# Analysis of Motion of Dynamic Scenes in Microscopy Images: Formalization, Criteria and Results

Olga Nedzved Belarusian State University Minsk, Belarus onedzved@bsu.by

Igor Gurevich Federal Research Center "Computer Science and Control" of RAS Moscow, Russia igourevi@ccas.ru Vera Yashina Federal Research Center "Computer Science and Control" of RAS Moscow, Russia werayashina@gmail.com

Ren Tiaojuan Zhejiang Shuren University Hangzhou, China rentj@zjsru.edu.cn Sergey Ablameyko Belarusian State University Minsk, Belarus ablameyko@yandex.by

Ye Fangfang Zhejiang Shuren University Hangzhou, China cliney@zju.edu.cn

Abstract. In this paper, we formalize the problem of the motion analysis of dynamic objects and scenes based on the algorithms and methods developed by the authors for analysis of the cell population behavior. Cell population is considered as a system of dynamic objects and motion is analyzed by using concept of an integral optical flow. On the base of the main types of motion, the key points of cell movement in the population are identified and stages of cell development and interaction are described. The formalization of operations on dynamic objects has been completed.

#### Keywords: dynamic object, scene, motion

#### I. INTRODUCTION

In the study of living cells, time-lapse microscopy is important. This is a sequential recording of microscopic images in long-term monitoring systems for the observation and analysis of the cell population in vitro, which allows to study the cell dynamics in detail.

Video-microscopy can be considered as one of the types of time-lapse microscopy. Its advantages include high temporal resolution and the ability to shoot continuously over long periods of time. Videomicroscopy allows you to obtain frame-by-frame recording of changes in the shape and mobility of living cells, as well as the brightness of their images. Cell motion can be described based on dynamic objects [1].

The currently existing technologies for video sequences analysis are focused mainly on the motion of individual objects rather their moving aggregates, which combine the motion of the entire system with the motion of its components [2].

One of the ways to track the motion of cells is the method of tracking dynamic objects, which is a continuous determination of the position of the object [3, 4]. In this paper, we analyze the motion of dynamic objects and scenes, formalize this process and demonstrate the results.

#### II. DYNAMIC OBJECTS AND SCENES

An elementary dynamic object is a small movable localized object with physical parameters such as volume, density or mass. Their motion can be rotational, rectilinear, accelerated, or even barely noticeable. The complexity of detection and tracking is determined by their size, change in shape, and the nature of motion.

Dynamic objects in microscopic images can be divided into the following classes:

 individual small objects that are a movable component of the background and can be removed when sorting by size,

– large background components with small displacement due to stochastic motions in a specimen,

- fragments of the environment around moving objects that are changed due to their optical characteristics.

Cells in microscopic specimens are mobile objects, three types of cell motility can be distinguished:

- real cell motion,

- displacement of intracellular structures,

- changes in cell shape.

Dynamic objects form scenes. The scene can be defined as static, with the added time variable. 3D motion is estimated by modeling forward and backward flow of objects as dense three-dimensional vector fields.

A continuous scene can be represented as a 5D vector function, where the input determines a three-

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dimensional location x = (x, y, z) and the viewing direction 2D ( $\theta$ ,  $\varphi$ ). The output corresponds to the color of the pixels and can be defined as c = (r, g, b) and volume density  $\sigma$ . In practice, the direction is expressed as a three-dimensional vector in Cartesian space d. This continuous 5D representation of the scene can be approximated by a network MLP F  $\Theta$  : (x, d)  $\rightarrow$ (c,  $\sigma$ ), where  $\Theta$  means the weight to match each 5D input coordinate with the corresponding volume density and corresponding color. Dynamic objects can be described in the same way.

The key problem of monitoring a dynamic scene is to separate the background and objects that can be static or change over time. In this case, the direction of motion plays an important role, it can be divided into the following levels:

- background motion generated by camera motion;

- background motion formed by a change in the surrounding space;

- object motion;
- motion inside objects;
- motion of groups of objects.

Thus, in the first step, it is necessary to divide the image field into different types of motion. For this purpose, motion maps based on optical flow calculations can be used [5].

Obviously, for a uniform motion of the optical system, the background is formed in the form of a constant flow, which has a unique image. Depending on the motion of the camera, the image has its own unique characteristics as in Fig. 1.



Forward movement of the camera

Movement of the camera to the right

Fig. 1. Direction of the optical flow field when the camera moves

The behavior of system of objects is determined by group motion. In this case, motion maps are used, they determine general trends and can indicate individual events in motion [6]. Motion mapping based on the summation of vectors in a local area of the image. But simple summation can lead to the same values for different motion patterns, as in Fig. 2. Thus, the result of the movement must be determined using several different maps.



Fig. 2. Two cases when the adder gives the same result for different patterns of motion

It is assumed that these directions of movement are spatially integrated by local cells in the image space in the same way as provided in the template model in Fig. 3.



Fig. 3. Scheme of the model of building motion maps. The optical flow field is converted to a vector map. The model cells form the setting of the Gaussian direction of movement in this coordinate system. Adders determine the directions of movement based on the direction of the cells

The fields in the flow analysis model combine vectors that encode similar directions of motion in the locally generalized space. The model assumes the selectivity of traffic flows, which is based on the properties of the local field, rather than on complex and specialized interactions. Model local cells calculate the main directions of optical flow. In particular, at each position the cell has a generalized field corresponding

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to the main direction of movement. This field makes each cell less sensitive to deviations from its main direction of motion. The local field model summarizes data with the same preferred direction of movement in a spatial area around their center, as in Fig. 3. In addition, the model forms the probability of the direction of motion. Thus, based on the summation of vectors, it is possible to generate motion maps that allow to determine the contours of a dynamic object and the types of its motion.

# III. MONITORING OF DYNAMIC OBJECT MOTION

The general scheme for analyzing images of dynamic objects includes five stages (Fig. 4):

- image capture and preprocessing;

- segmentation of the image scene and selection of cells areas;

- characteristics measurement;

- definition of laws for the formation of a general description of cell motion;

- cell state classification.

The most important characteristics for describing dynamic objects in microscopic images are: position characteristics (coordinates, speed of motion, direction change, trajectory), morphological characteristics (size, shape, degree of object shape change), optical characteristics (brightness, color, optical density), and also the duration of the observation.



Fig. 4. General scheme of video processing of movable cells

## IV. FORMALIZATION OF OPERATIONS WITH DYNAMIC OBJECTS

The formalized concept of the structural model of a dynamic object is an ordered set of patterns  $A = \langle A_1, A_2, ..., A_n \rangle \subseteq \Omega_n$ , which corresponds to the total structural description *A*, if object pattern  $A \in \Omega$  can be completely uniquely reconstructed by combining elements from *A*, which formally can change over time:

$$A = \delta(A) = A_1 \oplus A_2 \oplus \ldots \oplus A_n, \tag{1}$$

where  $\delta$  – operation of structural reconstruction of the pattern according to the structural description;  $\bigoplus$  – operation of combining patterns from  $\Omega$ , on which, in the general case, no additional conditions are imposed, except that  $\Omega$  closed with respect to  $\bigoplus$ . It should be noted that the effect of blocking some objects by others, typical for images in a video sequence, in contrast to many other areas of application of structural analysis, makes the order of combining elements of the visible scene fundamentally important. Therefore, the operation  $\oplus$  in the general case cannot be either symmetric or associative, although sometimes such a restriction is nevertheless imposed.

Thus, the definition of a model of dynamic objects is performed through a changing preimage L with timeconstant properties, which consists of n components:

$$L = \delta (L) = L1 \oplus L2 \oplus \cdots \oplus L_n.$$
 (2)

In this case, the types of dynamic elements are known, which are specified by the characteristic predicates like elements  $Mi(Li) \in \{0,1\}$ , i=1,...,n. In addition, given m conditions or connections predicates  $Mk(L) \in \{0,1\}$ , k=1,...,m. Then the preimage model takes the form:

$$M(L) = M_1(L_1) \cdot \dots \cdot M_n(L_n) \cdot M_1(L) \cdot \dots \cdot M_m(L).$$
(3)

The variables n and m are considered to be timevariable parameters that are also subject to optimization, and predicates are considered as probabilistic or fuzzy, taking values by [0,1]. In this case the problem of structural segmentation of dynamic objects will correspond to the most general case of structural image analysis.

The motion description of objects system can be performed not only at the pixel level, but also at the region level. The characteristics for describing the movement at the region level are the direction of movement, the speed of movement and the intensity of movement in the region determined on its basis.. The determination of motion at the region level and the corresponding motion maps are described in [5, 6].

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The motion maps are used to determine the type and directions of motion in a video sequence and to determine the number of pixels moving in the selected directions. In this case, the motion is described for all nodes (hypothetical centers of motion) through which the moving pixels pass. Thus, the dynamic object model is defined as:

$$A = \delta (A) = OP \oplus OCM \oplus ICM \oplus OQ \oplus IQ.$$
(4)

Here OP, OCM, ICM, OQ, IQ are corresponding motion maps. Thus, the simple motion of a dynamic object can be described using the concepts of an algebraic field and a ring.

There are three types of dynamic objects motion: directional motion, aggregation (moving towards a common center), and scattering (moving away from the center). The signs of aggregation are: a) several objects move to one image region from other areas; b) the speed of motion of these objects is greater than the speed of chaotic motion; c) at least two predominated directions of movement can be distinguished. The signs of scattering include: a) several objects move in the direction from their common center to other regions of the image; b) the speed of their motion exceeds the speed of chaotic movement; c) at least two directions of movement predominated can be distinguished.

For the last two types of motion, it is possible to define the algebraic concepts of a group and a ring based on the presence of a hypothetical center of motion. In this case, by analogy, we can define the additive operation for the group  $C(q^i, \vec{v}, \vec{a})$ , with the displacement operation specified on it  $\{\circledast\}$ :  $C + C \rightarrow C$ . Thus, it is possible to define an algebraic element as a motion towards the center, a neutral element as the absence of motion and an inverse element as a motion from the center. The associativity property is present in this case.

Thus, relative to motion, algebraic groups of events can be distinguished as simple motion and motion relative to a hypothetical center. It is allows to consider the problem of motion from an algebraic point of view with the vector as the reference concept. Pixel displacement vectors  $\delta q^i$  can be obtained based on the calculation of optical flows. The vectors of the integral optical flow are additionally used  $\tilde{q}^i$ . Therefore, the displacement vector obtained on the basis of the integral optical flux can be expressed as:

$$\tilde{q}^i = \sum_j \quad \delta q^i_{\ i},$$

where *j* is frame change index in the video sequence.

# V. PRACTICAL RESULTS

As experiments, the movement of vesicles containing the GLUT4 protein onto the cell membrane was considered. GLUT4, an insulin-regulated glucose transporter protein, is predominantly found in the cytoplasm of adipose and muscle cells in the absence of insulin. Understanding the effect of insulin on the spatio-temporal regulation of intracellular GLUT4 transport is important for elucidating the pathogenesis of type 2 diabetes in humans.

The GLUT4 intracellular transport movement dataset was derived from a total internal reflection fluorescence microscopy (TIRF) image sequence. TIRF microscopy is an optical technique used to observe the fluorescence of individual molecules, based on the phenomenon of total internal reflection. Parameters such as the speed of movement of vesicles in the nearmembrane region, their number and density were determined for the vesicles.

Registration of the appearance of vesicles is carried out on binary images obtained because of threshold segmentation, which is the result of calculating the difference between the brightness of the images of the vesicles and the background. Segmentation consists in the fact that the brightness value of each pixel is compared with a threshold value, which is determined based on the correspondence of its local environment to the Gaussian distribution and a predetermined user correction to determine the deviation from the Gaussian distribution.

The selected dynamic objects are controlled by means of motion maps, which make it possible to form a description and conduct monitoring at the level of the graph structure.

The construction of a set of protein transport graphs in cells corresponding to a set of all vesicles isolated at the segmentation stage consists of the following steps:

1) Selection of the initial branches of the graphs in accordance with the number of vesicles.

2) Splitting the video sequence into minimum time intervals is performed for the entire video sequence. The time for each event is determined, and then the minimum time interval according to which the timeline is split.

3) Construction a set of graphs (Fig. 5). For each node, the state of the cell is registered, according to which the number of inputs-outputs for each block is determined. The simple motion block has one input and one output, the intersection block has one input and two outputs, the membrane delivery block is the terminator of the graph branch. Thus, each branch of the graph represents a family tree for an individual vesicle.

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Fig. 5. A set of graphs describing the movement of individual vesicles

The use of graphs allows tracking the stages of GLUT-4 delivery in the cell. It also reduces the analysis time based on the transformation of dynamic objects into dynamic groups on the frame of the video sequence as in Fig. 6.



Fig. 6. Combining vesicle tracks into dynamic groups on a video sequence frame

# VI. CONCLUSION

The paper formalizes the problem of the motion analysis of dynamic objects and scenes based on the algorithms and methods developed by the authors for analyzing the behavior of the cell population as a system of dynamic objects, which are based on the use of the concept of an integral optical flow. The main types of movement have been determined, which make it possible to distinguish the key moments of the movement of cells in a population and describe the stages of development and interaction of cells. The formalization of operations on dynamic objects has been completed.

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