

Biochemical analysis of the saliva of patients with cognitive and physical disabilities as a practical Biochemistry class strategy in the Dentistry course

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ABSTRACT

Given the importance of teaching biochemistry in dental surgeon training courses, the objective was to present a pedagogical strategy that enables the articulation of basic molecular concepts within the subject, allowing a better understanding of the complexity of oral and systemic diseases, through the biochemical analysis of saliva from patients with cognitive and physical disabilities at the Dental Assistance Center for Disabled Persons (CAOE) of the School of Dentistry, Araçatuba - São Paulo State University (UNESP). For this purpose, groups of five students analyzed the saliva collected by a dental surgeon from CAO. Immediately after collection, salivary flow, pH, and buffering capacity were determined. Subsequently, the saliva was centrifuged, fractionated, and stored at -20 °C until the time of biochemical analysis. During the practical classes, using commercial kits, the samples were tested for: total protein, α -amylase, alkaline phosphatase, uric acid, glucose, aspartate aminotransferase, calcium, and phosphorus. At the end of the school year, students presented a report contextualizing the results with the patient's oral and systemic health. Tutoring sessions with monitors and teachers helped in the interpretation of the anamnesis form and the correlations of oral and systemic clinical parameters. For incoming students, this was the first opportunity to integrate theoretical knowledge with clinical perspectives. It is concluded that the adopted strategy is viable and can benefit educators who seek alternatives that allow the integration of basic and clinical sciences for teaching Biochemistry in Dentistry.

Descriptors: Learning. Teaching. Education, Dental. Biochemistry. Saliva.

1 INTRODUCTION

Saliva has been widely used for rapid and sensitive tests, which enable the early diagnosis and monitoring of diseases¹. While this highlights the importance of teaching biochemistry to students of the dentistry course, many students face difficulties in the process of learning and contextualizing the subject²⁻⁴. This difficulty was attributed by students to the lack of practicals for demonstrating the relevance of the contents taught. Consequently, it is not clear to them that biochemistry classes provide them the molecular bases that justify the need for good dental hygiene³.

Teaching based on clinical cases has proven to effectively facilitate the learning process in many disciplines, including biochemistry. In addition to improving students' interest⁵, it makes them feel more motivated to participate as active members of the class, help in decision making, and work with the group during discussions about clinical practice. In addition, it has been proven that this type of experience encourages students to develop fundamental skills for lifelong learning⁶. This teaching methodology actively involves the students, exposes them to the scientific process when studying real cases⁷, encourages the use of information in solving problems, and facilitates the retention of knowledge and improvement in communication⁸.

The National Curricular Guidelines for undergraduate courses in dentistry in Brazil were instituted on February 19, 2002, following the CNE/CES³ resolution, defining the principles, foundations, conditions, and procedures necessary for the training of dentists in Brazil. They state that the structuring of the dentistry course should use teaching or learning methods that allow students to play an active role in the process, in addition to integrating knowledge of

the basic sciences with that of clinical sciences⁹. Within the basic training of the dentistry course, the knowledge of physiological sciences such as; biochemistry, physiology, and pharmacology stands out⁹. Training in basic sciences is intended for the student to acquire the scientific bases necessary for the clinical treatment of patients, and to develop the ability to think and analyze data¹⁰.

Transforming students into competent and attentive professionals capable of improving the oral health care of the society is one of the main objectives of dental education¹¹, and a considerable challenge for educators in the field. For the students of this course, the learning cycle is also marked by setbacks, as the process of contextualizing theoretical knowledge and integrating it into practice, learning about pathologies, and enriching their skills¹², sometimes presents with adversities, and not everyone can complete it with ease.

Because of the challenges that punctuate this teaching-learning process, for both educators and students, several support strategies have been proposed to facilitate the better acquisition of knowledge by the student, and the transmission of the contents by the teacher. In the dentistry field, it has already been found that the traditional model of expository classes has several limitations, as unidirectional communication with a lack of interaction results in low student involvement¹³. In this context, a study on teaching human physiology showed that the discussion of clinical cases in basic areas is an important tool to improve students' learning and performance¹⁴.

Dental care for patients with special needs is different, given that they have physical and/or mental changes that can be temporary or permanent¹⁵. At our university unit, the possibility of working with the saliva of these

patients proved to be viable, due to the existence of the Dental Assistance Center for Disabled Persons (CAOE), which provides care to this specific portion of the population.

Given the importance of teaching biochemistry in the training course for future dental surgeons, the objective of this pedagogical strategy is to enable the articulation of basic molecular concepts within the subject, allowing a better understanding of the complexity of oral and systemic diseases, through the biochemical analysis of saliva from patients with cognitive and disabilities.

2 EXPERIENCE REPORT

This article deals with a descriptive report of an experience applied to first-year students of FOA-UNESP Full and Night Dentistry courses. This idea is the result of discussions leveraged by students regarding the difficulty encountered by teachers of the biochemistry classes in the teaching-learning process, which may be related to the inherent complexity of the content, its high abstraction, and the wide range of origins of the information presented^{16,17}.

The course is annual, mandatory, and includes structural, metabolic, and oral biochemistry. The weekly workload is three (3) hours/class, making an annual total of 90 hours. For the basic and oral contents to be better integrated, the subjects were grouped by similar themes, enabling the establishment of correlations between theory and practical, in order for the educational objectives to be achieved. The chronological sequence of activities is shown in Table 1.

The pedagogical strategies adopted in 2019 were theoretical lectures featuring a multimedia projector and a whiteboard, and practical laboratory classes and discussion of the data obtained in them. Theoretical classes

provided the basis for an interpretative approach for the analysis of the results obtained during the practical classes. This prior knowledge was of great importance for the students during practical classes¹⁸. For practical lessons, the class guides were made available to the students in advance, containing a brief introduction that recalled concepts covered in the theoretical lectures, in addition to the instructions and description of the different steps in the development of salivary biochemical analyses.

For practical activities, each class was subdivided into groups of five students, chosen by affinity, and maintained until the end of the school year. In the first two classes, biosafety, recognition, and the correct use of laboratory materials were addressed. Correct techniques for using graduated glass pipettes and automatic pipettes were taught and practiced. In the following weeks, three class hours were reserved for training in the collection of saliva from a student volunteer from each group. In this same encounter, students learned the procedures for determining salivary flow, pH, and buffering capacity, and finished by centrifuging the saliva and fractionating the supernatant for storage. In this way, students were prepared for poster work with the saliva of CAOE patients.

On a scheduled day and time for each class (A, B, and Night), a lecture at the CAOE amphitheater was given by the coordinator, to present the structure of the auxiliary unit, its logistics, and the peculiarities for the care of patients with special needs. Then, two random members from each group, with a duly attested, simplified anamnesis form provided by the discipline of Biochemistry, were sent to the dental offices of the Center.

Saliva collection was performed with

Salivette® by a dental professional who routinely attended to the patient, to avoid exposing him to situations that could generate stress. The sterile cotton rollers were placed on the floor of the mouth for a maximum of 5 mins and then transferred to the laboratory in a Salivette®, and kept in an ice bath. Salivary flow, pH value, and buffering capacity were then estimated. It is important to note that all patients involved had cognitive and/or physical impairments that require specialized care, but for different causes. Autism spectrum disorder, Down syndrome, and cerebral palsy were the predominant causes.

At the time of saliva collection, students were able to note that the subject's anamnesis formed the name and number of the patient's CAO record. Subsequently, they were instructed to finish filling out the form by consulting the Center's medical record, extracting the following information: name, age, gender, race, food habits, oral habits, the reason for consultation, medications in use, and past illnesses. Information on the presence of mouth breathing, oral hygiene conditions, dependence on others for oral hygiene, presence or absence of changes in salivary flow (sialorrhea, xerostomia), tongue characteristics, periodontal condition, and odontogram were also transcribed for the simplified version of the medical record. The high number of students in each class required well-structured and organized logistics from the entire team involved. During the collection of saliva samples, students in the class who did not go to the clinics watched the procedure via simultaneous transmission, accompanied by a professor of the discipline, for clarification.

After the determination of salivary flow, pH, and buffering capacity, the saliva samples were centrifuged, fractionated, and stored in 0.5

mL conical flasks, containing the necessary volumes for the determination, in weeks distributed throughout the school year, of the following parameters: total protein, α -amylase, alkaline phosphatase, glucose, uric acid, aspartate aminotransferase, calcium, and phosphorus. The aliquots duly identified with the group and class numbers were stored at -20 °C. No aliquot was defrosted more than once.

After analyzing the salivary parameters, the results obtained by the groups were transcribed to the whiteboard in practical classes. At that time, each group was asked about the patients' oral and systemic clinical conditions, and it was up to the teacher to assume a questioning and critical stance, encouraging students to search for information in the literature that would allow the establishment of correlations between the results obtained and the clinical data of the patient. In this way, reflection was encouraged after action. After the initial discussion, the students were driven again to action, validating their reflections through research in databases such as SciELO and PubMed. The development of a report, turned in by the group at the end of the school year, improved the students' ability to write a technical-scientific report.

To assign a grade to the report, factored into the final grade of the course, the following criteria were used: work format and quality of the graphic presentation; compliance with the rules for citing bibliographic references; quality of writing and formatting of the abstract and introduction; clarity in the definition of objectives; adequacy of the description of the methodology; clarity of results presented; reasoning of the discussion in works cited in the reference list; clarity and cohesion of the conclusion with the proposition, the results obtained, and the discussion presented.

The titles of the reports produced in 2019

are described in Table 2. It is worth mentioning that none of these reports or data contained herein have been or will be used for the preparation of the TCC or any research work.

Table 1. Theoretical content, practical activity, and educational objective of the activities carried out

Theoretical lecture	Practical activity	Pedagogical Objective
Buffer systems	pH and buffering capacity of saliva	Correlate the analyzed parameters with the maintenance of the mineralized structure of the teeth integrity
Enzymes	Determination of the activity of acid phosphatase in the saliva	Understand the importance of enzymes in metabolic processes in health and disease, as well as their uses as salivary biomarkers of oral and systemic diseases
Salivary proteins and peptides	Determining the content of total protein in the saliva	Contextualize the multifunctionality of salivary proteins and peptides in promoting the health of oral tissues (lubrication, buffering action, protection, inhibition of the formation of dental calculus, etc.)
Mineralized tissue proteins	Determination of the activity of alkaline phosphatase in the saliva	Reinforce the importance of enzymes as salivary biomarkers of damage to periodontal tissue
Digestion and absorption of the main components of the diet	Determination of α -amylase activity in the saliva	Identify the importance of α -amylase in digestive function and as a salivary biomarker of emotional and physical stress
Partial oxygen reduction and generation of reactive oxygen species	Determination of uric acid in the saliva	Understand the importance of uric acid as a component of the non-enzymatic salivary antioxidant defense's protection against oxidative damage
Halitosis	Determination of transaminase activity in the saliva	Associate the increased activity of amino acid metabolism enzymes with the production of odor vectors associated with halitosis
Metabolic effects of epinephrine, glucagon, and insulin. Integrated metabolism regulation	Determination of glucose in the saliva	Explore the correlation between salivary biomarkers and systemic diseases, especially diabetes
Tooth and saliva mineral composition	Determination of calcium and phosphorus in the saliva	Learn about the composition and the importance of maintaining the mineral balance between tooth structure and saliva in preventing tooth decay

Table 2. Titles attributed by students to the final reports of the practical activities of the Biochemistry discipline in 2019

Class A - Fulltime Dentistry Course
<ul style="list-style-type: none">- Analysis of salivary biochemical parameters in a female patient with cerebral palsy - Analysis of the biochemical parameters of the saliva of a patient with Down syndrome in relation to oral and systemic conditions- Biochemical parameters of a patient with cerebral palsy: salivary compounds and oral health relationship.- Salivary and biochemical parameters associated with oral health.- Salivary parameters of a patient with delayed neuropsychomotor development.- Biochemical analysis of saliva in relation to the oral and systemic health of a patient with attention deficit hyperactivity disorder.- Biochemical analysis of the saliva of a patient with Down syndrome to determine their oral health.- Comparative analysis of salivary biochemical parameters between Rett syndrome and cerebral palsy.
Class B - Fulltime Dentistry Course
<ul style="list-style-type: none">- Salivary biochemical parameters of patients with an intellectual deficit and the relationship between indices and systemic health.- Comparative analysis of salivary parameters and their relationship with oral and systemic health in patients with DS.- Biochemical analysis of saliva in autistic patients: correlation between saliva components and oral health- Analysis of the biochemical parameters of the patient's saliva from CAOE: correlation between saliva components and oral health.- Analysis of the biochemical parameters of saliva: Correlation between salivary components and cerebral palsy.- Biochemical analysis of the saliva of a patient with autism: comparison of the results obtained with his clinical condition.- Analysis of the biochemical parameters of saliva in relation to the oral and systemic health of a patient with cerebral palsy- Correlation between biochemical parameters of the patient's saliva from CAOE with his oral and systemic health.
Night Dentistry Course
<ul style="list-style-type: none">- Analysis of the salivary profile of a patient with epilepsy. Final Biochemistry Report.- Analysis of the biochemical parameters of saliva with the clinical picture of a patient with intellectual disability.- Salivary biochemical parameters in relation to attention deficit hyperactivity disorder.- Biochemical profile of a patient undergoing orthodontic treatment with an intellectual and epileptic deficit.- Biochemical analysis of saliva to determine oral health.- Analysis of systemic factors that influence the biochemical patterns of saliva.- Oral and systemic characteristics that influence the biochemical factors of saliva.

At FOA-UNESP, the first contact of students with pre-clinical activities occurs in the second year of graduation, which can generate some frustration. Therefore, this activity, as proposed and carried out by the Biochemistry discipline, softened expectations, kept students motivated, and consequently, strengthened interest in the course, which could be observed throughout the year due to the very low number of absences in practical classes and good performance of the students, which resulted in a high percentage of approval.

Considering that the activity was carried out by an expressive number of students per class, and that it is well established that the learning style differs from individual to individual and is influenced by previous experiences¹⁹, it is natural that some deficiencies have surfaced, such as the difficulty in reading and understanding articles in English; the lack of computer skills; restrictions on interpretation, and following of text formatting rules. The difficulty in interpreting the clinical procedures described in the medical record cannot be included above, as it is inherent to the

fact that students are in their 1st year of graduation.

Regarding the difficulty in interpreting salivary biochemical parameters and their association with oral and systemic health, there was a lack of validated reference values for saliva, which results, in certain cases, in the absence of a consensus regarding saliva collection and processing techniques in the literature^{20,21}. Despite these obstacles, the proposed objectives were still achieved.

Motivation and interest in biochemistry should, as much as possible, be maintained by the teachers responsible^{22,23}. Thus, all concepts were explored, so that the correlations with patients' systemic and oral clinical parameters were evidenced, thereby demonstrating their applicability. To further guide and assist students, meetings were held with the discipline's teachers and monitors to discuss specific points. We credit part of the success of this activity to the fact that real cases of patients, with whom the students had direct or indirect contact, were studied, which led to a greater sense of involvement that facilitated the acquisition, integration, discussion, and application of the concepts of biochemistry.

The epistemological bases of traditional teaching were maintained through pre-established content, unidirectional transmission of knowledge in theoretical classes, and transversal evaluation. However, the description of the experience report demonstrates the viability of incorporating a new component into the structure of the discipline, through the development of practical activities in which the student is an active participant who conducts the process of scientific investigation. This helps improve theoretical and practical knowledge and provide transformative actions in a professional training path. Although the analysis of the

epistemological status of biochemistry indicates problems related to the imagined combination of entities of chemistry and biology²⁴, the students' curiosity and interest in the concepts covered in the theoretical classes were stimulated by integrating the results of the salivary analyses of a real studied clinical case.

The set of activities carried out stimulated the cognitive learning domain, since intellectual development was fostered, and the psychomotor domain, since students had to learn to handle laboratory glassware and equipment previously unknown to them while performing the laboratory tests.

3 FINAL CONSIDERATIONS

Some factors contribute to the low performance and motivation in the study of biochemistry. Among these, we highlight the superficial approach on the part of the students at the time of the study, privileging a mechanical process of memorization, and the lack of effective knowledge about the importance of the teachings of the discipline in their future professional careers.

In the proposed and reported activities, there was satisfactory student involvement throughout the school year, greater appreciation and understanding of the importance of biochemistry, and an understanding of the complexity involved in caring for patients with special needs. Despite the longing for practical and clinical procedures, students demonstrated greater awareness of the importance of theoretical knowledge essential for the interpretation of the results obtained, valuing the importance of basic discipline in their training.

It was concluded that the adopted strategy is viable and can benefit educators who seek alternatives that allow the integration of basic and clinical sciences to the teaching of Biochemistry for Dentistry.

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RESUMO

Análise bioquímica da saliva de pacientes com deficiência cognitiva e física como estratégia de aula prática de Bioquímica no curso de Odontologia

Diante da importância do ensino de Bioquímica na formação do futuro cirurgião-dentista, o objetivo é apresentar uma estratégia pedagógica que possibilita a articulação dos conceitos moleculares básicos da disciplina no entendimento da complexidade das doenças bucais e sistêmicas, por meio da análise bioquímica da saliva de pacientes com deficiência cognitiva e física do Centro de Assistência Odontológica à Pessoa com Deficiência (CAOE) da Faculdade de Odontologia de Araçatuba- Universidade Estadual Paulista (UNESP). Para tanto, grupos de cinco estudantes realizaram as análises na saliva, coletada por um cirurgião-dentista do CAOE. Imediatamente após a coleta, foi determinado o fluxo salivar, pH e a capacidade tamponante. Na sequência, a saliva foi centrifugada, fracionada e

armazenada a -20 °C até o momento das análises bioquímicas. Durante as aulas práticas, realizaram-se os seguintes ensaios, utilizando *kits* comerciais: proteína total, α -amilase, fosfatase alcalina, ácido úrico, glicose, aspartato aminotransferase, cálcio e fósforo. Ao final do ano letivo, os estudantes apresentaram relatório contextualizando os resultados com a saúde bucal e sistêmica do paciente. Plantões de dúvidas com monitores e professores auxiliaram na interpretação da ficha de anamnese e nas correlações dos parâmetros clínicos bucais e sistêmicos. Para os estudantes ingressantes, foi a primeira oportunidade de integrar o conhecimento teórico às condições clínicas de um paciente. Conclui-se que a estratégia adotada é viável e pode beneficiar educadores que buscam alternativas que permitam a integração das ciências básicas e clínicas ao ensino de Bioquímica para a Odontologia.

Descritores: Aprendizagem. Ensino. Educação em Odontologia. Bioquímica. Saliva.

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