with the "LL Tukon" microhardness tester (see image below). This device provides high automatization of the measurement process thanks to a computer and screen which are used to automatically set and display both the size of the indentation and directly the Vickers hardness degree.

Statistical methods of determining tooth hardness

The statistical methodology applied in research can be provisionally presented in four stages:

Presentation of the research sample – picture of the sample: This section describes the sample. First the original measurements are processed, which will be given in statistical tables to give a more concise presentation. Table of original measurements: The first column shows the number of the measurement. The second column has the depth of the indentation from the hardness test. The third column gives the hardness value by Vickers (HV), and the fourth column is for the location of the indentation in micrometers. The chart (see image), represents the so-called hardness profile. The presentation of the research sample ends with a text additionally explaining the displayed variable and/or table and chart.

Data analysis – determining descriptive statistical indicators for measuring tooth hardness: The analysis primarily assumes determining derived statistical hardness indicators for equivalent

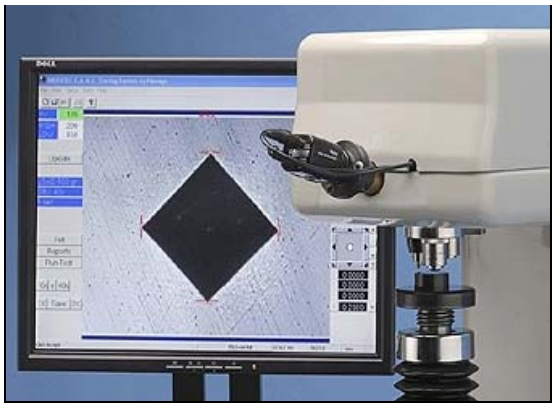

Fig. 1.

cross-sections, individual teeth, teeth groups and tooth location (up – down). The analysis also includes the so-called measurement homogenization. All results of the analysis are shown in the appropriate statistical tables.

The comparative statistical analysis of the derived statistical tooth hardness indicators. This consists of two parts: comparative analysis and testing implicit research hypotheses.

Determining tooth hardness rank: The final part of the methodological procedure for determining tooth hardness is ranking relevant hardness indicators, i.e. equivalent cross-sections, individual teeth, tooth groups and tooth location. For this purpose, ranking algorithms were developed. One is the algorithm for evaluating the rank of arithmetic means of equivalent cross-sections, and the other one is for evaluating the hardness rank for individual teeth, based on testing significant differences between the arithmetic means of individual teeth hardness.

I Algorithm hardness rank for equivalent cross-sections: $RANG_U$ ↑ ▪ $Rang(I)$, where $RANG_U$ denotes the total rank of the average hardness for equivalent tooth cross-sections, and $Rang(I)$ denotes the individual rank of the average hardness for equivalent tooth cross-sections. The criterion for selecting the hardness rank for equivalent tooth cross-sections is $\min(RANG_U)$.

II Algorithm hardness rank for individual teeth: $RANG\_SKOR_U = \sum Rang\_skor(I)$, where $RANG\_SKOR_U$ denotes the total hardness rank score for individual teeth, and $Rang\_skor(I)$ the individual hardness rank score for individual teeth. $Rang\_skor(I)$ is calculated based on the evaluation of the test result among certain teeth in the following way: if the result is of a) high statistically relevant difference, b) some statistically relevant difference or c) no statistically relevant difference, then the numbers 2, 1 or 0, respectively, are assigned to the result. The criterion for selecting the hardness rank for equivalent tooth cross-sections is $\max(RANG_U)$.

RESULTS

Descriptive statistical indicators for original tooth hardness measurements

Each series of measurements is followed by data in tables on the tooth hardness profile and the corresponding charts of the hardness profile for the tooth cross-sections depending on the location of the measurement. Tables are then given showing descriptive statistical parameters for individual measurement series. The descriptive parameters were average value, standard deviation, fifth and ninety-fifth percentile and variation coefficient.

Example: Dens incisivus primus superior, pars medialis radicis dentis

Table 1 shows that the average hardness value for the middle of the root dens incisivus primus superior is 76.06 HV with the standard deviation of 11.12 HV. It turns out that the series is homogeneous with the variation coefficient of CV=14.62%. The fact that the 5th and 95th percentiles are included in the interval encompassed by X_{sr} ± 2SD, indicates that the influence of extreme measurement values to descriptive statistical parameters is negligible. Based on the results of descriptive statistics and conclusions on the ranks of average hardness values

Table 1. Descriptive parameters of tooth hardness (dens incisivus primus superior, pars medislis radicis dentis

X_{sr}	76,06
SD	11,12
5th percentile	59,30
95th percentile	92,70
CV=SD/ X_{sr} *100	14,62

Table 2. Table showing tooth hardness profile (dens incisivus primus superior, pars medialis radicis dentis)

o.	Indentation depth (µm)	Hardness (HV)	Location (µm)
1	100	92.7	250
2	100	92.7	230
3	105	84.1	240
4	105	84.1	230
5	108	79.5	250
6	109	78	230
7	116	68.9	250
8	125	59.3	240
9	125	59.3	250
10	125	59.3	225
11	122	62.3	225
12	122	62.3	250
13	117	67.7	230
14	113	72.6	245
15	113	72.6	240
16	110	76.6	250
17	105	84.1	230
18	105	84.1	240
19	104	85.7	230
20	104	85.7	240
21	104	85.7	250

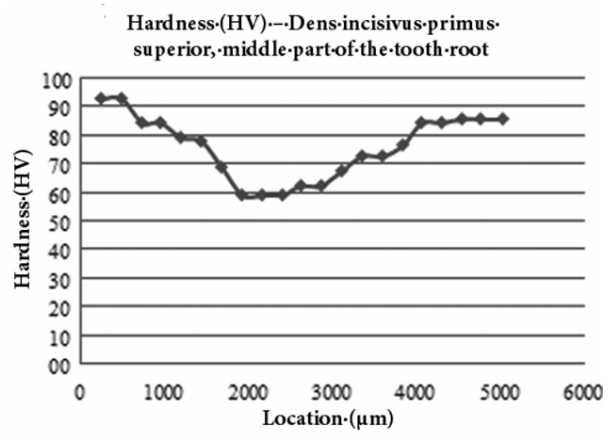

Fig. 1. Graphic chart of tooth hardness profile (dens incisivus primus superior, pars medialis radicis dentis)

Tooth hardness analysis – derived descriptive statistical indicators for tooth hardness measurement:

in equivalent cross-sections of certain tooth types, applying criteria for rank selection produces the rank of average hardness in equivalent tooth cross-sections such as: 1. Corona dentis, 2. Pars medialis radicis dentis, 3. Apex radicis dentis, 4. Cervix dentis.

Comparative statistical analysis of tooth hardness:

Comparative analysis of average hardness values in equivalent tooth cross-sections includes unreduced and unhomogenized measurements, but also reduced and homogenized measurements.

Table 2 shows that the total rank of tooth hardness for certain tooth types in homogenized measurements is no different than the total rank of hardness for certain tooth types in unhomogenized measurements, except for dens incisivus primus and dens molaris secundus which have switched places. It also shows that there is no difference in the rank of total hardness for tooth groups and tooth location between unhomogenized and homogenized measurements.

Testing the relevance of difference between arithmetic means of tooth hardness values (individually and among tooth groups)

In order for valid conclusions to be made on testing the relevance of the difference between arithmetic means of tooth hardness it is necessary to determine the rank of hardness based on the conducted tests. To evaluate the total rank of average hardness value for individual teeth we use the developed algorithms for rank evaluation based on results of mutual individual tests:

$$RANG\_SKOR_U = \sum Rang\_skor(I) \cdot$$

The $Rang\_skor(I)$ value is calculated based on the evaluation of the test result among certain teeth in the following way: if the result is of a) high statistically relevant difference, b) some statistically relevant difference or c) no statistically relevant difference, then the numbers 2, 1 or 0, respectively,

The rank of average hardness values in equivalent cross-sections according to tooth type

Equivalent cross-sections / tooth	The rank of hardness in cross-sections according to tooth type							
	Dens incisivus primus	Dens incisivus secundus	Dens caninus	Dens premolaris primus	Dens premolaris secundus	Dens molaris primus	Dens molaris secundus	Dens molaris tercius
Corona dentis	4	1	3	1	1	1	1	1
Cervix dentis	1	4	4	3	3	3	3	2
Pars medialis radicis dentis	3	3	2	2	2	2	-	3
Apex radicis dentis	2	2	1	4	4	4	2	-

Table 2. Comparative rank of certain tooth types and groups for unhomogenized and homogenized measurements

Name		Total rank	
		Unhomogenized measurements	Homogenized measurements
Tooth type	Dens incisivus primus	3	1
	Dens incisivus secundus	8	8
	Dens caninus	5	5
	Dens premolaris primus	7	7
	Dens premolaris secundus	6	6
	Dens molaris primus	2	2
	Dens molaris secundus	1	3
	Dens molaris tercius	4	4
Tooth group	Incisivus dentis	2	2
	Premolaris dentis	3	3
	Molaris dentis	1	1
Tooth group	Superior dentis	1	1
	Inferior dentis	2	2

are assigned to the result. The criterion for selecting the hardness rank for equivalent tooth cross-sections is $\max(RANG_U)$. Using the Spearman rank correlation shows that $\rho_{emp}=0,29<\rho_{gr}=0,738$ (for p=0,05) which indicates that there is no congruence in the ranks of original and homogenized measurements (p>0,05).

DISCUSSION

A total of 664 hardness measurements were performed on experimental cross-sections with total average hardness of 73.17 HV and standard deviation of 55.68 HV. The variation coefficient of 76.09% indicates that it is a highly inhomogeneous set. The so-called homogenization process was carried out, thus excluding measurement values lower than the 5^{th} and higher than the 95^{th} percentile. This process excluded extreme values and produced a homogeneous set with total average hardness of 63.76 HV and standard deviation of 14.66 HV. The homogenization process, removing 5% of the highest and 5% of the lowest hardness values, practically reduced the measurement to testing hardness of the dentin.

With individual teeth, the highest hardness of 73.90 HV belongs to the dens incisivus primus, followed by the dens molaris primus with 69.49 HV, dens molaris secundus with 66.11 HV, dens molaris tercius with 63.57 HV, dens caninus with 62.05 HV, dens premolaris secundus with 60.83 HV, dens premolaris primus with 59.25 HV and finally dens incisivus secundus with 53.02 HV. When it comes to tooth groups, the highest hardness value is in the molaris dentis with 63.73 HV, followed by the incisivus dentis with 63.33 HV and finally the premolaris dentis with 60.17 HV. Regarding tooth location, the highest hardness is that of the superior dentis with 63.48 HV and then the inferior dentis with 61.63 HV.

The hardness rank and tooth hardness value distribution by individual teeth, tooth groups and tooth location corresponds to the expected hardness model made on the basis of the physiological characteristics of permanent teeth.

Table 3. Rank score and individual teeth rank, based on testing the relevance of the differences in arithmetic means of hardness – homogenized measurements

Tooth type	Rank score	Rank
Dens incisivus primus	12	2
Dens incisivus secundus	14	1
Dens caninus	6	7
Dens premolaris primus	8	4,5,6
Dens premolaris secundus	8	4,5,6
Dens molaris primus	9	3
Dens molaris secundus	8	4,5,6
Dens molaris tercius	5	8

CONCLUSIONS

Original methodology was developed for the preparation of the experimental material – experimental plates (cross-sections) in four characteristic tooth locations, i.e. the corona dentis, cervix dentis, pars medialis radicis dentis and apex radicis dentis. Hardness was measured along the larger axis of each cross-section. This methodology implicitly determined the focal point of the hardness testing. Tooth hardness was measured in different cross-sections and different layers.

Based on the hardness measurements the so-called hardness profile was introduced and shown in a table and diagram, linking tooth hardness and location along the tooth cross-section. Along with the hardness profile, we have tables of the descriptive statistical parameters.

The derived descriptive statistical indicators of tooth hardness were calculated for equivalent cross-sections, individual teeth, tooth groups and tooth location.

An algorithm was developed for determining the tooth hardness rank for each of the observed categories, i.e. equivalent cross-sections, individual teeth, tooth groups and tooth location, and an algorithm was developed for determining hardness rank for certain tooth categories based on test results of implicit research hypotheses on the existence of a statistically relevant difference between arithmetic means of tooth hardness values. The results of the research show that the hardness rank for individual teeth derived from the original measurements and the hardness rank from the homogenized measurements differ. The Spearman rank correlation shows that there is no congruence between ranks in the original and the homogenized measurements ($p>0,05$).

REFERENCES

Caldwell R.C., Muntz M.L., Gilmore R.W., and *W. Pigman,* Microhardness studies in intact surface enamel, *J. Dent Res*, No.: 36 732-738, 1957.

Cirano F.R., Romito G.A., and *J.H. Todescan*, Determination of enamel and coronal dentin microhardness, *Braz J. Oral Sci*, Vol 2, Number 6, July/September 2003.

Featherstone J.D.B., ten Cate J.M., Shariati M., and *J. Arends,* Comparison of artificial caries-like lesions by quantitative microradiography and microhardness profiles, *Caries Res,* No.: **17**, 385-391, 1983.

Fuentes V., Toledano M., Osorio R., and *R.M Carvalho.,* Microhardness of superficial and deep sound human dentin, *J. Biomed Mat Res*, Vol **66A(4)**, 850-53, August 2003.

Fukuda K., Higashi S., Miake K., and *T. Matsui,* Studies on microhardness of the human third molar especially that of the enamel, Bull Tokyo Dent Coll **16**, 11-33, 1975.

Gaspersic D., Enamel microhardness and historical features of composite pearles of diferent size, *J. Oral Pathol Med,* **24**, 153-8, 1995.

Kodaka T., Debary K., Yamada M., and *M. Koroiwa,* Correlation between microhardness and mineral content in sound human enamel *Caries Res* **26**, 139-41, 1992.

Krage T.L., Stiefel A., Stephan B.M., Zimmer S., Lambrichts I., and *H.M. Raab,* Microhardness changes in dentine after neonatal capsaicin application, *I. Endodontic Journal,* Vol **38**, Issue 8, Page 570, August 2005.

Meredith N., Sherriff M., Setchell D.J., and *S.A Swanson.,* Measurement of microhardness and Yung's modulus of human enamel and dentine using an indentation technique, Arch Oral Biol **41,** 539-45, 1996.

Pashley D., Okabe A., and *P. Parham,* The relationship between dentin microhardness and tubule density, *Endod. Dent. Traumatol.* **1**, 176-9, 1985.

Saunsbury P., and *H.F. Atkinson,* An investigation into the hardness od human enamel, Part II, Studies of deciduous and permanent teeth, *Brit Dent, No.* **251**-**253**, 1953.

Shinji Hata, and *Hiroko Hata,* Validation of hardness measurement of deltal hard tissue using a tactile sensor: In vitro study, *Pediatric Dental Journal* **10(1),** 149-154, 2000.

ПРИЛОГ МЕТОДОЛОГИЈИ ИСТРАЖИВАЊА ТВРДОЋЕ СТАЛНИХ ЗУБА

САЊА ГЊАТО

Дом здравља, 78000 Бања Лука, Република Српска, Босна и Херцеговина

Примјеном Викерсове методе испитивана је тврдоћа (микротврдоћа) сталних зуба. Развијена је и усвојена оригинална методологија припреме експерименталног материјала, тј. попречних пресјека на четири карактеристичне локације на зубу: corona dentis, cervix dentis, pars medialis radicis dentis и apex radicis dentis. Уведен је „профил тврдоће“ који повезује тврдоћу и локализацију дуж попречног пресјека зуба. Израчунати су изведени дескриптивни статистички показатељи тврдоће зуба за: еквивалентне попречне пресјеке, појединачне зубе, зубне групе и зубну локализацију. Развијена су два алгоритма за одређивање ранга тврдоће зуба; један за оцјену ранга аритметичких средина тврдоће попречних пресјека зуба, а други за оцјену ранга тврдоће појединачног зуба.

Укупно је извршено 664 мјерења тврдоће на експерименталним попречним пресјецима са укупном просјечном тврдоћом од 73,17 HV и стандардном девијацијом од 55,68 HV. Коефицијент варијације од 76,09% говори да се ради о високо нехомогеном скупу. Извршен је поступак тзв. хомогенизације тако да су искључене вриједности мјерења које су биле мања од 5. и већа од 95. перцентила. На тај начин из разматрања су искључене екстремне вриједности и добијен је хомогени скуп са укупном просјечном тврдоћом од 63,76 HV и стандардном девијацијом од 14,66 HV. Поступком хомогенизације, уклањањем 5% највећих и 5% најмањих вриједности тврдоћа, мјерење се практично своди на испитивање тврдоће дентина.

Код појединачних зуба, највећу тврдоћу има dens incisivus primus са тврдоћом од 73,90 HV, затим слиједи dens molaris primus са 69,49 HV, dens molaris secundus са 66,11 HV, dens molaris tercius са 63,57 HV, dens caninus са 62,05 HV, dens premolaris secundus са 60,83 HV, dens premolaris primus са 59,25 HV и на крају dens incisivus secundus са 53,02 HV. Код зубних група, највећу тврдоћу имају molaris dentis са 63,73 HV, затим слиједе incisivus dentis са 63,33 HV и на крају premolaris dentis са 60,17 HV. Код зубне локализације, највећу тврдоћу имају superior dentis са 63,48 HV а затим inferior dentis са 61,63 HV.

Ранг тврдоће и дистрибуција вриједности тврдоћа зуба по појединачним зубима, зубним групама и зубној локализацији одговара очекиваном моделу тврдоћа изграђеном на основу физиолошких карактеристика сталних зуба.