Algorithm for Evaluation of Crystal Figures Obtained after Drying of Mixed Saliva

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Algorithm of qualitative description of crystallographic aggregations of mixed saliva was developed. The efficiency of this method for the diagnosis of gastrointestinal diseases was demonstrated.

Key Words: *saliva; dendritic (skeletal) microcrystals; morphology*

Formation of crystal structures in biological fluids (blood, lacrimal fluid, saliva, urine, *etc.*) is intensely studied now [6,9]. The main purpose of these studies is to evaluate the adaptation status: whether the subject is healthy or ill, and to diagnose the disease. These studies are based on the following paradigm: different pathologies change the crystallization characteristics of biological fluids.

The method historically called "crystal deposition method" consists in drying biological fluid on an open surface; it is widely used in experimental dentistry. Its disadvantage is the absence of precise criteria of crystallization. We improved the method of native crystallization, which enabled us to create an algorithm for description of the visual images of the most incident variants of microcrystal aggregations of mixed saliva.

MATERIALS AND METHODS

The method for obtaining videofiles of salivary pool crystallograms was described previously [1-5,7]. The photographs were made using a Leica DM-LS microscope with a Sony SSC-DC30P camcorder. The files were saved as scanning gray images (256 gray gradations) with 300 points/inch resolution in the BMP format. A total of 581 videofiles were studied and the algorithm of data analysis was developed.

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RESULTS

Videofiles of the resultant crystallograms are presented in Fig. 1, *1*, *6*, *8*, *10*; Fig. 2, *5*. The rest figures show fragments detected using Adobe PhotoShop graphic software. The fragment was rotated around its axis until it looked as a tree or a bush.

Normally four main types of crystals can be seen in mixed saliva (Fig. 1).

Type 1 (Fig. 1, *1*) is characterized by the absence of crystal structures. Normally this type occurs in women of fertile age during phase 2 (luteal phase) of the menstrual cycle.

Type 2 crystals (Fig. 1, *2*) are box-shaped. These crystals are normally rare. We distinguished 4 variants (Fig. 1, *2-5*). We failed to identify these crystals.

Type 3 crystals (Fig. 1, *6*) are cross-shaped. They are normally rare. Four variants of cross-shaped crystals were distinguished (Fig. 1, *6-9*). According to some data [8], these are variants of growth of the same crystal type.

Type 4 crystals (Fig. 1, *10*) are the main type of salivary crystals, looking like a tree or bush. They are called skeletal crystals in crystallography and dendrites in physics. In our case the skeleton of salivary crystals is flat (lying in one plane).

The main branches of the skeleton (MBS) or first-order branches with second-order branches originating from them, *etc.*, are distinguished in dendritic crystals. Besides these, irregular processes are forming during crystallization. These variants can be expertly evaluated (Fig. 1, *11*).

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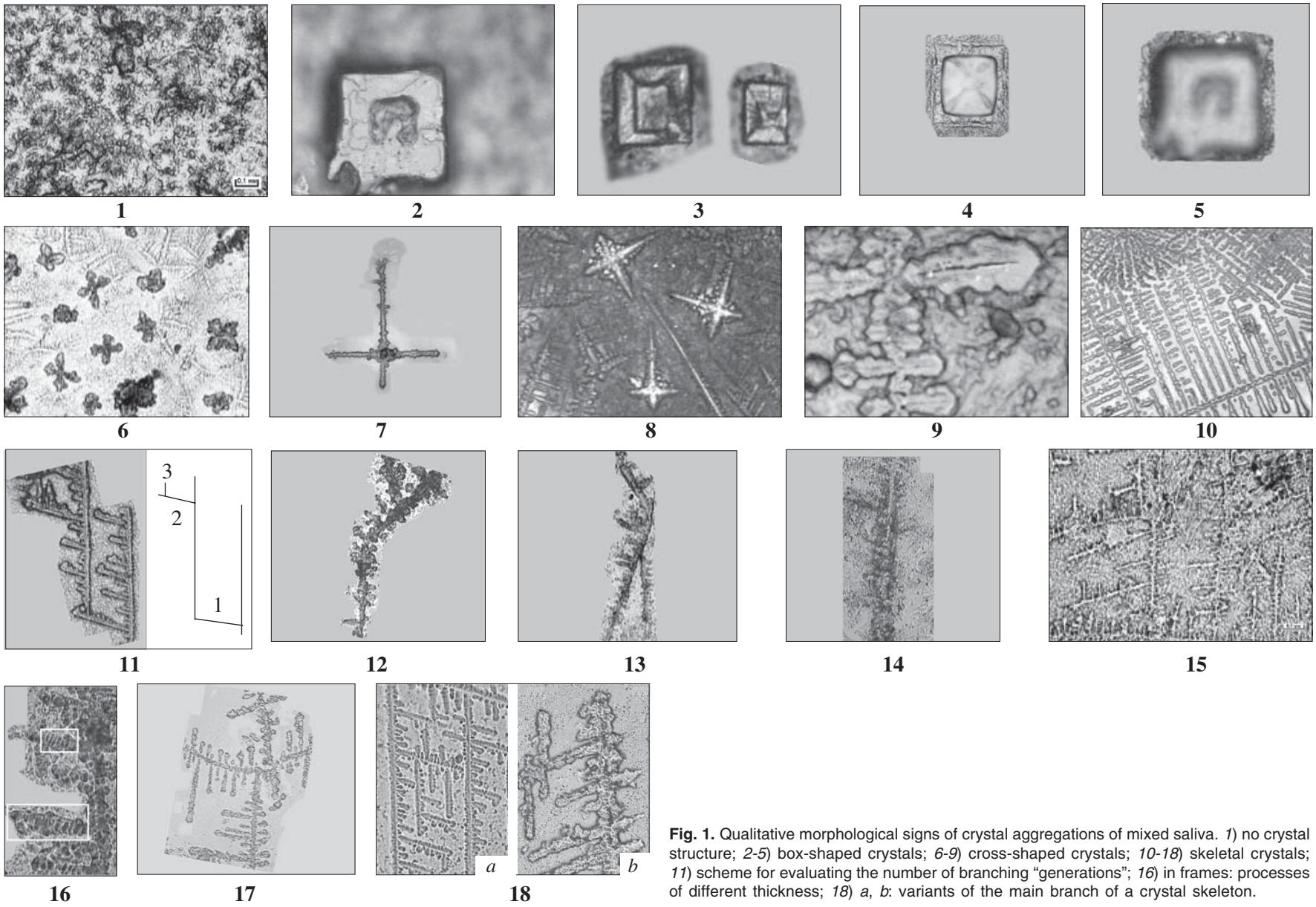

Fig. 1. Qualitative morphological signs of crystal aggregations of mixed saliva. *1*) no crystal structure; *2-5*) box-shaped crystals; *6-9*) cross-shaped crystals; *10-18*) skeletal crystals; *11*) scheme for evaluating the number of branching "generations"; *16*) in frames: processes of different thickness; *18*) *a*, *b*: variants of the main branch of a crystal skeleton.

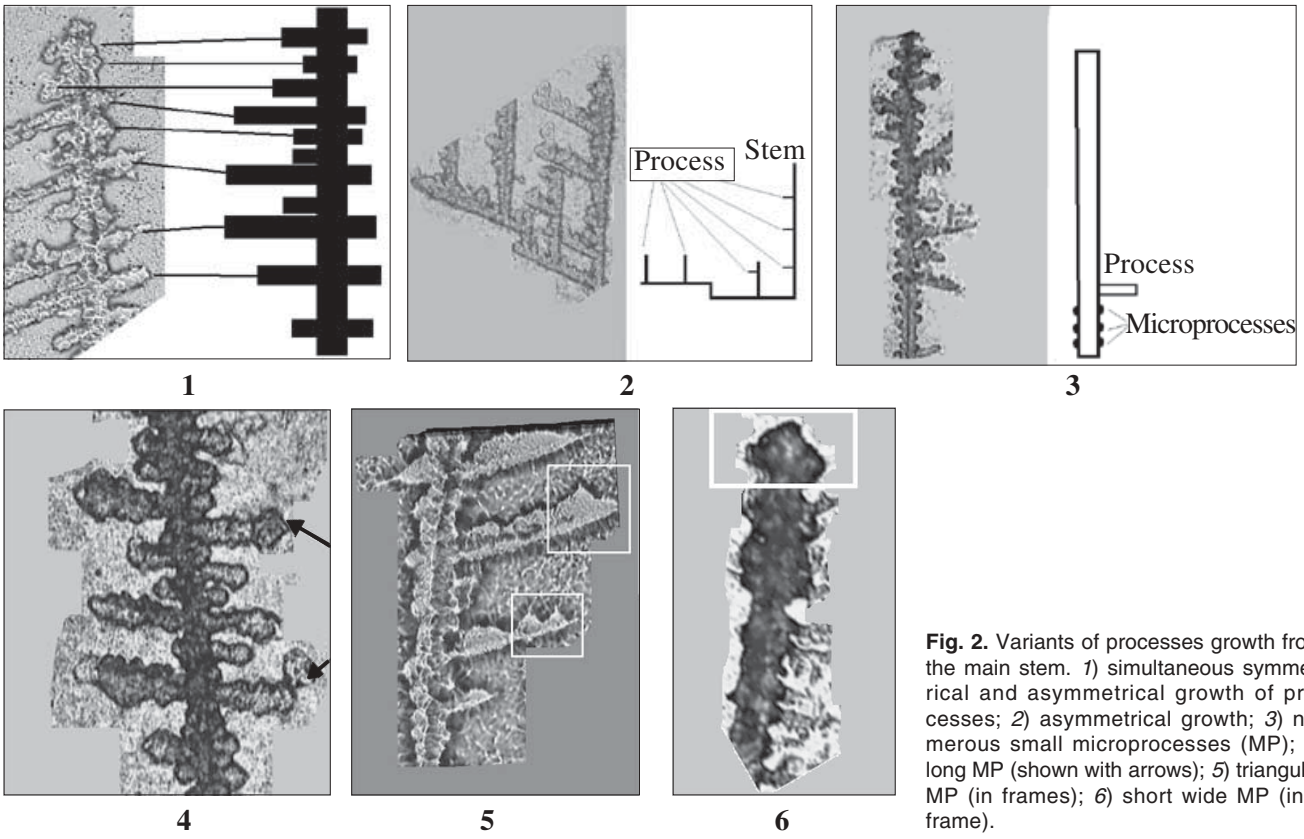

Fig. 2. Variants of processes growth from the main stem. *1*) simultaneous symmetrical and asymmetrical growth of processes; *2*) asymmetrical growth; *3*) numerous small microprocesses (MP); *4*) long MP (shown with arrows); *5*) triangular MP (in frames); *6*) short wide MP (in a frame).

The MBS growth is straight in the majority of cases, sometimes with distortions (Fig. 1, *12*) or doubling at the ends (Fig. 1, *13*). Crystals with blurred contours (Fig. 1, *14*), thinned crystals (Fig 1, *15*), crystals with lamellar structure or structure of different density (Fig. 2, *16*) can form during crystallization; sometimes a crystal can look fragmented (Fig. 1, *2*, *17*). The length and width of MBS can also vary greatly (Fig. 1, *18a* and *b*).

Processes start growing from MBS during dendritic growth of a crystal. It is a probability process depending on the conditions of crystallization; several variants of this process are probable.

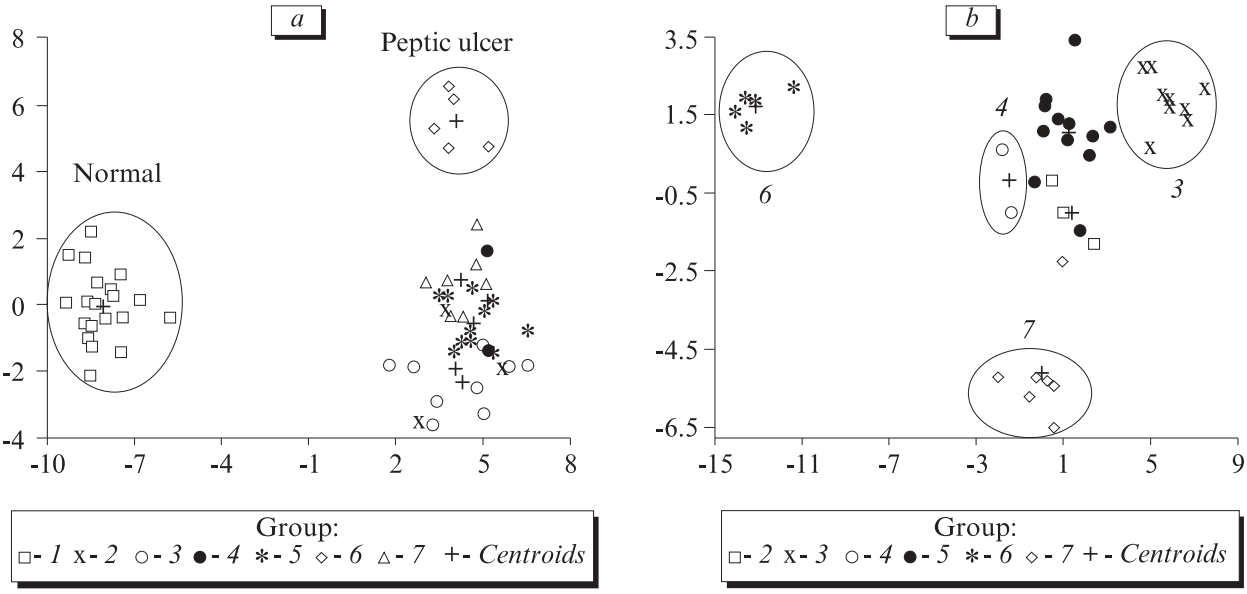

Fig. 3. Dissemination of morphological signs of crystallograms of normal subjects and patients with gastrointestinal diseases. Discriminant analysis, two-dimensional diagram. Abscissa: first discriminant function; ordinate: second discriminant function. *1*) normal; *2*) gastroduodenitis; *3*) chronic pancreatitis; *4*) duodenal ulcer; *5*) cholecystitis; *6*) peptic ulcer; *7*) chronic gastritis. "+": center of a cluster.

The result of the crystal morphogenesis can be evaluated by the following characteristics:

1) by the mode of branching. Two transverse processes are most often formed symmetrically and one process asymmetrical (Fig. 2, *1*). "Asymmetry" of crystal branches is sometimes observed: transverse processes from MBS growing from only one side (Fig. 2, *2*);

2) by the length of processes. Numerous microprocesses are situated on the MBS body (Fig 2, *3*), including long transverse microprocesses (Fig. 2, *4*);

3) by the ends of processes. Triangular microprocesses (Fig. 2, *5*) or short wide processes (Fig. 2, *6*) can form.

All qualitative data on morphological signs are introduced in the program of multidimensional statistical analysis (in our case Statgraphics Plus 5.0) and discriminant analysis is carried out. The "norm" forms an independent clearly distinguished group not crossing over other classes (Fig. 3, *a*) during solution of the two former discriminant functions of the data massive. Moreover, these data are very distant from all other classes by the first discriminant function. The group of patients with peptic ulcer forms one more independent class.

Analysis of the data massive from patients with gastrointestinal diseases by means of the multidimensional statistics method distinguished the majority of diseases (Fig. 3, *b*). Peptic ulcer and chronic gastritis are characterized by a very high degree of signs discrimination. The grouping of signs in patients with chronic pancreatitis and duodenal ulcer is less demonstrative. Such a disease as gastroduodenitis could not be distinguished as an independent group.

Hence, staged morphological analysis of crystals formed from salivary pools helps to create a base of morphological signs. Subsequent multidimensional statistical analysis of this database helps to clearly discriminate between health and disease and even diagnose some diseases [5,7].

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