http://dx.doi.org/10.35630/2199-885X/2022/12/2.2

DIGITAL TECHNOLOGY FOR PROCESSING DRIED DROPS OF BIOFLUIDS

Received 23 January 2022;
Received in revised form 19 February 2022;
Accepted 21 February 2022

Andrew Martusevich*[1,3]✉, *Lida Kovaleva*[4], *Konstantin Karuzin*[2], *Maria Feofilova*[6], *Ivan Bocharin*[1,3], *Alexandra Surovegina*[1,3], *Vladimir Nazarov*[1,5], *Anastasia Kashirina

[1] *Privolzhsky Research Medical University, Nizhny Novgorod, Russia*
[2] *Bioniq Health-Tech Solutions, London, United Kingdom*
[3] *Nizhny Novgorod State Agricultural Academy, Nizhny Novgorod, Russia*
[4] *Kuban State Medical University, Krasnodar, Russia*
[5] *Insitute of Applied Physics of Russian Academy, Nizhny Novgorod, Russia*
[6] *Tula State University, Tula, Russia*

✉ *cryst-mart@yandex.ru*

ABSTRACT — Attempts to digitize samples and apply artificial intelligence and machine learning methods to analyze crystalloscopic (dried drops of biological fluids) and tesigraphic (dried drops of biological fluids with crystallogenic substance) facies have not yet been successful. In this regard, there is a need to develop a simplified algorithm for describing the facies of biological fluids, which can be used for a unified computer study of the results of crystallization of biological objects, which served as the purpose of the work. To develop and test the method presented in this paper, we used more than 16,000 images of dried biological fluids of the human and animal body, including both crystalloscopic and tesigraphic facies. The algorithm is based on determination of 4 main parameters (crystallizability, structure index, facies destruction degree and clearity of the marginal zone), graded on three-point scales. In addition, a facies integral parameter combining the values of all criteria is proposed.

KEYWORDS — biocrystallomics, facies, digital analysis, crystallization

INTRODUCTION

Currently, there are numerous studies devoted to the study of the crystallogenic properties of biological fluids in humans and animals [1–4, 7]. It is shown that the application of biocrystallomics as a new discipline that comprehensively considers the processes of crystallization and structuring in biological systems has broad prospects [5, 6]. They are associated with the diagnosis and differentiation of diseases of various profiles, personification of the appointment and monitoring of the effectiveness of treatment, prediction of the further course of pathology [1–7]. The fundamental basis for the diagnostic informativeness of the technology is the specificity of the dehydration structuring of the components of biological fluids in the form of a set of organo-mineral aggregates [3, 5, 6]. In conditions of pathology, the component composition and physicochemical properties of the biological media are significantly transformed, which is recorded in shifts in the morphology of their dried samples (facies) [1–3, 5–7].

To date, two main options for assessing the crystallogenic properties of liquid media have been identified: the study of their own crystallogenic activity (crystalloscopy) and the study of co-dehydration with a crystal-forming agent (graphic tesigraphy), which is performed by various salts (most often isotonic sodium chloride solution and copper sulfate) [6]. On the other hand, the greatest difficulties are caused by the correct description of the crystallization result, for which numerous methods have been proposed (from a morphological approach involving the search and manual counting of the number of individual structures to the application of various quantitative and semi-quantitative parameters). For a long period, we have been developing and testing on various biological objects a system of multiparametric description of crystallograms (result of dehydration of biofluid drops) and tesigrams (result of co- dehydration of biofluid drops with special inducer of crystallization), which is independent of the type of biological fluid and the morphology of the elements formed from it during drying [5, 6]. However, the attempts made by us and other specialists to digitize samples and apply artificial intelligence and machine learning methods to analyze crystalloscopic and tesigraphic facies have not yet been successful [3, 4]. This is primarily due to the multiplicity of criteria, the complex and wide gradation of their values, the presence of direct and inverse scales for various parameters and other factors [3–5]. In this regard, there is a need to develop a simplified algorithm for describing the facies of dried samples of liquid biological media, which can be used for a unified computer study of the results of crystallization of biological objects.

The purpose of this work was to create a unified algorithm for evaluating crystalloscopic and tesigraphic facies based on basic criteria.

MATERIAL AND METHODS

To develop and test the method presented in the work, we used more than 16,000 images of dried

biological fluids of the human and animal body (blood serum and plasma, saliva, urine, sweat and tear fluids, etc.) obtained over the past 14 years [5, 6]. The specified volume of the material included both crystalloscopic facies, which are the result of their own dehydration of biosubstrates, and tesigrams, in which 0.9%; 0.1%, 3% and 10% solutions of sodium chloride, 0.1N solutions of hydrochloric acid, sodium hydroxide and other compounds were used as crystal-forming agents (crystallization inducers). In micro-preparations of dried biological media, the most informative features were identified, which were used to develop a substrate-independent, compact (in terms of the number of indicators) parameter system based on the minimum number of gradations of each evaluation feature.

RESULTS

In-depth analysis of a large array of crystalloscopic and tesigraphic facies made it possible to form the most compact and simple algorithm for describing dried microsampling of biological fluids. It includes 4 main indicators, graded on three-point straight scales:

I. ***Crystallizability*** (Cr) is a parameter characterizing the density of crystalline and amorphous elements in facies; the main quantitative criterion for the intensity of crystallogenesis. Composition of the scale:

— 0 points — no signs of crystallization (less than 5 structures in the field of vision) or hypercrystallogenesis (more than 30 structures in the field of vision);

— 1 point — moderately suboptimal crystal density (5–10 or 20–30 elements in the field of view);

— 2 points — optimal intensity of crystalization (10–20 crystals in the field of view).

II. ***The structure index*** (SI) is an indicator that characterizes the complexity of the structure of the facies. Gradations of the parameter:

— 0 points — only amorphous bodies or numerous branched dendrites filling the entire field of view;

— 1 point — single crystals (it is possible to include single polycrystalline structures);

— 2 points — dendritic and single-crystalline elements are present in facies in almost equal proportions.

III. ***The facies destruction degree*** (FDD) is a criterion for the correctness of the processes of crystallogenesis. It includes 2 evaluation options:

— 0 points — total destruction of structural elements in the dried microsamples of biological fluid;

— 1 point — pronounced changes in crystal morphology while preserving the possibility of identifying their type;

— 2 points — there are no signs of structural destruction or are fleeting, morphology and interaction with other elements can be determined for all elements.

IV. ***The clearity of the marginal protein zone*** (Mz) is an indicator describing the state of the proteome of a biological fluid. Gradations of the parameter:

— 0 points — the marginal belt is absent or has an excessive size (more than ⅔ of the facies radius), numerous, randomly located faults are found in it;

— 1 point — the radius of the marginal zone from ⅓ to ⅔ of the radius of the dried sample, there are moderately chaotic faults;

— 2 points — the marginal zone occupies less than ⅓ of the facies radius, contains evenly distributed, centripetal faults.

For the tasks of digitizing the result of own and initiated crystallization of biological fluids and obtaining a single value for each sample in order to form a conclusion on it, a facies integral parameter (FIP) was additionally proposed, calculated by the formula:

$$FIP = Cr + SI + FDD + Mz,$$

where *Cr* is crystallizability; *SI* is the structure index; *FDD* is the facies destruction degree; *Mz* is the clearity of the marginal zone.

Based on subsequent studies, the informativeness of the created algorithm for describing crystalloscopic and tesigraphic facies was confirmed.

CONCLUSION

A detailed analysis of an extensive array of crystallograms and tesigrams of biological fluids in humans and animals (blood serum and plasma, saliva, urine, sweat and tear fluids, etc.) has been created and we tested a comprehensive unified algorithm for their evaluation. It is based on the determination of 4 main parameters (crystallizability, structure index, facies destruction degree and clearity of the marginal zone), graded on three-point scales. In addition, a facies integral parameter combining the values of all criteria is proposed. It should be noted that the developed approach for the first time makes it possible to describe both crystalloscopic and tesigraphic facies according to uniform criteria.

FUNDING

This work was financially supported by the Ministry of the Health of Russian Federation (№AAAA-A20-121030100012-9).

REFERENCES

Denisov A.B., Pushkar' D.Y., Denisov S.A. Use of saliva crystallogenic properties for early diagnostics of

prostate cancer // Bull. Exp. Biol. Med. – 2006. – Vol. 142, no. 2. – P. 242-245. DOI: 10.1007/s10517-006-0338-2

Jordanishvili A.K. Oral liquid adult: age peculiarities of the physicochemical properties and micro crystallization // Adv. Gerontol. – 2019. – Vol.32, N3. – P. 477-482.

Kokornaczyk M.O., Bodrova N.B., Baumgartner S. Diagnostic tests based on pattern formation in drying body fluids - A mapping review // Colloids Surf B Biointerfaces. – 2021. – Vol. 208. - 112092. doi: 10.1016/j.colsurfb.2021.112092

Li Z.X., Zha Y.M., Jiang G.Y., Huang Y.X. AI aided analysis on saliva crystallization of pregnant women for accurate estimation of delivery date and fetal status // IEEE J Biomed Health Inform. – 2021. - Dec 15. doi: 10.1109/JBHI.2021.3135534

Martusevich A.K., Kamakin N.F. Crystallography of biological fluid as a method for evaluating its physicochemical characteristics // Bull. Exp. Biol. Med. – 2007. – Vol. 143, no. 3. – P. 385-388. doi: 10.1007/s10517-007-0118-7.

Martusevich A.K., Kamakin N.F. Unified algorithm for the study of free and initiated crystallogenesis of biological fluids // Clinical laboratory diagnostics. - 2007. - No. 6. - P. 21-24.

Pancu G., Lăcătuşu S., Căruntu I.D., Iovan G., Ghiorghe A. Evaluation of caries activity using the micro-crystallization saliva index (IMK) // Rev. Med. Chir. Soc. Med. Nat. Iasi. – 2006. – Vol. 110, no. 1. – P. 206–211.