ACCESSIBILITY OF HYDROXY GROUPS OF PECTIN SUBSTANCES FOR DEUTERATION

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An apparatus for the deuteration of solid samples and for recording their IR spectra in a current of D_2O vapor, and an IR-spectroscopic method of determining the degree of deuteration (q) of the hydroxy groups of pectin substances (PSs) are described. It has been established that there are no zones of structurally different nature in films of derivatives of PSs at the carboxy group. The rate of deuterium-exchange of the water of hydration of PSs is higher than for the hydroxy groups of the pyranose rings. The rate of deuterium exchange depends on the density of packing of the polymer chains of the pectin derivatives.

The capacity of the hydroxy groups of pectin substances (PSs) for exchanging their hydrogen with deuterium can be used, as in the case of any polysaccharides (PSCs) for determining ordered regions in the reticular structure of the polymer formed as the result of intermolecular interactions and to identify in the IR spectrum the frequencies due to vibrations with a contribution to the distribution of the potential energy of a proportion of the OH groups.

Methods are known for deuterating PSCs with D_2O vapor and liquid [1]. The degree of deuteration of the OH groups is calculated from the ratio of the optical densities at the absorption maxima $\nu(OD) = 2530\ cm^{-1}$ and $\nu(OH) = 3360\ cm^{-1}$ [2] or from the ratio of the areas of these bands [3]. However, for example, in the IR spectrum of partially deuterated cellulose, several bands relating to $\nu(OH)$ have been detected [4]. In such a case the determination of the degree of deuteration according to [2] loses its sense. A necessary condition of those methods [2, 3] is the elimination of D_2O residues from the cell after the end of deuteration; otherwise its absorption will mask the $\nu(OD)$ bands of the PCS. All this limits the use of the given procedures for investigating the process of deuteration in time.

An improved procedure, in comparison with [5], for deuterating PSs in a current of nitrogen saturated with D_2O vapor permits the rapid achievement of the maximum substitution and enables the dynamics of deuteration to be followed.

The apparatus (Fig. 1) consists of a glass distributor with five cocks (1-5), a flask for the liquid reagent (8) with cocks (6, 7), a tube furnace (9), and gas cells (14, 16). All the elements are connected by flexible tubing, which ensures the lability of the apparatus. The quartz tube furnace (9) is connected with the measuring cell (14) by means of a spherical ground joint (10). A rotameter (18) is included to regulate the rate of flow of the mixture of gases. The temperature in the cell (14) which is measured by the thermometer (19) the block of which is placed between the cells, is regulated by the voltage of the electric current to the furnace (9) and by the rate of flow of the mixture of gases. If it is necessary to dry the sample (12) before deuteration, dry nitrogen is passed through the system with the open cocks (1, 11, 13, 15, and 17). The dehydration of the sample is monitored from the change in the optical density of the $\delta(H_2O)$ or $\nu(OH)$ bands. With cocks (2, 3, 6, 7, 11, 13, 15, 17) open and flask (8) in an inclined position, nitrogen, passing through D_2O, is saturated with its vapor, bathes the sample (12), and entrains the water formed on deuterium exchange. As can be seen from Fig. 1, the system of cocks permits any part of the apparatus to be evacuated without affecting the hermetic sealing of the apparatus while vessels with other reagents are connected operatively through the reserve cocks (4, 5).

The process of deuterating the OH groups can be followed from the change in the spectrum without stopping the flow of D_2O vapor through the cell.

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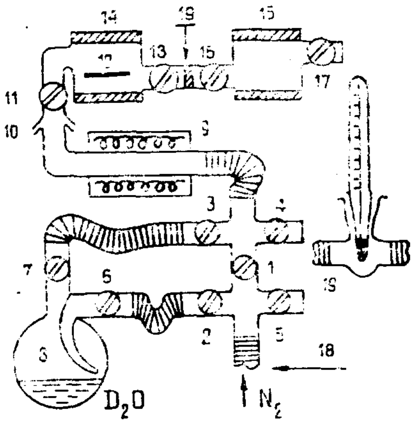

Fig. 1

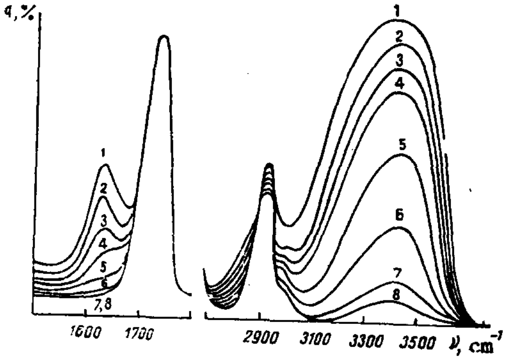

Fig. 2

Fig. 1. Sketch of the apparatus for deuterating films in D_2O vapor (explanation in the text).

Fig. 2. IR absorption spectra of films of CH_3-PS with different degrees of deuteration (q): 1) 0; 2) 0.18; 3) 0.33; 4) 0.43; 5) 0.64; 6) 0.84; 7) 0.93; 8) 0.97.

The proposed method has permitted the complete deuteration of the hydroxy groups of films of pectic acid and of its derivatives at the carboxy group to be achieved. At any degrees of deuteration of PSs (Fig. 2) no well-defined structure is observed in the broad ν(OH) band. This has served as a basis for assuming the structural homogeneity of pectin films and the possibility of determining the degree of deuteration of the OH group by measuring the optical density D at any point of the spectrum — for example, at the maximum of the new (OH) band.

If the degree of deuteration of the OH groups is denoted by q, and if $Q = q_{\max}$ (the maximum achievable value of q in a given experiment, $Q \leq 1$) and D' is the optical density relative to the base line passing through the 3800 cm⁻¹ point parallel to the axis of abscissas, then

$$Q = \frac{D'(0) - D'(Q)}{D'(0)}, \tag{1}$$

$$q = \frac{D'(0) - D'(q)}{D'(0)}, \tag{2}$$

where D'(0) applies to the undeuterated sample.

In the spectra of the initial and the deuterated samples, the base lines do not usually coincide, which is probably connected with a change in the light scattering of the film during deuteration. In an investigation of the dynamics of deuteration, when the process can be followed from the change in D only at some particular point* of the spectrum, the position of the base line can be determined only for the initial and final states, when it is possible to record the complete spectrum. For other, intermediate, values of q in the investigation of the dynamics of deuterium exchange we calculated the correction for the change in the position of the base line. For this we first recorded the complete spectra of the initial state, a number of fixed intermediate states, and the final state of the PS and plotted the dependence of the optical density at 3800 cm⁻¹ on q: $D_0(q)$. It was found that for the derivatives that we investigated it was linear:

$$D_0(q) = D_0(0) + nq, \quad \text{where } n = \frac{\Delta D_0}{\Delta q}. \tag{3}$$

On the basis of the linear relationship (3) in the investigation of the dynamics of deuteration n can be determined from the values of D_0 for the initial and final states alone:

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*What are under consideration are ordinary spectrometers, the rate of scanning of the spectrum of which is comparable with the rate of deuterium-exchange.

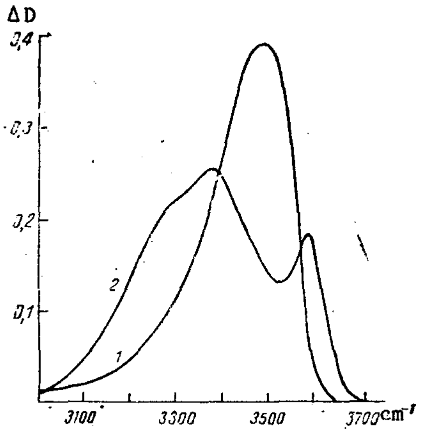

Fig. 3

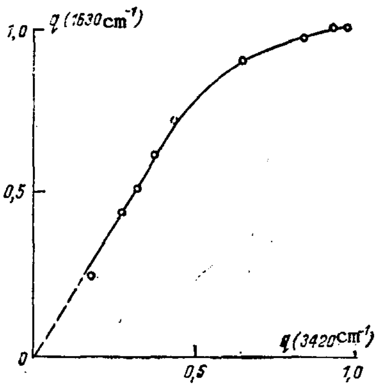

Fig. 4

Fig. 3. Differential IR absorption spectra in the ν(OH) region: 1) films of CH_3-PS (difference between the spectra of an anhydrous and a completely deuterated film); 2) water of hydration in a film of CH_3-PS (difference between the spectra of an air-dry and an anhydrous film.

Fig. 4. Isochronous values of the degree of deuteration of an air-dry film of CH_3-PS calculated from D(1635) and D(3420).

$$n = \frac{D_0(Q) - D_0(0)}{Q}, \tag{4}$$

$$D'(q) = D(q) - D_0(q), \tag{5}$$

where D(q) is relative to the line T = 100%.

Substituting (3) in (5), and then (5) in (2), we obtain:

$$q = \frac{D'(0) - D(q) + D_0(0) + nq}{D'(0)},$$

$$q = \frac{D'(0) - D(q) + D_0(0)}{D'(0) - n}.$$

Since $D'(0) + D_0(0) = D(0)$, then

$$q = \frac{D(0) - D(q)}{D'(0) - n}. \tag{6}$$

Because the absorptions at the maxima of ν(OH) of the ordered and unordered regions of cellulose differ appreciably [4], which indicates a difference in the H-bonds, differences must also be expected in the intensities of these bands. Thus, for PSCs with an inhomogeneous system of H-bonds the probability is not excluded that the values of q and the rate of deuterium exchange determined by measuring D at different points of the ν(OH) band will not be identical.

The rates of deuterium exchange calculated from optical density measurements at various points of the ν(OH) band of PS derivatives at the carboxy group scarcely differ. This confirms the point of view that there are no structurally differing zones in films of PS derivatives and that it is justified to determine q by measuring D at the maximum of the ν(OH) band.

In an investigation of the deuteration of a hydrated film of methyl pectate, CH_3-PS (Fig. 2), it was found that the value of q calculated from the optical densities of the band at frequencies of 3300 and 3520 cm^{-1} differed. The contribution to the optical density at the 3300 cm^{-1} point of the ν(OH) band of water is greater than that of the ν(OH) band of pyranose rings, while the situation at 3520 cm^{-1} is the opposite (Fig. 3), from which follows the conclusion that the rates of deuterium exchange of the water of hydration and of the hydroxyls of the pectin are different.

To confirm this hypothesis, we compared the isochronous values of q calculated from the change in D at the points $\delta(H_2O) = 1635\ cm^{-1}$ and $\nu(OH) = 3420\ cm^{-1}$, partially masked by the

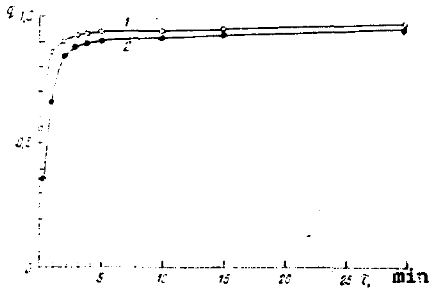

Fig. 5

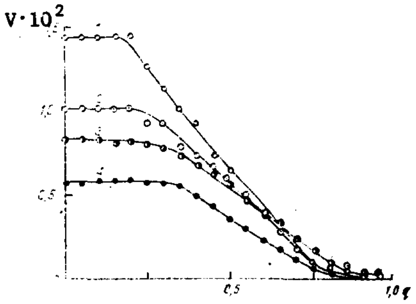

Fig. 6

Fig. 5. Change with time of the degree of deuteration (q) of films of K-PA 3 μm (1) and 8 μm (2) thick.

Fig. 6. Dependence of the rate of deuterium exchange on the degree of deuteration of films: 1) CH_3-PS; K-PA; 2) Ca-PA; 3) H-PA; 4) Cu-PA.

absorption of water. At the same rate of deuteration of water and hydroxy groups of the pyranose rings, the isochronous values of q should lie on a straight line equidistant from the q(1635) and q(3420) axes. The experimental curve (Fig. 4) for values of $q \leq 5$ is displaced closer to the q(1635) axis. Consequently, the rate of deuterium exchange for the water of hydration of pectin is greater than the rate for its hydroxyls. At larger values of q the slope changes in connection with a decrease in the proportion of H_2O in comparison with hydroxy groups.

The process of deuterating anhydrous films of CH_3-PS, of pectic acid H-PA, and of its salts K-PA, Ca-PA, and Cu-PA has been investigated by the method developed. Since the process of deuteration depends on the film thickness (Fig. 5), which is connected with the diffusion of the D_2O molecules into the film, to exclude this effect we studied the dynamics of deuteration in films of the same thickness.

In the initial stage, the rate of deuterium exchange

$$V = \frac{d(1-q)}{d\tau}\ \text{sec}^{-1}$$

is constant (Fig. 6). Here the stage limiting the process is the diffusion of D_2O vapor into the film. Then the second phase begins and the rate of deuterium exchange falls, since it is now limited by the concentration of OH groups. The rate depends linearly on the concentration of OH groups $(1 - q)$ and it is described by the equation of a reaction of the first order relative to $1 - q$. The rate constants of deuterium exchange do not correlate with the values of ν(OH) and with the energies calculated from them [4] of the H-bonds involving the hydroxy groups.

Thus, for example, the rate of deuterium exchange for CH_3-PS and for K-PA are identical, and the positions and intensities of the ν(OH) bands of these derivatives differ substantially [6]. At the same time, at smaller differences in ν(OH) for CH_3-PS and H-PA a difference is observed in the rates of deuterium exchange. These rates for derivatives of PS at the carboxy group are most probably governed by the density of packing of the polymer chains. The structure is loosest in K-PA and CH_3-PS films. In neither case are there carboxyl—carboxyl bonds between the polymer chains. In H-PA, binding takes place through carboxyl—carboxyl interactions with the formation of dimeric groupings and in the salts Ca-PA and Cu-PA the chains are cross-linked at the carboxyl groups through the metal ions.

EXPERIMENTAL

IR spectra were recorded on a Perkin-Elmer spectrometer at 1500-4000 cm^{-1}. The rate of passage of nitrogen saturated with D_2O vapor was calculated so that the concentration of D_2O was not the limiting factor. At a rate of 0.7 liter/min a tenfold exchange per minute of the mixture of gases in the cell was achieved, which ensured that the rate of deuterium exchange was independent of the rate of passage of the D_2O vapor.

The investigation of the deuteration of a PS film is made up of the following operations:

1. Where necessary, the film is dehydrated in the cell by means of a stream of hot dry nitrogen, after which the furnace is switched off and nitrogen is passed at room temperature.

2. The spectrum of the initial state with $q = 0$ is recorded, and from this $D(0)$, $D'(0)$, and $D_0(0)$ are determined.

3. A current of nitrogen saturated with D_2O vapor is passed through the cells, and the change in $D(q)$ with time is recorded simultaneously.

4. After the limiting value of $D(Q)$ has been reached the final spectrum is recorded, and from this $D(Q)$, $D'(Q)$, and $D_0(0)$ are determined.

In the investigation of the dynamics of deuterium exchange, the time τ is reckoned from the moment of the simultaneous opening of cock (3) and shutting of cock (1) (Fig. 1). Cocks (2, 6, and 7) are opened beforehand. Then the D_2O vapor does not come directly into contact with the sample but only after the displacement of the dry nitrogen from the volume of the connecting tubes and of the cell. The true initial reaction time τ is therefore determined by extrapolating the curve $q = f(\tau)$ to the value $q = 0$. τ_0 corresponds to the intercept cut off by the curve on the axis of abscissas. $\tau = \tau' - \tau_0$, where τ' is the time reckoned from the moment of shutting cock (1).

SUMMARY

1. An apparatus has been constructed for the deuteration of solid samples (films) in a stream of D_2O vapor.

2. An IR-spectroscopic method of determining the degree of deuteration of the hydroxy groups of films of pectin substances by measuring the optical density of the maximum of the $\nu(OH)$ band has been developed.

3. No structurally distinct zones have been detected in films of derivatives of pectin substances by the method developed.

4. The rate of deuterium exchange of the water of hydration of pectin is greater than the rate of exchange of the hydroxyls of the pyranose rings.

5. Differences in the rates of deuterium exchange for different derivatives of pectin substances of the carboxy group are most probably due to differences in the density of packing of the polymer chains.

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