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A DEVICE FOR UNIAXIAL ORIENTATION OF GELATINOUS MEDIA AND FOR MEASURING THE ANISOTROPY OF THEIR PHYSICAl PROPERTIES

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The design characteristics of the device suggested here provide the possibility of accomplishing uniaxial orientation of gels with retention of the constancy of their volume, thus making it possible to carry out highly accurate measurements of their anisotropic physical parameters in directions parallel and perpendicular to the axis of orientation of the gel. Using as examples polyacrylamide gels that contain biologically important molecules and membranes, we demonstrate the possibility of measuring the anisotropy of their luminous absorption and refraction.

Important information on the structure of molecules and microparticles in gelatinous media can be obtained by investigating the anisotropy of their physical properties in an orientationally ordered state. It is known that under the action of shear stresses anisometric molecules and microparticles in a liquid or gelatinous medium can align in a laminar hydrodynamic flow [1]. Here the anisotropy of thermophysical, mechanical, and optical properties of the medium is observed.

The description of numerous devices for uniaxial orientation of liquid and gelatinous media is available (such as [2-11]). They have different efficiencies and different measuring capabilities, but they do not allow one to measure the physical parameters of a specific aligned specimen in directions parallel and perpendicular to the axis of its orientation. The proposed device – a cuvette – provides the possibility of carrying out simultaneously the orientation of gels and the above-indicated measurements.

The basic structural elements of this device are shown in Fig. 1. Here 1, 2 and 3, 4 are pairwise parallel and coaxial windows. Windows 2, 3,4 are made in pistons 5, 6, 7 in such a manner that they can move in the mutually perpendicular directions $A-A$ and $B-B$. The components of the gel and the molecules or microparticles under investigation are subjected to joint polymerization directly in the measuring volume (V) of the cuvette. The orientation of the specimen obtained is carried out by moving piston 7 toward the inside of the measuring volume along the $B-B$ axis. Here windows 1, 2 slide apart along the $A-A$ axis of gel orientation.

A necessary condition for efficient operation of this device is continuous contact of piston 7 with pistons 5, 6 in order to ensure closure of the measuring volume. To perform highly accurate measurements, it is necessary that the gel should not crack in orientation, that it could not withdraw from windows 1-4, and, which is extremely important, that provision be made that the measuring layer of the gel should be plane-parallel simultaneously in the directions $A-A$ and $B-B$. All this can be fulfilled only if in the process of simultaneous movement of pistons 5, 6, 7, by which the gel is oriented, the initial volume of the latter remains unchanged. We will try to find a geometric shape for the profile of the contact surfaces of pistons 5, 6 such that the volume V would be constant.

By virtue of the design characteristics of the device, the problem of determining a geometric shape for the profile of the contact surfaces is plane and symmetric with respect to windows 1, 2 (see Fig. 1). Suppose x and y are the coordinates of the point of contact of piston 7 with piston 5 in the coordinate system XY; l, h are the distance from the plane of window 4 to the lower and upper points of contact between piston 5 and piston 7, respectively; m is the thickness of piston 5; d is the thickness of piston 7. The condition of invariance of the volume of the gel in its deformation orientation is described by the equation (see Fig. 1)

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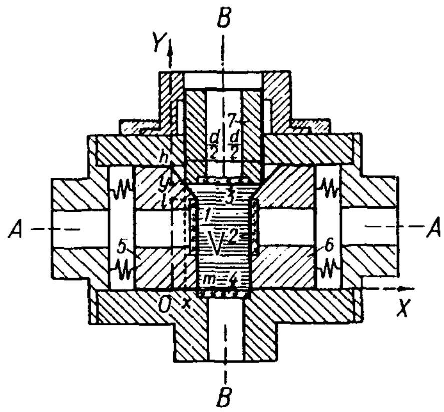

Fig. 1. Basic design characteristics of the cuvette for orientation and measurement of the anisotropic physical properties of oriented gelatinous media (a central cross section).

$$V = 2\left(\frac{d}{2}y - \int_x^m y\,dx\right) = \text{const}\,. \tag{1}$$

Differentiation of this equation with respect to x gives the equation $y + (2/d)y = 0$, whence

$$y = C\exp\left(-\frac{2x}{d}\right), \tag{2}$$

and the constant $C = h$ (its value can be found from the boundary conditions $x = 0$, $y = h$). Thus, the desired mathematical form of the profile of the contact surface of pistons 5 and 6 in the coordinate system XY (see Fig. 1) is described by the equation

$$y = h\exp\left(-\frac{2x}{d}\right). \tag{3}$$

A relationship between the geometric parameters of the cuvette can be obtained from Eq. (3) on condition that $x = m$, $y = l$:

$$\frac{h}{l} = \exp\left(\frac{2m}{d}\right). \tag{4}$$

The cuvette was made of D16T-grade Duralumin and had the parameters $m = 10$ mm, $l = 12$ mm, $d = 20$ mm, $h = 32.62$ mm. The windows of the cuvette were manufactured from high-quality KU-grade fused quartz, each having an area of 64 mm^2 with a thickness of 0.5 mm.

We will show the capabilities of this device as applied to the orientation of gelatinous media and to measurement of the anisotropy of absorption and refraction of light waves in oriented polyacrylamide gels that contain molecules of met-hemoglobin and microparticles of membranes of chromatographs. A measure of the anisotropy of absorption of linearly polarized light by a substance is the linear dichroism:

$$P = (D_{\parallel} - D_{\perp})/(D_{\parallel} + D_{\perp}) = (D_{\parallel} - D_{\perp})/2D\,. \tag{5}$$

One of the measures of the anisotropy of light refraction by a substance is the measure of circular birefringence, namely, the optical activity of the substance measured in angular degrees:

$$\Psi = \frac{\pi L(n_+ - n_-)}{\lambda}\,. \tag{6}$$

TABLE 1. Linear Dichroism and Optical Activity of Oriented Gels Containing Molecules and Microparticles

Object under unvestigation	Measurements in the direction of passage of light with respect to the orientation axis of polyacrylamide gel			
	perpendicular		parallel	
	P, units 10^3	Ψ, angular degrees 10^2	P, units 10^3	Ψ, angular degrees 10^2
7% polyacrylamide gel containing intracytoplasmic membranes of a Chromatum minutissimum culture at a concentration of 16 mg/cm^3	5.2(300) 0.0(335) −4.8(371) 0.0(399) 7.5(480) 5.8(585)	0(300) 0(335) 0(371) 0(399) 0(480) 0(585)	0(300) 0(335) 0(371) 0(399) 0(480) 0(585)	0(300) 0(335) 0(371) 0(399) 0(480) 0(585)
5% polyacrylamide gel containing met-hemoglobin at a concentration of 0.4 mg/cm^3	−0.99(402) −1.27(412)	−3.0(398) 0(412) 1.8(422)	0(402) 0(412)	−3.0(398) 9(412) 1.8(422)
3–7% polyacrylamide gel	0 (300–528)	0 (300–528)	0 (300–528)	0 (300–528)

The degree of optical activity was determined using a Spectropol-1 (Fica-France) polarimeter; the linear dichroism was measured on a Jasco-20 (Japan) dichrograph; the isotropic optical density was determined on a Beckman UV-5270 (USA) spectrophotometer. In the measurements procedures developed earlier were used [12-14].

Data on the investigation of the linear dichroism and optical activity of polyacrylamide gels that contain different substances are summarized in Table 1. The values of P and Ψ were measured at the extrema (maximum, minimum, zero) of the functions $P(\lambda)$, $\Psi(\lambda)$ and were normalized to the thickness of the gel layer of 1 cm. The wavelength of the transmitted light (in nanometers) is given in brackets. The measurements conducted allow a conclusion on the angles of mutual orientation of the dipole moments of transitions of the pigments, which play an important role in photosynthesis and which enter into the composition of chromatophore membranes, and on the presence of impurity substances in preparations of met-hemoglobin, and they make it possible to evaluate their concentrations. This information is required for calculating the structural organization of membranes and for monitoring the purity of preparations of the hemoglobin series. Similar investigations were carried out by us for polyacrylamide gels containing a large class of medicinal preparations and pectin substances.

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NOTATION

$D_{||}$, $D_{\perp}$, optical densities of the oriented substance for linearly polarized light passing through the substance with the polarization vector parallel ($||$) and perpendicular ($\perp$) to the orientation axis; D, isotropic optical density; n_+, n_-, refractive indices of the substance for light passing through it and polarized around the circle to the left (+) and to the right (−); λ, light wave length; L, thickness of the layer of the substance.

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