ACTA STEREOL 1982; 199-202 STEREOL 82 SHEFFIELD

MEASUREMENT OF RESPONSE FUNCTION IN CELL BEHAVIOR BY LASER FOURIER TECHNIQUE

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ABSTRACT

When a laser beam is fired at a moving cell, the diffraction pattern created corresponds to a Fourier transform of the scattering density of the moving cell. A stimulus applied to the cell causes several types of reaction which can be separated as factors in the Fourier representation. Deformation of motile organelles causes change in the diffraction pattern, but its translation causes no change. This enables the response function of motile organelles to be identified, when they may be hidden by changes of position.

INTRODUCTION

The Fourier technique has been developing from classical crystal stereology that Raue, Friedrich and Knipping, and Bragg established by means of X-ray diffraction. The distribution of diffraction fringes on two-dimensional reciprocal range are turned to the exact distance between lattice surfaces on three-dimensional pictorial domain by Fourier analysis [Bragg, 1913]. Recently, laser with high monochromity, high coherency and high intensity is diffracted to fine pattern not only by exact periodic structure but also by irregular. Remarkably, when a cell is swimming in a field of laser beam, the diffraction pattern created corresponds to Fourier transform of the image of the swimming cell. In this method, structural change of the motile organelles and the resultant translational and rotational

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movement of the whole cell body are soparated [Ishizaka, 1981]. This is a new face of stereology that bridges between structure and function [Weibel, 1969].

In this paper, it is stated that another bridge from structural change to the inducing stimuli is real-time imaged by Fourier transform method, to analyze the response function in cell behavior.

RESPONSE FUNCTION IN CELL BEHAVIOR

A structural change of motile organelle, $\stackrel{\wedge}{,}$, $M(t, \vec{x})$, is induced by a stimulus from the environment, $S(t, \vec{x})$. It was remarked that there was a time-lag, t-t', and a dislocation, $\vec{x}-\vec{x}$, between the structural change and the inducing stimulus. When the stimulus is experimentally given at a time t' on a locus \vec{x} of the cell, the structural change $\stackrel{\wedge}{,}$, $M(t, \vec{x})$ does not always occur at the same time and locus, it usually occurs after a time t' on the other loci. The response relation between the structural change and the inducing stimulus,

 $\stackrel{\Delta}{\xrightarrow{+}} M(t, \vec{x}) = R(t-t^2, \vec{x}-\vec{x}) S(t^2, \vec{x})$

was connected by a function of both the time-lag, t-t', and the dislocation, $\vec{x} - \vec{x}'$. $R(t-t', \vec{x} - \vec{x}')$ is called the response function.

Conversely, the structural change at a time t on a locus \vec{x} arises by integrating response to stimuli before the time t on every locus of the cell,

$$M(t, \vec{x}) = \begin{array}{c} \sum & \sum & \Delta \\ t', \vec{x}, & t', \vec{x} \end{array} M(t, \vec{x}) \\ = \int_{-\infty}^{t} \int_{-\infty}^{\infty} R(t-t', \vec{x}-\vec{x}) S(t', \vec{x}) dt' d\vec{x} \end{array}$$

It is found that the equation expresses the mechanism of cell behavior.

SEPARATION BETWEEN STRUCTURAL CHANGE AND TRANSLATION AND ROTATION

In cell behavior, structural changes of motile organelles effect translational, \vec{x}_{T} , and or rotational, \tilde{R} , movement of the whole cell body. We describe the structural change as a function M (t, \vec{x}_{i}) of time, t, and the coordinates of central mass system of the cell, \vec{x}_{i} , and also as a function of time and observational coordinates, M(t, \vec{x}).

$$\vec{x}_{1} = \vec{R}(\vec{x} - \vec{x}_{T})$$

The Laser Fourier transform turns the image of the structural change from the pictorial domain (time,t, and locus, \vec{x}) to the reciprocal range (frequency, ω , and wave vector, \vec{k}) by interfero-micro-diffractometry [Ishizaka,1981].

$$\begin{split} \hat{\mathbf{M}}(\boldsymbol{\omega}, \vec{\mathbf{k}}) &= \int_{-\infty}^{\infty} \int_{-\infty}^{\infty} \mathbf{M}(\mathbf{t}, \vec{\mathbf{x}}) \exp \mathbf{i}(\boldsymbol{\omega} \mathbf{t} - \vec{\mathbf{k}} \vec{\mathbf{x}}) \, \mathrm{d} \mathbf{t} \, \mathrm{d} \vec{\mathbf{x}} \\ &= \int_{-\infty}^{\infty} \int_{-\infty}^{\infty} \mathbf{M}_{\mathrm{s}}(\mathbf{t}, \vec{\mathbf{x}}_{\mathrm{i}}) \exp \mathbf{i}(\boldsymbol{\omega} \mathbf{t} - \vec{\mathbf{k}} \vec{\mathbf{x}}) \, \mathrm{d} \mathbf{t} \, \mathrm{d} \vec{\mathbf{x}} \\ &= \int_{-\infty}^{\infty} \int_{-\infty}^{\infty} \mathbf{M}_{\mathrm{s}}(\mathbf{t}, \mathbf{\widetilde{R}}(\vec{\mathbf{x}} - \vec{\mathbf{x}}_{\mathrm{T}})) \exp \mathbf{i}(\boldsymbol{\omega} \mathbf{t} - \vec{\mathbf{k}} \vec{\mathbf{x}}) \, \mathrm{d} \mathbf{t} \, \mathrm{d} \vec{\mathbf{x}} \\ &= \exp -\mathbf{i} \mathbf{k} \vec{\mathbf{x}}_{\mathrm{T}} \, \hat{\mathbf{M}}_{\mathrm{s}}(\boldsymbol{\omega}, \mathbf{\mathbf{\widetilde{R}}} \vec{\mathbf{k}}) \\ |\hat{\mathbf{M}}(\boldsymbol{\omega}, \mathbf{\vec{k}})|^{2} &= |\hat{\mathbf{M}}_{\mathrm{c}}(\boldsymbol{\omega}, \mathbf{\mathbf{\widetilde{R}}} \vec{\mathbf{k}})|^{2}, \end{split}$$

separating the structural change from the translational movement of the whole cell body.

Evidently, the separation was illustrated by a experiment, as shown in Figure 1. One TV camera caught normally pictorial image of the swimming cell (O in Figure 1.) and the other camera reciprocally diffraction pattern (D in Figure 1.). Both were superimposed on the optical axis, and mixed on one CRT. The image of the cell moved about in the picture, but the diffraction pattern never translated, although, deformed and/or rotated about optical axis. It was found that the structural change is free from the translation.



Figure 1. Pictorial image and its diffraction pattern superimposed on a picture [Ishizaka & Yamashita,1980]

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Insertion of reversely rotation k-mirror separated the diffraction pattern of the structural change from the rotation,

 $|\hat{\mathbf{M}}_{\mathbf{S}}(\omega, \widetilde{\mathbf{R}} \mathbf{k})|^2 \Rightarrow |\hat{\mathbf{M}}_{\mathbf{S}}(\omega, \mathbf{k})|^2.$

Thus a bridge between the cell movement and the structural change has been constructed by interfero-microdiffractometry.

DECONVOLUTION OF RESPONSE FUNCTION BY FOURIER TRANSFORM

The equation of mechanism of cell behavior was Fourier transformed. The left side of the equation has already been obtained by Laser Fourier transform. The right side of the equation factorizes. It is found that the Fourier transform of structural change is equal to the product of the Fourier transform of response function and the Fourier transform of the inducing stimulus,

 $\widehat{\mathbf{M}}(\boldsymbol{\omega}, \vec{\mathbf{k}}) = \widehat{\mathbf{R}}(\boldsymbol{\omega}, \vec{\mathbf{k}}) \quad \widehat{\mathbf{S}}(\boldsymbol{\omega}, \vec{\mathbf{k}}).$ Algebraically, this is $\widehat{\mathbf{R}}(\boldsymbol{\omega}, \vec{\mathbf{k}}) = \widehat{\mathbf{M}}(\boldsymbol{\omega}, \vec{\mathbf{k}}) / \widehat{\mathbf{S}}(\boldsymbol{\omega}, \vec{\mathbf{k}}).$

The inverse Fourier transform leads to the response function

$$R(t,\vec{x}) = \int_{-\infty}^{\infty} \int_{-\infty}^{\infty} \hat{R}(\omega,\vec{k}) \exp -i(\omega t - \vec{k}\vec{x}) d\omega d\vec{k}$$

$$= \int_{-\infty}^{\infty} \int_{-\infty}^{\infty} (\hat{M}(\omega,\vec{k}) / \hat{S}(\omega,\vec{k})) \exp -i(\omega t - \vec{k}\vec{x}) d\omega d\vec{k}$$

Therefore, the response function in cell behavior can be identified from the Laser Fourier transform of the structural change divided by the Fourier transform of the stimuli experimentally given, by this Fourier transform method.

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