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Original Article

# Chemical and physical features of biological fluids in treatment of hydatid disease

## Propriedades físico-químicas dos fluidos biológicos no tratamento da doença hidática

G. Widjaja[a,b*], L. A. Younus[c], W. K. Abdelbasset[d,e], D. D. Ibragimov[f], A. V. Yumashev[g], M. N. Shalaby[h], Y. F. Mustafa[i] and I. Fardeeva[j]

[a]Postgraduate Study, Universitas Krisnadwipayana, Jatiwaringin, Indonesia
[b]Faculty of Public Health, Universitas Indonesia, Depok, Indonesia
[c]Department of Clinical Laboratory Sciences, Faculty of Pharmacy, Jabir Ibn Hayyan Medical University, Al Najaf Al Ashraf, Iraq
[d]Prince Sattam Bin Abdulaziz University, College of Applied Medical Sciences, Department of Health and Rehabilitation Sciences, Al Kharj, Saudi Arabia
[e]Cairo University, Kasr Al-Aini Hospital, Department of Physical Therapy, Giza, Egypt
[f]Department of Oral and Maxillofacial Surgery, Samarkand State Medical Institute, Samarkand, Uzbekistan
[g]Sechenov First Moscow State Medical University, Moscow, Russia
[h]Suez Canal University, Biological Sciences and Sports Health Department, Faculty of Physical Education, Ismailia, Egypt
[i]University of Mosul, College of Pharmacy, Department of Pharmaceutical Chemistry, Mosul, Iraq
[j]Kazan Federal University, Kazan, Russia

**Abstract**

The aim of this paper is to better understand the dynamics of crystallogenic and starting activity in biological fluids of patients throughout surgery and the late postoperative phase in alveococcosis. Samples of saliva from 22 individuals with alveococcosis were included in the research. Biological fluid samples were taken at the time of admission and before the patient was discharged. Following that, slides were made utilizing the teziocrystalloscopy method, which incorporates the investigation of the crystal forming activity of mixed saliva with its starting characteristics using a 0.9 percent sodium chloride solution as the foundation ingredient. Using our own set of criteria, we evaluated the outcomes of crystalloscopic and tezigraphic experiments. Specrophotometric examination of tezigraphic and crystalloscopic facies was done using a PowerWave XS microplate spectrophotometer at wavelengths of 400, 350, and 300 nm to augment the results from ocular morphometry of dried saliva micro slides. Surgical therapy results in a partial normalization of physical and chemical parameters, as well as the composition of the patient's biological fluids after the patient is discharged from the hospital.

**Keywords:** crystallization, saliva, surgical treatment, alveolar hydatid disease, alveococcosis.

**Resumo**

O objetivo deste trabalho foi compreender melhor a dinâmica da atividade cristalogênica e inicial nos fluidos biológicos dos pacientes durante a cirurgia e na fase pós-operatória tardia da alveococose. Amostras de saliva de 22 indivíduos com alveococose foram incluídas na pesquisa. Amostras de fluidos biológicos foram coletadas no momento da admissão e antes da alta do paciente. Em seguida, foram confeccionadas lâminas utilizando a técnica de teziocristaloscopia, que combina a investigação da atividade formadora de cristais da saliva mista com suas características iniciais, utilizando uma solução de cloreto de sódio a 0,9% como ingrediente-base. Usando nosso próprio conjunto de critérios, avaliamos os resultados de experimentos cristaloscópicos e tezigráficos. O exame espectrofotométrico da fácies tezigráfica e cristaloscópica foi feito usando um espectrofotômetro de microplaca PowerWave XS nos comprimentos de onda de 400, 350 e 300 nm para aumentar os resultados da morfometria ocular de microplacas de saliva seca. A terapia cirúrgica resulta na normalização parcial dos parâmetros físicos e químicos, bem como da composição dos fluidos biológicos do paciente, após a alta hospitalar.

**Palavras-chave:** cristalização, saliva, tratamento cirúrgico, doença hidática alveolar, alveococose.

## 1. Introduction

The liver is the largest solid organ in the body. Due to the production and secretion of bile, the liver is a gland. The liver is protected by the chest and is placed in the upper part of the abdominal cavity on the right side. The liver

*e-mail: widjaja_gunawan@yahoo.com; sfmsmu@mail.ru
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is made up of two major lobes, each of which is made up of smaller lobules. Liver cells have two various sources for blood supply. One is the hepatic artery, which carries oxygen-rich blood from the heart, and the other is the portal vein, which carries blood and nutrients from the gastrointestinal tract and spleen to liver cells. Liver disease is a general definition of all potential problems that prevent the liver from performing its functions properly. Usually, the disease should affect more than 75% of the liver tissue so that the liver does not function properly. It should be noted that the liver is the only organ in the body that can easily replace its damaged cells. However, if the number of damaged cells in the liver exceeds a certain level, the liver will not be able to replace them and, as a result, will not be able to perform its functions properly. The liver may be damaged in several ways, including:

- Inflammation of cells such as hepatitis;
- Obstruction of bile flow such as cholestasis;
- Accumulation of cholesterol or triglycerides such as fat change or steatosis;
- Impaired blood flow to the liver;
- Harm to liver tissue by abnormal cells such as cancer cells or harm to liver tissue by minerals and chemicals.

Exploratory laparotomy and invasive palliative results show that a significant number of patients with localized liver disorders are profoundly inoperable (Perinel and Adham, 2019). Doctors recently restricted themselves to excision of fistulas, dissection of purulent cavities, and the injection of parasitotropic medicines during repeated liver surgeries (Voskanyan et al., 2018; Bebezov et al., 2019; Zagaynov et al., 2018; Panteleev et al., 2019). Repeated radical procedures on patients who were previously thought to be inoperable appear to be a genuine possibility. Yudin (1929) is said to be the first to conduct a radical liver dissection following an exploratory laparotomy for alveococcosis (Martusevich and Zhdanova, 2020).

Increased surgery causes that most people with localized liver lesions do not receive treatment at a specialist medical center (Błaszkowska and Wójcik, 2011; Błaszkowska and Góralska, 2016; Menna et al., 2018). Concurrently, most of them are restricted to exploratory laparotomy or palliative surgery due to technical problems and erroneous assessments of the operability of the lesion focus, leaving patients disabled and with a bad result (Skipenko et al., 2012; Dybicz et al., 2018; Pielok et al., 2020). A number of Russian surgeons' papers dedicated to repeated procedures primarily on liver alveococcosis. have been published since 1964 (Tumol'skaia, 2010; Bebezov et al., 2019; Panteleev et al., 2019). Experts demonstrated that there is a major difficulty with delivering a radical therapy to inoperable patients with focal lesions of the liver, which happened at the same time as different skills in the surgical treatment of patients with focal lesions of the liver emerged (Zagaynov et al., 2018). On the other hand, it's critical to back alveococcosis surgery with complete diagnostic guidance. The application of current instrumental approaches to the study of liver morphology (ultrasonography, magnetic resonance imaging, computed tomography, and X-ray diagnostics with contrasting) allows for a high grade of precision in determining the nature, extent, and intensity of structural damage to the organ connected to disease progression (Gossios et al., 1997; Imankulov et al., 2015; Patkowski et al., 2016). Latex agglutination responses and a particular ELISA alveococcosis diagnosticum can be used to verify the difficulty of the immune response to the presence of the parasite (Wnukowska et al., 2011). Sputum may also include alveococcus scolexes. Simultaneously, the characteristics of metabolic alterations in alveococcosis, such as differences in biological fluids' chemical and physical properties, surgical therapy, and treatment in the postoperative period, have previously been understudied. In this context, it is demonstrated that the introduction of alveococcus alters the crystallogenic characteristics of biological substrates in a substantial and directed manner and that the nature of this transformation results in equivalent alterations in saliva and urine crystallostasis (Martusevich et al., 2006; Gorbachev et al., 2019). Explaining the dynamics of crystallogenic and starting activity of patients' biological fluids in alveococcosis after surgery and in the late postoperative phase is the subject of present study and is of considerable practical and scientific importance.

## 2. Materials and Methods

We examined saliva samples from 22 individuals medicated for alveococcosis. Conventional instrumental (MRI, CT, and ultrasound) and laboratory (ELISA, latex agglutination) testing were used to confirm the diagnosis (Martusevich et al., 2014; Vuchev, 2014; Filippou and Tsoumpas, 2018; Damião et al., 2021; Yang et al., 2021). All of the individuals had surgery successfully (partial hemihepatectomia, lobectomia, hepatectomia). Sampling was done at the time of admission and before the patient was discharged from the hospital. Patients had not engaged in any physical exercise or been in a condition of psycho-emotional stress within 3 hours of the research. Patients washed their mouths completely with about 100 mL of $dH_2O$ for 5 minutes prior to biological fluid collection. Following that, 1 mL (milliliter) of oral fluid was spat into dry and clean test tubes. Following that, images were produced utilizing the teziocrystalloscopy technique (Kamakin and Martusevich, 2002; Martusevich et al., 2019), which integrates the investigation of mixed saliva crystal formation activity and its starting characteristics using a 0.9 percent sodium chloride solution as the foundation ingredient. The outcomes of tezigraphic and crystalloscopic tests were evaluated using the researchers' criteria (Martusevich et al., 2008, 2016b, c). It may be used to determine the regularity of their distribution on the contexture of the sample, the degree of amorphous and crystalline component destruction, the intensity of particular zones of the facia, the biological substrate's crystallization specialties, etc. Spectrophotometric examination of teziographic and crystalloscopic facias, conducted on a PowerWave TM XS microplate reader at wavelengths of 400, 350, and 300 nm (nanometer), was used to augment the results from visual morphometry of dried saliva micro slides. We estimated the amount of optical density of facies by deducting the optical density of pure glass from the overall value of the indicator to

neutralize the impact of glass properties on the findings of spectrometric investigations of biological crystals. Microsoft Excel 2007 spreadsheets and the Primer of Biostatistics version 4.03 application were used to analyze the statistical data.

## 3. Results and Discussion

Saliva seems to be the most efficient non-invasive marker of changes in the chemical and physical characteristics and composition of blood, as well as biological substrates produced in the digestive system (Nater and Rohleder, 2009; Nunes et al., 2015; Martí-Álamo et al., 2012). The last one is the alveococcosis locus morbi, which appropriately represents its metabolic and functional condition. As a result, a recent study of crystallogenic activity in the biological fluid was of special practical and scientific relevance (Vorobyov et al., 2009; Martusevich et al., 2016a). According to the findings, the main morphometric characteristics of the crystalloscopic facies of patients' saliva show dynamics of changing parameters (see Figure 1).

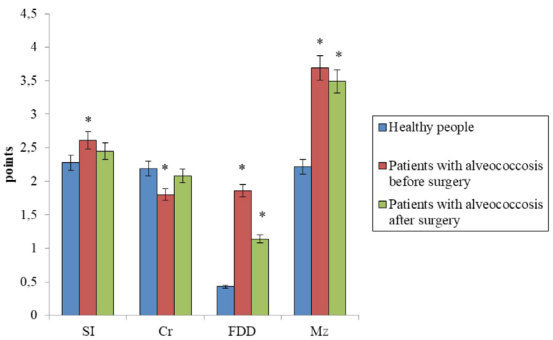


**Figure 1.** The outcome of a parametric investigation of patients' saliva crystallization during surgery for alveococcosis (Mz – clearity of marginal zone, FDD – facia destruction degree, Cr – crystallizability, SI – structure index; "*" – Statistical disparities in the level of healthful individuals is $p<0.05$).

The multidirectional alterations in the crystallizability (Cr) and structural index (SI) of biological substrate components, which are indications of dehydration structuring of the biological medium (Martusevich et al., 2020, 2021). As a result, on the oral fluid slides of patients with alveococcosis, there are more complex by the fernlike character of the organization elements, dendritic crystals with a branching order of 3.3 or higher, which is mirrored in the structure's development in comparison to the level of the index micro slides of the saliva of nearly healthy individuals. The expansion of the primary structural components of the saliva facies in individuals with alveococcosis causes a reduction in the density of the crystals (see Figure 2), which is measured as a crystallizability parameter. The pathological nature of the identified transitions in crystalogenic potential in the patients under consideration is made apparent in the dynamics of facies collapse, which reaches nearly two relative units (rel.un.), corresponding to the formation of extreme disruption of crystal element formation whilst still preserving type differentiation. The mechanical characteristics of the parasite's growing liver and other organs and the parasite's immunological responses to its existence are known to play a role in the pathogenesis of alveococcosis (Patkowski et al., 2016; Voskanyan et al., 2018).

As a result, higher concentrations of immunoglobulins (Schroeder Junior and Cavacini, 2010) have been seen in the blood, and to a lesser extent, in other biological substrates, resulting in higher protein concentrations. Information on salivary indicators of metabolic changes in the peripheral zone of the slides (parameter Mz), where the dehydration protein macromolecules (McPherson, 2017) are concentrated, is shown by the crystallogram (Sanchez-Weatherby and Moraes, 2016). This index was discovered to have a high value in alveococcosis patients and to be substantially lower than the normal level ($p<0.05$).

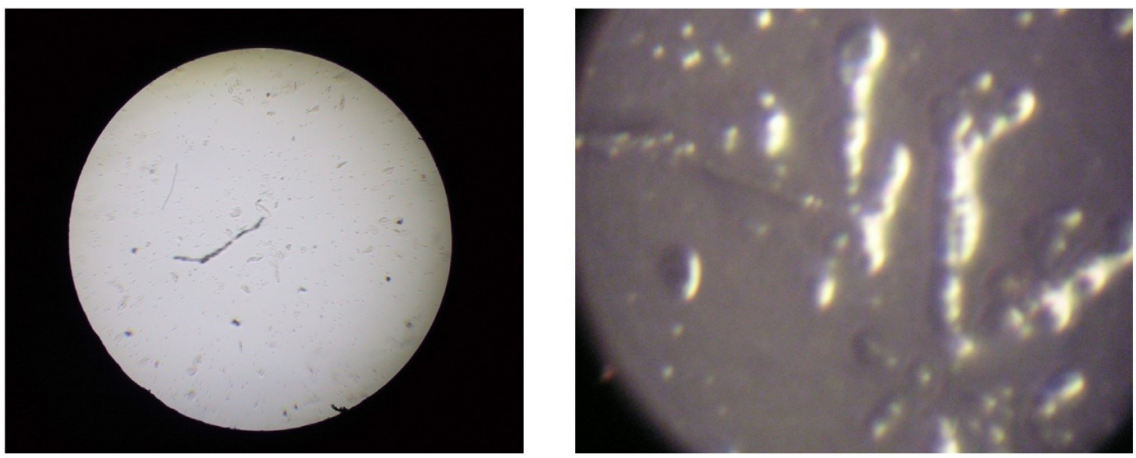


**Figure 2.** Healthy and alveococcosis patients' saliva crystallosopic facias.

Generally speaking, the crystallogenic characteristics of saliva before surgery change considerably from a healthy individual's crystalloscopic pattern. The crystallogenic characteristics of the investigated biological fluid are dramatically altered during the early postoperative phase (see Figure 1). Therefore, the parameters describing the density and complexity of structure-building crystal components have been nearly completely normalized (Cr and SI, respectively). Both of these metrics do not alter substantially from the baseline when compared to the control (pre-surgery) level ($p<0.05$). The facia destruction degree (FDD), which is the key measure characterizing the accuracy of crystallogenesis, is also lowered considerably when compared to the preoperative crystalloscopic pattern ($p<0.05$), although it does not approach physiological levels. It's noteworthy to note that the intensity of saliva microslides' marginal zone varies very little after the procedure. This pattern may be linked to the persistence of high anti-alveococcus antibody concentrations in the blood and, as a result, in other bodily fluids. As a result, the parameter Mz, which is directly reliant on the amount of a protein, has a comparable effect. In individuals with alveococcosis, partial normalization of salival crystallogenic characteristics occurs after surgery. The most common trend in shifts in crystallogenic characteristics of saliva in alveococcosis is the augmentation of crystal elements, as well as the depth of their construction. This study was the first to demonstrate the occurrence of such changes. The overall optical density of the crystalloscopic facies of saliva reduces after surgery, although it remains substantially greater than the values found in healthy people (see Figure 3).

This process is also most plainly evident in the spectroscopic research at $\lambda$= 300 nm, but it is less strong at $\lambda$= 350 nm, and at the maximum of $\lambda$= 400 nm, it is nearly completely gone due to relatively low absolute values. Compared to healthy participants ($p<0.05$), the fundamental indication of biological fluid starting potential, tezigraphic index (TI), was found to be substantially greater in the saliva of the considered cohort of patients following surgery. This measure decreases considerably after surgical therapy, although the differences are not statistically significant. The complexity marker of structure-building elements, crystallinity (C), was found to have a similar pattern. However, the indicator's postoperative level doesn't really change considerably from the pre-surgery and typical tezigraphic pattern of saliva. The belt coefficient (BC), which is a criterion of dispersion of molecular weight of biological substrate components, is revealed to have the least fluctuation among the fundamental tezigraphic indicators (see Figure 4). The efficacy of surgical therapy may be seen in the dynamics of the main tezigraphic criteria of the accuracy of structure creation processes, as well as the clarity of the slides' marginal zone (see Figure 4). These values were substantially lower after surgery compared to pre-surgical levels, but they did not approach the levels seen in healthy people ($p<0.05$). This pattern suggests that alveococcus-related metabolic changes in bodily fluids' content and physical-chemical characteristics are safe following severe surgery. The outcomes of a spectroscopic examination of the tezigraphic facies of patients' saliva in the dynamics of surgical therapy confirm these changes completely (see Figure 5). At all wavelengths utilized in this example, the dynamics of the total optical density of tezigrams are visible.

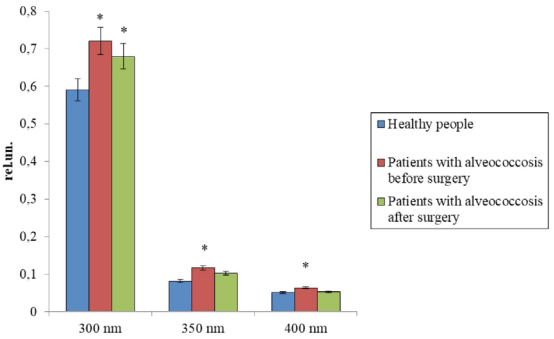


**Figure 3.** During surgery, saliva crystalloscopic facias of patients with alveococcosis were analyzed using spectrometric methods ("*" – Statistical disparities in the level of healthful individuals is $p<0.05$).

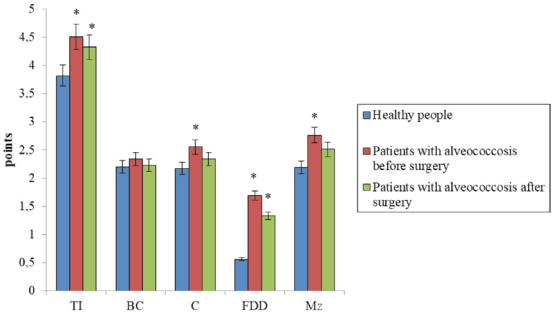


**Figure 4.** During surgical therapy of alveococcosis, results from tezigramms parametric saliva analysis were collected (Mz – clearity of marginal zone, FDD – facia destruction degree, C – crystallinity, BC – belt coefficient, TI – teziographic index; "*" – Statistical disparities in the level of healthful individuals is $p<0.05$).

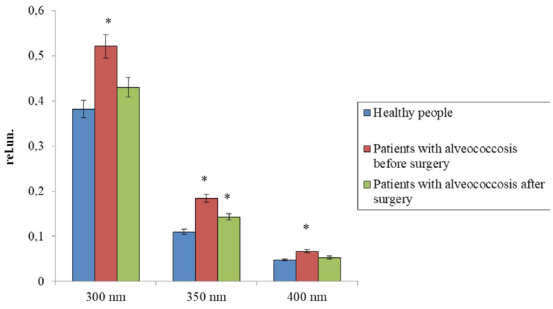


**Figure 5.** Spectrometry results of alveococcosis patients' saliva tezigraphic facias during surgery ("*" – Statistical disparities in the level of healthful individuals is $p<0.05$).

## 4. Conclusion

We were able to determine that there are substantial changes in the crystallogenic characteristics of saliva in alveococcosis of the liver after conducting crystalloscopic

investigations. Decrease in crystallizability, the grade of the destruction of individual structures, and the severity of the sample marginal zone, as well as an increase in the structure index, are all signs of these changes. Facias' optical density rises as a result of these changes. Almost all of the parameters are normalized after surgery, with the exception of the grade of facias destruction and the clarity of the sample's marginal zone, which remain high. Simultaneously, only at a wavelength of 350 nm does the optical density of micro-preparations increase. The discovery of such crystalloscopic indicator dynamics enables us to use them as a secondary indication of the efficacy of liver alveococcosis therapy. Subsequent confirmation of the diagnostic information content of the technology we employed could be used to build a strategy for quickly determining if liver resection is beneficial in alveococcosis. Our findings also add to our knowledge of the metabolic alterations that occur in patients with hepatic alveococcosis.

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