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The Applications of Chromadistillation in the Injection of Large Samples in the Gas-chromatographic Determination of Impurities

S.M. Yanovskii

The characteristic features of the injection of large samples into the chromatographic column with allowance for chromadistillation are considered. The theory of the chromadistillation processes occurring in the initial section of the column, which permits a rational choice of the injection parameters, is discussed and the methods and devices for the direct injection of the sample into the column are surveyed.
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I. INTRODUCTION

The application of chromatography for the preparative isolation of pure substances, high-temperature analysis, concentration of impurities, and various physicochemical measurements has required a continuous development of methods for the injection of large amounts of the substance to be analysed into the column. It became possible to meet these requirements with effective solution of these problems using a new version of chromatographic separation—chromadistillation (CD), developed in the USSR in recent years.[1-6] In the course of CD in a column on an inert solid filler and in a stream of carrier gas, numerous processes involving the condensation and evaporation of the components in the mixture analysed take place. Complete mixture separation is achieved either as a result of the application of a temperature field with a negative temperature gradient (thermal CD)[1,7] or as a result of preliminary injection into the column of a specified dose of a substance more volatile than any of the mixture components (limiting CD).[2,8,9] Isothermal elution (IE) of a mixture without the introduction of a limiting agent can also be included among CD processes.[10-14] The so-called "chromatography of nearly saturated vapours" (CNSV), which the present author has proposed,[15,16] occupies an intermediate position between traditional gas chromatography and CD. As in the usual chromatography, the CNSV process takes place on an adsorbent or a stationary phase (SP). However, owing to the large amount of liquid on the sorbent, the concentrations of the substances analysed in the gas phase approach the values corresponding to saturation. The limiting[9] and thermal[17] versions of CNSV have also been realised.

The development of the theory of CD methods[18,19] and the explanation of the mechanisms of the processes occurring[20-22] demonstrated their possible application for the analysis of mixtures,[23,24] continuous preparative synthesis of pure substances,[25-27] fractional distillation of mixtures of petroleum products in terms of their boiling points,[28-37] and the determination of the physicochemical characteristics of pure substances and solutions.[6,20,21,29,38-40] The characteristics of the CD process can be used to simplify the detection system[20-22,36,37,39] and also to concentrate and determine the "light" and "heavy" impurities in liquids;[33,45-50] a method has been described for the use of a CD pre-column with injection of large samples into a capillary column (CC) in the determination of impurities.[51]

A large series of studies by Grob and co-workers, who introduced the so-called "solvent effect" (SE)[52] manifested in the direct (without splitting of the stream) [splitless] injection of the sample into the column,[53-55] can be regarded as a separate special application of CD to the determination of impurities on CC. From the standpoint of classical chromatographic ideas,[56,57] a 2 μl sample exceeds the maximum permissible size for packed columns by a factor of at least 10[54] and according to estimates[56,58] the dose introduced into a CC should not be greater than 2×10^{-4} μl. However, the sample size in studies on the SE reaches 10 μl for CC and in studies on CD using filled columns even several millilitres can be employed without impairing the efficiency of the separation. The sharp increase in the dose leads to new qualitative results. The presence of liquid components on the column (including the solvent as the main mixture component) creates conditions for the formation of narrow fronts at the boundary between the zones of the components being separated and hence for the concentration of the impurity components at these boundaries. The solvent must be regarded as the most important factor determining the efficiency of the analysis; its presence is essential for injection and it is therefore unnecessary to take steps for its removal from the system, which is usually done in the traditional injection methods.[59,60,79]

The direct injection of a large sample into the chromatographic column and also the employment of the entire initial sample (without any being discarded) in a single analytical cycle creates a new direction in the field of injection, since it makes it possible to improve the quantitative characteristics of the analysis. The amount of the component to be determined injected into the column with the sample, is then increased and the errors associated with the diversion of the stream and the fractionation of the components on evaporation are eliminated, and the probability of the thermal decomposition in the analysis of unstable compounds is reduced, since the injection is carried out at lower temperatures. With increase of the dose, the sensitivity of the

determination of the impurity component increases. In view of the great possibilities of the direct methods for the injection of the sample into CC, there has been a sharp increase in recent years of the number of publications on this field (in 1982 there were more than 100)[61] and special symposia and schools designed to learn the new injection technique have been organised.[62,63] In connection with the development of the injection methods, studies have been carried out by major investigators such as Zhukhovitskii, Grob, Schomburg and others.

The choice of the the injection system (with or without stream-splitting) depends on the specific analytical problem.[62] The direct injection methods using the SE constituted a significant step forward in the field of injection. However, the rational development of these methods was appreciably held back by the fact that the SE theory was erroneous (see below), because the processes occurring at the initial section of the column were regarded as purely chromatographic. In fact they are to a large extent of the distillation type and are covered by the new version of chromatography—chromadistillation. From our point of view, the studies on the SE can be regarded as an important application of CD for the determination of impurities in solvents. In these methods the high efficiency of the CC is combined with the possibility of greatly increasing measurement sensitivity. The fact that the development of the CD and SE processes has occurred independently in different chromatographic schools can be accounted for by the vast flood of available information which created conditions such that investigations could occur independently even in closely related fields.

The aim of the present review is to examine the entire range of problems arising in the injection of large samples into the chromatographic column. It became possible to explain all the results obtained by the SE method from the standpoint of the CD theory. Such a treatment permits a deeper understanding of the mechanisms of the processes occurring during the direct injection and indicates further ways of improving the injection technique.

II. PRINCIPAL CONCEPTS AND TERMS

For subsequent exposition it is essential to establish a definite correspondence between the terms used in CD[5,6] and in the SE methods.[64] The processes occurring in the injection of large samples can be most rationally classified in accordance with the character of the sorption isotherms of the substances being separated. In the injection of large amounts on the SP, concave sorption isotherms are always observed (Fig.1) and are described by the equation[65]

$$a = \frac{mp/p^0}{1 - p/p^0}, \qquad (1)$$

where a is the degree of sorption, m the amount of the SP, p the partial pressure of the component, and p^0 the vapour pressure. At low relative pressures p/p^0 Eqn.(1) becomes identical with the linear equation for the classical gas–liquid chromatography (GLC). With increase of p/p^0, the non-linearity of the sorption isotherms is manifested to an increasing extent and, when $p/p^0 \to 1$, a tends to infinity. Sorption isotherms of an analogous character are observed also in adsorption in capillary condensation.[66] The concave section of the sorption isotherms corresponds to the conditions in the chromatography of vapours near saturation.[15] In the absence of the SP ($m = 0$) and when pure substances are injected onto an inert layer, the sorption isotherms have the form of vertical straight lines passing through the $p = p_i^0$ points (Fig.1). The principal version of CD corresponds to this case.[1,6,7]

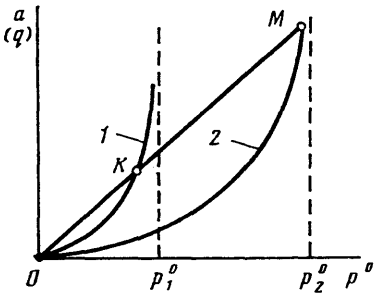


Figure 1. The sorption isotherms for components *1* and *2* at high concentrations;[9] continuous line—CNSV; dashed lines—CD; the straight line *OM* corresponds to the material balance of the components at the boundaries between the zones; q is the amount of liquid per unit volume of the bed in CD.

CD processes occur in CC in two versions of the sample injection without splitting the stream (splitless injection[67]): injection with preliminary evaporation of the sample (via an evaporating injector) and with its subsequent condensation on a cooled column[53,54] or direct injection (without preliminary vaporisation) into the cooled column (direct cold on-column injection).[67] CD processes are sometimes observed even in the injection of large samples with small stream splitting coefficients (split injection).[68] The process occurring on the surface of a clean CC wetted by the liquid (flooded inlet)[69] corresponds to ideal CD.[6,7] Analysis on a SP saturated by the solvent (phase soaked)[70,71] should be included in CNSV according to the present author's ideas.[15] The isothermal process in the distribution of the sample in the form of a film (sample film)[64] corresponds to isothermal CD elution.[10–14,20,21] The preliminary deposition of the pure solvent on the layer (solvent film)[64,72,73] corresponds to the limiting versions of chromatography and CD.[8,9]

Depending on the character of the solutions of the impurity components formed with the main mixture component (solvent), different elution sequences of the impurities are observed on the chromadistogram (CDG). If the solution is ideal, the components emerge in accordance with the increase of their boiling points (t_b): less volatile impurities (heavy) emerge at the closing solvent front and more volatile impurities (light) emerge at the leading front.[49] In solutions with marked positive deviations from Raoult's law (with azeotropy or layer formation in the system) the elution sequence may not correspond to the t_b of the components.[74] The components with t_b greater than that of the solvent, which emerge at the leading front of the main component, should then be classified as light impurities. In the isothermal elution of the mixture there is a possibility of either the emergence of a less volatile impurity component after the solvent (solute lagging)[21,75] or the loss of light components simultaneously with the disappearance of the solvent zone (solute escape).[6,76] Retaining the term "solvent effect" as being more general, Grob and co-workers[77,78] introduced the concept of "solvent trapping" for all processes in which

a large sample can influence the results of sample injection, by analogy with low-temperature extraction (cold trapping). It is therefore suggested that impurities be divided into those fully retained by the solvent (trapped) and those which are only partly retained or not retained at all, which corresponds to our classification of impurities into heavy and light.[49]

Several approaches to the investigation of the impurity zone formation processes on injection of a large sample onto the column have now arisen. In studies on CD [6] the theory of processes occurring on a column with an inert filler for concentrated solutions of the components of the initial mixture has been mainly considered (on the basis of balance equations). In experiments designed to test the theory, the inlet to the column was connected directly to the detector (Fig. 2*a*), which made it possible to record the changes in the concentrations of the components in the zones formed. Additional information about the mechanism of the distribution of the liquid was provided by visual studies on the wetted section of the column. The determination of the impurities can be considered, within the framework of CD, as the limiting case where the concentrations of the components to be determined are small compared with that of the principal component. For the separate determination of the impurity components, a portion of their sample was directed to the chromatographic column after the preliminary enrichment on the CD column (Fig. 2*b*); the main bulk of the solvent was then discarded to the atmosphere.

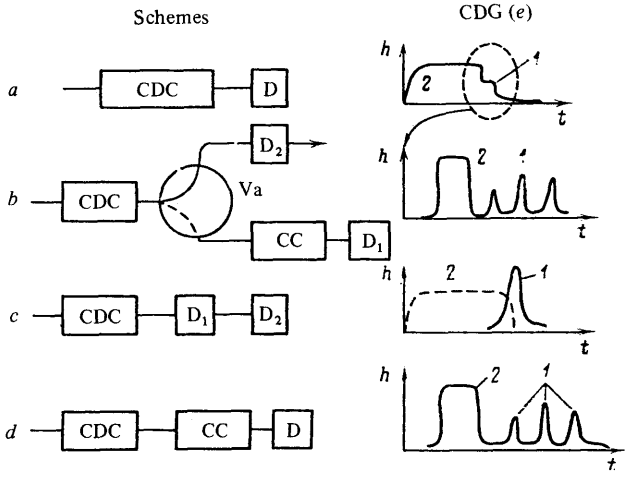


Figure 2. Schematic illustration of the experimental apparatus for the investigation of CD processes (*a*–*d*) and the corresponding elution curves (*e*); *h* = height of the signal on the recorder strip; *t* = time; CDC = chromadistillation column; CC = capillary column; Va = valve; D = detector; 1) impurity; 2) solvent.

The isolation of an impurity from the solvent has been studied both theoretically and experimentally.[72] In order to determine the location of the impurity on the elution curve, the author used an additional selective detector (Fig. 2*c*).

The approach which arose in the development of the SE is strictly experimental and involves the determination of impurities solely on CC. The samples were deposited directly in the CC and the processes occurring along its initial section could be inferred solely from the results of the separation of the components in the main section of the column containing the SP.[79] In later investigations[52,70] special attention was paid to the section for the sample distributed (in Fig. 2*d* we have specified it as the CD column) and the processes occurring along this section were studied by both visual and chromatographic (sampling) methods. The principal data were obtained as a result of detailed and laboratory experiments and the treatment was confined to the framework of development chromatography.

In the subsequent exposition we shall consider initially the theory of the process and then its experimental test and possible applications.

III. ISOTHERMAL ELUTION OF THE MIXTURE

In isothermal elution the mixture analysed is deposited in the liquid form on an inert carrier (without SP) and is eluted from the column by a stream of carrier gas at a constant temperature. Isothermal elution (IE) is the initial stage in any of the versions of the CD separation and it is therfore essential to consider its mechanism in detail.

1. Theory of Isothermal Elution

Our studies in the field of CD were preceded by Kiva's investigations[10–14] on the azeotropy of solutions in which the IE of the mixture from the surface of an inert carrier was first carried out (Fig. 3*a*). The elution curve has in this instance a stepped form (Fig. 3*b*). Under IE conditions, only partial separation of the mixture takes place. The less volatile component does not emerge in a pure form until the final zone is reached. However, the result of such separation is by no means trivial. According to the theory of customary distillation,[80] when a solution evaporates from a flask, the composition of the effluent mixture must vary continuously, becoming gradually enriched in the less volatile component *1*, which is evolved in a pure form only with the last drop of the solution.

The observed picture of separation under IE conditions is determined by the occurrence of distillation processes under chromatographic conditions where a multiplicity of mixture condensation and evaporation steps are attained as the component bands move along the bed. Despite the fact that in the given case there is no sorbent familiar to workers in the field of chromatography, the processes occurring can be considered by approaching them from the standpoint of frontal-desorption chromatography,[56] i.e. in the given instance the mixture analysed is, as it were, resolved "spontaneously" as it moved along the bed. If it is supposed that the mixture components *1* and *2* have markedly different vapour pressures and that component *1* is almost involatile, then the rear edge of the zone of component *2* will migrate forwards along the column in accordance with chromatographic laws (Fig. 3*c*). On the other hand, if the less volatile component *1* has an appreciable vapour pressure, then its rear boundary will also move along the column (Fig. 3*d*). The rate of migration of the zone boundaries u_i as well as the composition of the solutions formed in the zones are in general determined by the material balance equations at the zone boundaries.[6,21] This approach

is equivalent to considering the theory of the equilibrium process on the assumption that the height equivalent to the theoretical plate (HETP) is zero. In the IE of a binary mixture, the material balance equations with respect to both components at the boundary where component *2* ends yield

$$u_2 = \frac{\alpha c_2^0}{q^{II}} = \frac{\alpha c_1^0 (1 - N_1)}{q^I - q^{II} N_1}, \qquad (2)$$

where α is the linear flow rate, c_i^0 the concentrations of the saturated vapour of components *1* and *2*, q^I and q^{II} the amounts of liquid per unit volume of the column for bands I and II, and N_1 the mole fraction of component *1* in the initial mixture. Here and henceforth we shall neglect the amount of substance in the gas phase compared with the amount in the liquid phase. Having expressed q^I in terms of q^{II} with the aid of Eqn.(2), we obtain

$$q^I = q^{II}[\beta_{1,2} + (1 - \beta_{1,2}) N_1]. \qquad (3)$$

The equation for the rate of migration of the rear boundary of the zone of component *1* then assumes the following form:

$$u_1 = \frac{\alpha c_1^0}{q^I} = \frac{\alpha c_1^0}{q^{II}[\beta_{1,2} + (1 - \beta_{1,2}) N_1]} \qquad (4)$$

where $\beta_{1,2} = c_1^0/c_2^0$. All the principal characteristics of the IE process follow from Eqns.(2)–(4). The existence of a unique solution to the system of material balance equations shows that the zone corresponding to the mixture should be followed in the bed by the appearance of the zone of the pure less volatile component *1*. Within the limits of this zone, component *1* is present on the carrier in the liquid state and its concentration in the gas phase corresponds to the saturated vapour (c_1^0). According to Eqn.(3), the amount of liquid per unit volume of the column in the zone of component *1* is less than in the initial zone ($q^I < q^{II}$), since the multiplier in square brackets in Eqn.(3) is less than unity. It must be emphasised that, under the conditions of the rapid attainment of equilibrium between the vapour and the liquid, for which the equations presented are significant, the zone of component *1* is formed immediately after the introduction of the mixture. Indeed, comparison of Eqns.(2) and (4) shows that $u_1 < u_2$. For this reason, however short the time interval Δt from the start of the experiment (provided that there is no spreading and on the assumption that the HETP is zero), a certain distance Δx arises between the boundaries and increases as the process proceeds. This explains Fig.3*e*, which presents parts of the migration of the boundaries of the zones of components *1* and *2*.

The formation of the zone of the pure component *1* with a definite amount of liquid within the limits of the zone q^I [see Eqn.(3)] and at the concentration in the gas phase corresponding to the saturated vapour (c_1^0) occurs as a result of repeated mixture condensation and evaporation steps. One cannot assume that the more volatile component *2* is eluted from the section where it was initially located, as usually happens in chromatography. The less volatile component *1* evaporates simultaneously. Its concentration in the vapour phase of the zone formed is higher than that in the initial zone. Component *1* condenses (dissolves) at the boundary between the zones and forms a corresponding zone I (Fig.3*d*) with a definite thickness of the liquid film. The components are separated by distillation at the boundary between the zones in such a way that the entry of one of the components into a neighbouring zone (as a result of diffusional spreading) leads to its removal as a result of the difference between the vapour pressures of the substances being separated. The opposed effects of these processes give rise to a stationary distribution of the substance between the zones whose boundary migrates in accordance with the laws governing CD equilibria.[18] By virtue of the distribution of liquid in the form of a thin layer on the packing, conditions are created which ensure the effective saturation of the gas stream by the vapours of the substances analysed. As a result, the spreading of the stationary fronts is insignificant.[19] According to the material balance equation (4), the zone of the pure component *1* should arise at the rear edge of the band. For this, it is sufficient that the two fronts of components *1* and *2* (Fig.3*d*) should traverse a distance equal to the width μ_2 of the diffuse boundary (between the zones of components *1* and *2*). In the absence of equilibrium, under conditions where the HETP is not zero, the relaxation time required for the appearance of the zone of component *1* is

$$t_{rel} = \frac{\mu_2}{u_2 - u_1}. \qquad (5)$$

In non-equilibrium fractionation,[80] the attainment of a stationary distribution with formation of the zone of the pure component *1* requires that the mass-exchange and countercurrent hydrodynamic processes should go to completion throughout the volume of the sample. As in chromatography, numerous separation steps are achieved more readily in CD than in distillation.

The theory of the IE of the impurity component follows directly from the general theory of equilibrium isothermal CD[18] as a special case where the fraction of one of the components in solution tends to zero. In this case the usual CDG (Fig.3*b*) is converted into the CDG of the solvent (Fig.3*f*), at the closing front of which only a small hump due to the impurity component is observed. If the main component *2* is not registered by the detector, the observed zone of the impurity *1* should have an extended low, stepped section terminating in a narrow peak (Fig.3*g*).

We shall consider special cases of the application of Eqn.(3).

(1) When $N_1 \to 0$, the impurity is the less volatile component. The amount of liquid in the neighbouring zones is then linked by the equality $q^I = \beta_{1,2} q^{II}$. As mentioned above, in the general case $q^I < q^{II}$, since $\beta_{1,2} < 1$. This result confirms the conclusion that the zone of component *1* arises immediately (under equilibrium conditions) after the start of the IE process and that the amount of liquid within the limits of this zone should be rigorously defined and constant as a function of time (for a uniform initial distribution of the liquid).

(2) When $N_1 \to 1$, component *1* is the main one in the mixture and the impurity is more volatile than the solvent. The amount of liquid in the zones then remains virtually unchanged, i.e. $q^I = q^{II}$.

(3) When $\beta_{1,2} \cong 1$, the vapour pressures of the components are similar and the equality $q^I = q^{II}$ holds regardless of mixture composition.

(4) When $\beta_{1,2} \to 0$, the less volatile component has an extremely low vapour pressure compared with the solvent. The amount of liquid in the zone of component *1* formed then depends on its mole fraction N_1 in the initial mixture.

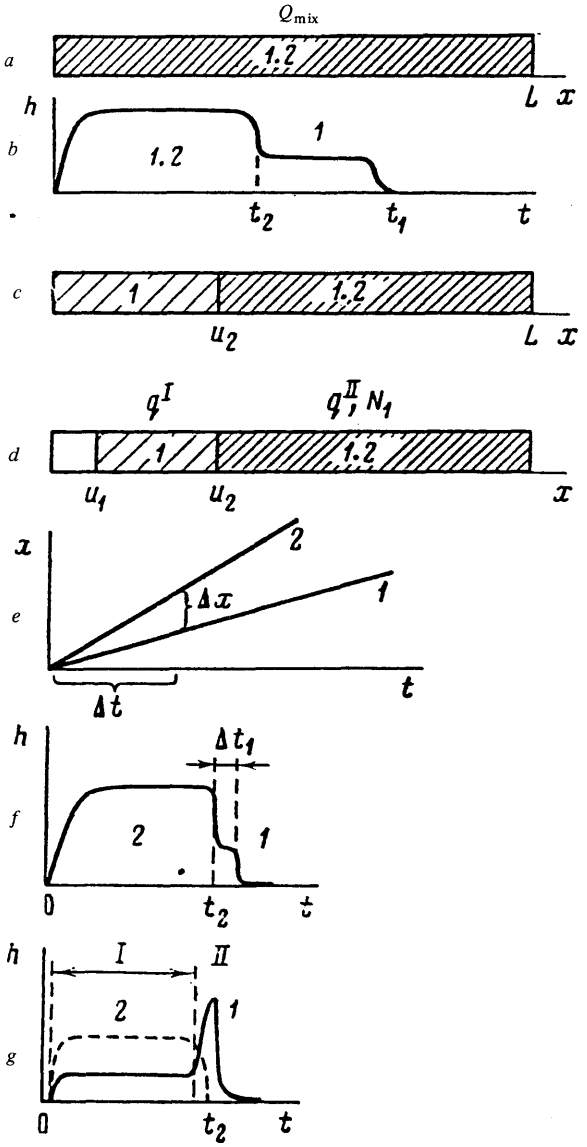


Figure 3. Schematic illustration of isothermal elution;[6,21] x = distance along the bed; L = length of wetted section of the bed.

We shall now consider the factors which influence the impurity zone width. The width of the rear zone on the CDG (Fig.3*b*) corresponds to the less volatile component *1*. Its length ($\Delta t_1 = t_1 - t_2$) depends on the total amount of the deposited mixture (Q_{mix}), which wets a section of the column having a length L. Each boundary of the zone of component *1* crosses this section at its own rate determined by Eqns.(2) and (4). As a result, the equation for the component *1* band width on the CDG is

$$\Delta t_1 = \frac{L}{u_1} - \frac{L}{u_2} = t_2(\beta_{2,1} - 1)N_1, \qquad (6)$$

where $t_2 = Q_{mix}/v_0 c_2^0$, $\beta_{2,1} = c_2^0/c_1^0$, $Q_{mix} = q^{II}SL$, $v_0 = \alpha S$ is the volume flow rate, and S the cross-sectional area of the column. It follows from Eqn.(6) that, with decrease of the fraction of component *1* in solution ($N_1 \to 0$) and as the vapour pressure of the impurity component approaches that of the solvent ($\beta_{2,1} \cong 1$), the width of the impurity zone Δt_1 diminishes. However, it must be emphasised that, under equilibrium elution conditions, component *1* is present in the liquid form within the limits of its zone, however small the latter may be, and its concentration in the gas phase corresponds to the saturated vapour (c_1^0). The conditions governing the formation of the pure component *1* band are defined by the relations

$$\beta_{1,2} \neq 1 \quad \text{or} \quad c_1^0 < c_2^0. \qquad (7)$$

Indeed, when $\beta_{1,2} = 1$, it follows from Eqns.(2)–(6) that $q^I = q^{II}$, $u_1 = u_2$, and $\Delta t_1 = 0$. This means that the rates of migration of the boundaries are the same and that the bands of components *1* and of the mixture of components *1* and *2* are indistinguishable in terms of the amount of liquid. When $\beta_{1,2} < 1$ and $N_1 \to 0$, the step corresponding to component *1* becomes narrow and, on spreading, it is converted into a peak on the rear solvent edge (Fig.3*g*).

Losses of the less volatile component *1* are possible in IE simultaneously with the emergence of the *1,2* solvent band (Fig.3*g*, Section 1). Knowing the rate of migration of the rear boundary of the solvent zone along the bed [Eqn.(2) for u_2], it is possible to obtain an expression for the retention volume at this boundary:

$$V_2 = t_2 v_0 = \frac{L}{u_2} v_0 = \frac{Lq^{II}}{\alpha c_2^0} v_0 = \frac{Q_{mix}}{c_2^0}. \qquad (8)$$

Eqn.(8) shows that the retention volume V_2 is independent of the way the liquid is distributed along the bed (q^{II} does not enter into the final expression) and is determined by the total amount of the mixture Q_{mix} introduced into the column and by the vapour pressure of the most volatile component *2*. Furthermore, regardless of the composition of the initial mixture, V_2 remains constant and equal to the retention volume of the pure solvent sample with a dose equal to Q_{mix}. This finding is associated with the fact that, when the solution is diluted, the amount and vapour pressure of the solvent in the mixture diminish to an equal extent, in proportion to its mole fraction N_2 in the solution.

In order to calculate the losses of the impurity component *1*, it is necessary to take into account the concentration at which it is evolved from the column during the elution period of the volume V_2:

$$Q_{loss,1} = V_2 c_1^0 N_1 = Q_{mix} N_1 \beta_{1,2} = Q_{init,1}\beta_{1,2}, \qquad (9)$$

where $Q_{init,1}$ is the initial content of component *1* in the initial mixture. It follows from Eqn.(9) that the fraction of component *1* lost ($Q_{loss,1}/Q_{init,1}$) is independent of mixture composition and is determined by the ratio of the concentrations of the saturated vapours of the components and the solvent ($\beta_{1,2} = c_1^0/c_2^0$). An analogous formula has been obtained in other studies.[72,76]

We shall consider an example illustrating the employment of the formulae derived. Suppose that 2 μl of hexane containing 20 p.p.m. of dodecane ($N_1 = 2 \times 10^{-5}$) has been injected into the column under normal conditions. We shall now estimate the width of the impurity zone and the losses. The amount of solvent corresponding to the dose indicated

is 2×10^{-5} mole. For the hexane vapour pressure $p_6^0 \cong 120$ mmHg, the concentration of the saturated hexane vapour in the gas phase is 0.7×10^{-2} mol litre^{-1}. If the flow rate is $v_0 = 2$ ml min^{-1}, then the evaporation time of the main component t_2 is ~10^2 s. With increase of the number of carbon atoms in the series of alkanes by unity, the relative volatility changes by a factor of ~3 and we therefore have for hexane and dodecane $\beta_{2,1} \cong 3^6 \cong 10^3$. Having substituted the numerical data in Eqns.(6) and (9), we find that $\Delta t_1 = 2$ s (i.e. the impurity zone is quite long) and the losses are ~0.1%.

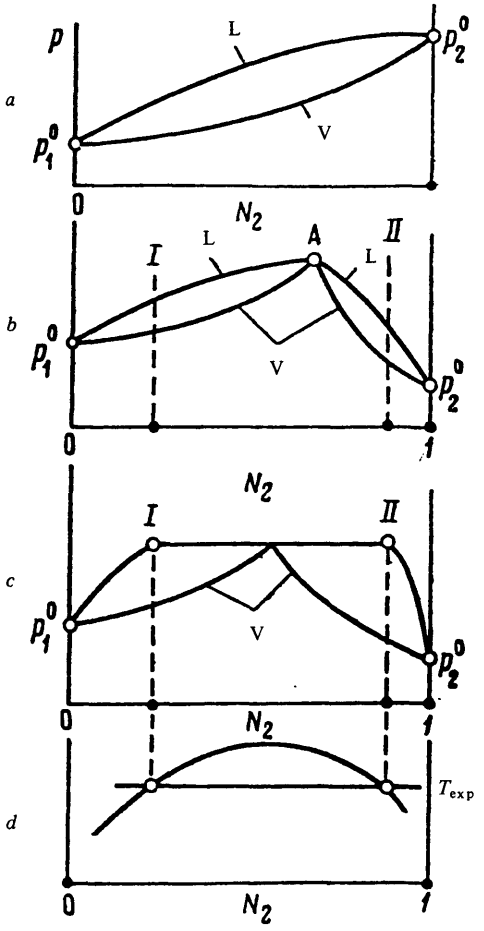


Figure 4. Schematic representation of the equilibrium diagrams for various systems:[80] L = liquid line; V = vapour line; *A* = point corresponding to the azeotrope: *a*) ideal solution; *b*) azeotropic mixture; *c*) heterogeneous liquid mixture (two layers); *d*) solubility curve.

We shall consider the case where the main system component (component *1*) is less volatile than the impurity (component *2*). The form of the elution curve of the impurity component and its location relative to the main mixture component will depend on the type of vapour–liquid equilibrium diagram (Fig.4). If the solution is ideal (Fig.4*a*), then, as shown in a number of studies,[10–14,74] the more volatile component *2* is evolved first and is then followed by the less volatile solvent *1* in a pure form ($p_2^0 > p_1^0$), in conformity with distillation laws. The occurrence of azeotropy or layer formation in the system (Figs.4*b*–4*d*) can alter the elution sequence of the zones on the CDG. In this case the azeotropic mixture or the solution separating into layers behaves (as in the usual distillation) as a single most volatile component. Depending on the mixture composition (I or II in Fig.4*b*) in relation to the azeotrope (points *A* in Fig.4*b*), IE can lead to the formation of the zone of one of the mixture components at the rear edge.[14,74] Thus, even if the impurity has a lower vapour pressure than the solvent ($p_2^0 < p_1^0$), there is a possibility of the evolution of the impurity component at the leading solvent edge for azeotropic and heterogeneous liquid systems.

In conformity with the above features of the IE under equilibrium conditions, the band of the volatile impurity should have a rectangular shape on the CDG. It remains to discover the amplitude and duration of this signal on the elution curve. In contrast to the heavy impurity, the concentration of the light impurity in the IE of the mixture is not higher than the concentration of the initial mixture. The concentration of component *2* in the gas phase over the solution is defined in accordance with the law governing the equilibria in real solutions:

$$c_2 = c_2^0 \gamma_2 N_2, \qquad (10)$$

where γ_2 is the activity coefficient of component *2*.

It must be emphasised that the CD analytical methods based on IE bear some similarity to the widely used vapour phase analysis,[81,82] since in both methods the components are not enriched in the gas phase. In contrast to the vapour phase analysis, in the CD methods for the direct determination of impurities[45,49] the sample injection is carried out more rationally: all the impurity is extracted from the sample and there is no need to know exactly the equilibrium parameters and hence to maintain rigorously the experimental parameters.

According to Eqns.(8) and (10), for an ideal solution the duration of the rectangular pulse of the impurity component remains constant with decrease of the concentration N_2 ($V_2 = \text{const.}$), while the amplitude diminishes. The activity coefficient of the impurity in a real solution can influence significantly the retention volume in the impurity band:[38,49]

$$V_2 = \frac{Q_{cm}}{c_2^0 \gamma_2}. \qquad (11)$$

As a rule, dilution of solutions with pronounced positive deviations from Raoult's law (Figs.4*b* and 4*c*) entails a sharp increase of the activity coefficient γ_2 (γ_2 may amount to tens and hundreds of units[83,84]), which may lead to an appreciable decrease of the width of the impurity component zone.

We shall quote an example of the determination of the butanol impurity in water. Suppose that the sample size is 0.1 ml (5×10^{-3} mole). At room temperature (25 °C), the vapour pressure of butanol is 7 mmHg, which corresponds to the gas-phase concentrations $c_2^0 = 4 \times 10^{-3}$ mol litre^{-1}. According to Eqn.(11), the component forming an ideal solution ($\gamma_2 = 1$) would be evolved under these conditions in a volume $V_2 = 10^3$ ml or after 50 min for a volume flow rate $v_0 = 20$ ml min^{-1}. Since $\gamma_2 \cong 45$ for butanol,[84] the time required to evolve the impurity is ~1 min.

We shall consider the studies on the SE mechanism in the light of the IE theory described above. In a series of communications by Pretorius and co-workers,[64,75,76,86] an attempt was made to derive an equation for the rates of migration of the solvent and sorbate boundaries and also

to determine the conditions governing the appearance of various SE regimes. A general disadvantage of the approach employed by Pretorius and co-workers,[64,75,79,85] which led to a series of incorrect conclusions, is their attempt to regard the processes in which the sorbate moves in the solvent as purely chromatographic. For example, it is impossible to set up the material balance equation at the closing solvent front immediately with respect to both components [see Eqn.(1) in Ref.85]. According to the IE theory which we described above, two cases are possible. If the impurity is lighter than the solvent, it moves ahead along the column and should not be taken into account in the material balance. For impurities heavier than the solvent, it is essential to formulate the material balance equations for both boundaries formed (Fig.3*d*). For this reason, Eqn.(6) quoted by Pretorius et al.[85] for the rate of migration at the closing edge of the solvent film can be valid only if one neglects the sorbate concentration in the gas phase.

The stages involving the formation of the sorbate film (initial and final) have been arbitrarily differentiated.[75] As mentioned above, a film of the less volatile impurity component is formed immediately (under equilibrium conditions) simultaneously with the onset of elution. Eqn.(12) obtained by Pretorius et al.[75] for the ratio of the thickness of the films in the solvent and impurity zones has narrow limits of applicability. The authors[75] assumed that the solvent, moving out of a wetted section, leaves on the surface an amount of impurity corresponding to its fraction in the initial solution. As shown above, this is possible only for $\beta_{1,2} \cong 0$ [see the special case 4 of Eqn.(3) described above]. As a result an inequality [inequality (14)[75]] was obtained, which imposes unduly rigorous requirements on the conditions for the appearance of the impurity zone:

$$\frac{p_1^0}{p_2^0} < \frac{n_1}{n_2}, \qquad (12)$$

where p_1^0 and p_2^0 are the vapour pressures of the impurity and the solvent and n_1 and n_2 are the molar concentrations of the impurity and the solvent in the initial mixture. It follows from condition (12) that the probability of the appearance of an impurity film is extremely low and it can be formed only for hydrocarbons with a molecular weight in excess of 500.[75] The CD theory presented above and the numerical example show that the impurity zone can be formed only when condition (7) is fulfilled and its formation is entirely realistic for a dodecane solution in hexane at a concentration of 2×10^{-2} g litre^{-1}.

In order to compress the peaks, it has been suggested[86] that a section of CC be established in which the so-called "retention gap" is present. For this purpose, the SP is partly or fully washed out of the initial section I of the column (Fig.5). The possible mechanism of the operation of such a section has been examined: the sorbate band *1* is hardly retained and rapidly traverses section I after the solvent *2* and then, when the substance passes from the SP to the CC (Section II, Fig.5b), it is compressed. If programmed temperatures are employed, then the heavy impurity mixture components begin to be eluted from the initial section I of the column only after the attainment of the appropriate temperature.

In another communication[87] schemes designed to explain the authors' idea about the mechanism of the "retention gap" are described. In their view, the sorbent is concentrated because in section I one edge of the sorbate band (the rear edge) moves at a higher rate (u_1', Fig.5*c*), while in the next section II, containing the SP, the other edge of the sorbate band (the leading edge) migrates at a lower rate (u_1''). However, this explanation of the concentration process is clearly unsatisfactory. It follows from the analysis of the operation of compound columns[56,88] that the degree of narrowing of the band is predetermined by the ratio of the rates of migration of the sorbate in the zones indicated. It must be emphasised that the concentration of the test substances in the gas phase does not change under these conditions. The sorbate band may be compressed without a change in the sorbate concentration in the gas phase on passing through the boundary at which the sorption capacity (the Henry coefficient Γ_i, Fig.5*d*) changes. Suppose that the initial width of the sorbate band is μ_1 for a uniform distribution of the substance with the concentration c_0 in the gas phase. The rate of migration of the band along section I is $u_1' = \alpha/\Gamma_1$. The entire band passes through the boundary during the period $\tau = \mu_1/u_1' = \mu_1\Gamma_1/\alpha$. During the same period, the leading edge of the band for the same concentration in the gas phase c_0 traverses a distance $\mu_2 = \tau\alpha/\Gamma_2 = \mu_1\Gamma_1/\Gamma_2$ in section II at a rate $u_1'' = \alpha\Gamma_2$. If the degree of sorption in the second section is greater ($\Gamma_2 > \Gamma_1$) the band is compressed by a factor Γ_2/Γ_1, although the concentration of the test substance in both parts of the column remains unchanged (c_0).

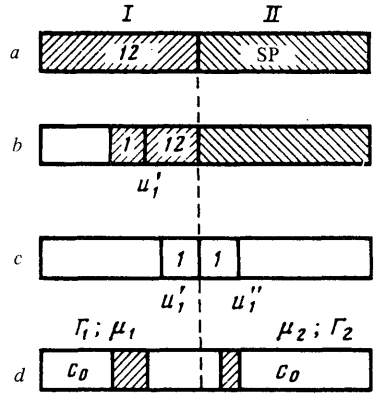


Figure 5. Schematic explanation of the "retention gap" mechanism;[56,86] u = rate of migration of boundary; μ = width of the sorbate band; Γ = Henry coefficient; c_0 = concentration of sorbate in the gas phase.

From the standpoint of the CD theory, the results described by Grob[86,87] can be readily accounted for in the following manner. The removal of the SP brings the experimental conditions closer to those of the isothermal CD on an inert carrier[6,21] (Fig.3). In the IE of the mixture on section I (Fig.3*b*), the solvent (*1*, *2*) is followed by the impurity component *1* in the liquid form. As a result of this, its concentration in the gas phase becomes a maximum and corresponds to the vapour pressure at the given temperature. Consequently, long before the rear solvent edge approaches section II with the SP (Fig.5), an increased sorbate concentration will have already appeared on section I. This increase of the concentration is in fact responsible for the narrowing of the band on the chromatogram. Furthermore, when the results of these experiments are considered, it is also essential to take into account the transition to zone

II of part of the impurity component during the elution of the mixture on section I. The final formation of the narrow peak corresponding to the entire amount of the impurity component in the sample takes place as a result of the subsequent CNSV processes (Section VI).

The explanation of the mechanism of the compression of impurities reduces, according to Grob,[52,79] to the consideration of the elution of the sorbate on a phase containing the solvent with a variable amount of liquid along the bed (Fig.6). When the sample is injected via an evaporating injector, the solvent forms, when condensed, a mixture on the SP with a constantly increasing (in the direction of motion) film thickness (zone I, Fig.6). In the first stage of the separation process, the solvent *2* moves forward relative to the sorbate 1. Next the leading edge of the migrating sorbate band in section I encounters a greater amount of the liquid phase than the closing edge. As a result of the movement at different rates of the sorbate band boundaries, the band is compressed and peaks which are narrower than in the case of split injection are formed.

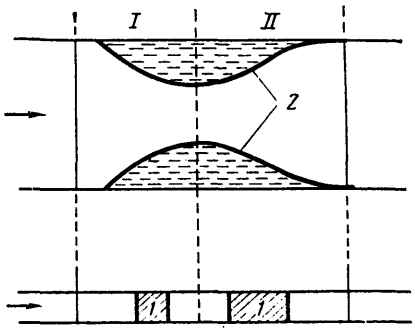


Figure 6. Schematic explanation of the SE mechanism in the gradient distribution of the solvent in the column:[52] 1) sorbate; 2) solvent; I) and II) direct and reverse SE zones.

The previously proposed picture of the concentration process (for section I in Fig.6) was subjected to a quantitative treatment[60,89] on the basis of the concepts adopted in GLC. It was shown that the retaining capacity of the liquid phase increases in parallel with the variation of its distribution profile. On this basis, it is concluded that the band of the impurity component on the elution curve is compressed (and the component is therefore concentrated), The correct explanation of the increase in the concentration of the impurity can be achieved on the basis of the CD theory.

The question of the concentration of the sorbate band for a longitudinal gradient of parameters (sorbability, capacity, and rate) has been frequently considered in the literature in connection with the development of versions of chromatography such as chromathermography,[56] where a moving temperature field with a negative gradient is used, and chromarheography,[90] in which a flow rate gradient is employed. It was shown in these studies that, in the absence of the flow rate gradient under the conditions of a stationary gradient of the Henry coefficient (achieved by altering the temperature or the amount of the phase), the concentration of the component cannot increase. The compression of the sorbate band does not lead to an increase in concentration, because the degree of adsorption of the component increases by a factor which is exactly the same as the factor by which the band has been compressed. Only the change in sorption capacity with time leads to an increase of concentration, as happens in chromatography with a programmed temperature[91] and in stationary chromathermography.[56] In both these versions thermal enrichment takes place, its extent being determined by the ratio of the Henry coefficients at the temperatures at which the sorbate is injected into the column and at the outlet from the latter. Furthermore, in chromathermography the "width enrichment", induced by the difference between the rates of migration at either boundary of the sorbate band, is manifested in the course of the migration of the temperature field. Grob[52,60] took no account of the capacity gradient migration factor. Only processes associated with the migration of the solvent front, the removal of the solvent by distillation, and the subsequent programming of the column temperature lead to the concentration of the impurity in the gas phase. We shall consider below the experimental data which confirmed the validity of the theoretical ideas presented above concerning the CD processes occurring in the course of IE as well as certain examples of the determination of impurities in solvents.

2. Distribution of the Liquid in the Column

Visual methods are important for the understanding of the mechanisms of the processes occurring in the injection of large samples. In a glass column filled with an inert carrier, which may consist of glass (etched) or metallic beads as well as diatomaceous carriers for chromatography, the section soaked with the liquid is readily seen.[21,22] A technique has been developed for the visual observation of the condensation process along the initial section of a CC.[52] The CC was first filled with a 1 *N* NaOH solution and was kept in boiling water for 4 h. As a result of leaching, the walls of the CC became appreciably clouded. It was established empirically that a thin film of the SP on the column (with a thickness $d_f = 0.12$ μm) is invisible—the column has a milky colour and is opaque. Large amounts of liquid ($d_f > 0.25$ μm) fill the pits in the column making it transparent and can be readily observed from the width of the transparent section of the column. The experiments were performed with the column thermostat at room temperature and the length and position of the ends of the soaked zone were measured after each 12 s.[52] Immediately after the injection of several microlitres of hexane through an evaporating injector, a relatively short soaked zone spread rapidly along the CC. After several seconds, the movement of the leading edge slowed down and the edge began to migrate in jerks. Within the limits of the soaked zone, waves arise and are carried forward in the form of a liquid plug which breaks up along the leading edge. After a time, the film at the closing edge became thinner and vanished. As both edges of the soaked zone move along the bed and the solvent evaporates, the leading edge slows down, whilst the rear edge accelerates until it overtakes the leading edge and the soaked zone vanishes completely.[69] The above picture of the distribution of liquid along the bed fully confirms our observations in experiments on columns filled with an inert carrier.[21] The distribution of the liquid on the surface of the filler could be assessed quantitatively from the rate of migration of the rear boundary of the soaked layer with the aid of

Eqns.(2)–(4) obtained previously. The amount of liquid q retained under isothermal conditions on the carrier surface in the presence of a gas stream was calculated. This quantity amounted to 0.05 ml of liquid in 1 ml of the column for various solvents and was almost independent of the flow rate and the grain size of the carrier. An appreciable increase of q at the bend in the U-shaped column is observed when the latter is positioned vertically. The constancy of q enabled us[21] to develop on the basis of the theoretical equations (2) and (4) a simplified method for the analysis of a binary mixture and for the calculation of the physicochemical parameters of liquids (the vapour pressures of the components and the activity coefficients). By comparing the velocity of the soaked front and that of hexane along the SP, it was possible to estimate approximately the thickness of the solvent film in the CC, which amounted to 40 μm according to Grob's data.[52] This quantity is close to that calculated on the basis of geometrical considerations and corresponds to a fraction of liquid in the column of 0.25.

The dependence of the length of the soaked section L and of its evaporation time t_2 on the experimental parameters has been studied.[21,69] It was found that L depends only slightly on the volatility of the solvent, the carrier gas flow rate, the inlet pressure, the flow rate and method of injection of the sample, and the grain size of the carrier in the column. According to Eqn.(8), the quantity t_2 is proportional to the dose and decreases in inverse proportion to the volatility of the solvent.

3. Distribution of the Sorbate along the Bed and Determination of Heavy Impurities

When the sample is injected directly into the column, the liquid and sorbate are distributed uniformly. This can be inferred from the stepped CDG obtained when the detector is connected to the outlet from the CD column (Figs.2*a* and 2*c*). The constancy of the signal during the period of the elution of the different zones indicates the constancy of the concentrations within the limits of the zone of the distributed liquid. The application of a selective detector with respect to the impurity[72] made it possible to demonstrate this yet again. In the above study the author actually repeated Kiva's principal results[14] in the determination of impurities and, on the other hand, to some extent foresaw certain CD and SE postulates. Experiments were performed on an inert carrier (Teflon) using two detectors—a flame-ionisation detector with a high sensitivity with respect to organic substances and virtually unresponsive towards water as well as a thermal conductivity detector for water. The impurity component (Carbitol) was readily soluble in water and was less volatile. The column was maintained at a temperature below the boiling point of water. The volume of the aqueous samples was 40 μl. When the mixture was injected, the following remarkable fact, which is valid for dilute solutions, was observed. Whereas pure Carbitol in the absence of water is hardly retained in the column, when its aqueous solution is injected part of the Carbitol (*1*) is eluted together with water (zone I in Fig.3*g*), while the main bulk of the Carbitol is evolved in the form of a narrow peak (zone II) at the rear edge of water (*2*). The size of the peak formed on the "tail" of the main component is proportional to the concentration of the impurity and sample size. Deans[72] (see also his later investigation[92]) recommends that the injection of large samples into one of the columns be combined with the ejection of the main component into the atmosphere and subsequent transfer of the impurity (by means of special switching devices) to another separating column, which facilitates the operation of the latter. An example of the determination of 0.04 p.p.m. of 1,2-dimethylcyclohexane in isooctane from a 0.4 ml sample was quoted.

In our determinations of heavy impurities in organic solvents by the CD method[44,93] the sample was injected several times in succession under isothermal conditions into the column. A step associated with the impurity component (octane), whose height corresponded to its vapour pressure, was observed on the CDG after the solvent (hexane) zone. The width of the step increased in proportion to the dose, whose maximum size was about 10 ml. These experiments showed that CD makes it possible to obtain high enrichments—the concentration of the impurity increases from the initial value in solution (N_1) to the maximum value corresponding to the pure liquid ($N_1 = 1$) and the enrichment coefficient is 10^3–10^4. In the development of an indirect CD method[45] for the determination of impurities in water (with preliminary extraction of the sample by a volatile solvent and its subsequent CD analysis) the losses of the impurity component arising in isothermal analysis [see Eqn.(8)] were taken into account in constructing a linear calibration.[93]

Whereas on filled columns the solvent and the sorbate are as a rule distributed uniformly, on CC, in particular when the sample is injected through an evaporating injector, certain characteristic features are observed. The use of a solvent during the concentration of the sorbate induces in many instances a distortion of the peaks—they are greatly broadened and even split into two components on the chromatogram. The distribution of the sorbate within the limits of the soaked section of the CC has been investigated using the method described by Grob.[69] The distribution of perylene on the surface of the CC after the introduction of concentrated perylene solutions was investigated in ultraviolet light (in a separate chamber). Subsequent experiments with dilute perylene solutions were performed under the same injection conditions with recording of the chromatogram and were compared with visual observations. When the sample is injected directly into the CC at 25 °C, the impurity is not always distributed on the column in the form of a uniform film. Depending on the conditions, most of the impurity component is concentrated at the ends of the liquid zone with a length amounting to several centimetres. In this case split peaks are observed on the chromatogram, the first of which is usually higher than the second. When perylene is injected through an evaporating injector, it is concentrated mainly in the hot zone at the inlet to the CC. The peaks then have the form of a "stool" with the flat portion turned towards the start of the experiment. The distorted peaks of all the components were clearly seen in experiments on a mixture of C_{12}–C_{36} n-alkanes diluted with acetone ($1:2 \times 10^5$) in which the temperature was programmed in the range from 30° to 300 °C.

The conditions governing the formation of a narrow band of impurities when the sample was injected into the CC through an evaporating injector have been investigated.[79] The evaporator and column temperatures during the injection period are kept constant (25 °C). Ten solvents of different classes with t_b up to 69 °C were investigated. In order to improve separation, the author recommends that the column temperature be reduced, that a less volatile solvent be chosen for the analysis of heavier alkanes, that a fairly large amount of liquid be deposited, etc.

The main principles governing the broadening of the bands on the bed in CC and the distortion of the peaks on the elution curve have been formulated.[86] It has been shown in relation to the analysis of the C_{22}, C_{26}, and C_{30} n-alkanes that repeated decrease of the thickness of the SP film (by washing it off) in the range from 2 to 0.03 μm leads to a proportional decrease of the peaks. In fact the author proceeded to the examination of CD on an inert carrier in this instance.

Experience gained in the application of the SE showed that the injection of large doses in CC washes the SP out of the initial section of the column, leads to its accumulation at some distance from the point of injection, and finally results in the impairment of the efficiency of the column and even in its blocking. In order to eliminate these undesirable phenomena, a method has been developed in which the SP is washed out of the first two metres of the CC in a preliminary step.[94] For this purpose, approximately 150 μl of methylene chloride, containing 0.1% of Carbowax 1000, was introduced from a microsyringe via an elastic connection, and then, after several seconds, the liquid was blown out in the opposite direction. The operation was repeated three times. Along the section of the CC 2 m long, it is possible to deposit 5–8 μl samples. Various procedures for the preparation of CC without SP along the initial section, in which the SP is extracted or burnt off, have been described in detail.[87] Empty pre-columns connected to the CC either by a flexible Teflon tube or via screw thread or glued junctions have also been used. The MEGA Company supplies prepared CC with an empty end section (3 m long).

A series of new factors leading to the broadening and distortion of the peaks have been treated systematically[77,78,95] and the mechanism of the formation of the sorbate band as a function of the experimental conditions has been considered. The only injection method ensuring sharp initial sorbate bands involves split injection. The remaining methods require a concentration process over a period up to at least 0.1 s (or over several mm along the bed). The broadening of the bands in the injection of large samples can arise for two reasons. Firstly, when samples are injected through an evaporating splitless injector, the process in which the sample is transferred from the evaporator to the CC bed entails an appreciable increase in the injection time (on average up to 30–60 s), which induces the spreading of the peaks on the chromatogram—the so-called "band broadening in time". The peaks of the components on the chromatogram have the same width. The broadening of the peaks is eliminated by improving the condensation conditions, by employing an additional solvent (limiter) (see Section IV), and by programming the CC temperature (the broadening of the peaks is not observed for substances which are eluted at a column temperature 60–80 °C higher than the initial temperature). The second cause of the broadening of the band is associated with the distribution of the liquid and together with it also of the sorbate over a definite fairly long section of the CC—the so-called "band broadening in space". On average, the soaked section can occupy between 10 and 60 cm of the length at the inlet to the CC. In this case the magnitude of the effect depends on the ratio of the lengths of the wetted and SP-coated sections of the column. Thus approximately 2% of the length of a 15 m column is wetted, which decreases the number of theoretical plates (NTP) by a factor of ~5. The "band broadening in space" is manifested for peaks of heavy components (with a relative capacity $k' > 30–50$) and is independent of the programming of the temperature. It has been noted[77,78] that all the types of spreading indicated above depend on the ratio of the vapour pressures of the substance analysed and the solvent. The processes on the solvent are regarded as the first stage of the chromatographic separation. In order to investigate the process involving the isolation of the impurity from the solvent, a method has been developed for sampling at different stages of the distillation of the solvent.[78] A capillary section 1 m long, deactivated with Carbowax, was placed between the injector and the CC. This pre-column, containing the entire sample, was maintained for a definite period at the inlet of the CC and then the residue from the sample was blown out into the atmosphere and the experiment was terminated. When a non-polar solvent (hexane) was used, the higher-boiling alkanes always emerged in the form of narrow peaks and were eluted from the pre-column attached to the CC together with the last portion of the solvent. If the experiment was performed under conditions where there was no condensation (using pentane as the solvent injected via an evaporator), then all the peaks were broadened to the same extent, corresponding to the time taken to transfer the sorbate onto the CC. The formation of narrow alkane peaks can be accounted for by the occurrence of two types of CD processes. In the first place, less volatile impurities, having a much lower vapour pressure than the solvent, are concentrated near the rear solvent edge along a short section between the evaporator on the CC. Furthermore, as a result of limiting CNSV processes (see below), the possibility of the appearance of steps before the peaks is eliminated. The influence of these two CD processes on the formation of narrow peaks was not considered by Grob.[77,78]

4. Distribution of the Sorbate along the Bed and Determination of Light Impurities

In the development of a direct method for the analysis of organic impurities in water, it was established that all hydrocarbons sparingly soluble in water emerge from an isothermal CD column almost at once in a single peak.[45,46] The possibility of evolving the impurity in the form of a narrow zone, enabled the authors to develop a series of CD methods for the determination of petroleum products in water.[49] The possibility of determining the solubility of C_6–C_9 alkanes in water at concentrations of 1–10 mg litre^{-1} and also of determining the content of petrol fraction hydrocarbons in water has been demonstrated.

In the determination of light impurities on CC, their evolution with the first portions of the solvent was also observed.[78] Thus approximately 90% of 2-propanol was transferred from hexane to the CC over a period of 20 s. As a rule, such conditions arose when there was a marked difference between the polarities of the solvent and the sorbate (for example, for ethanol as the impurity, hexane as the solvent).

Together with the normal SE, the "reverse solvent effect", manifested by the broadening and distortion of the peaks due to substances emerging before the main component, has also been discussed.[96] Both light impurities (C_7–C_{10} n-alkanes and certain aromatic compounds) and heavy impurities (durene and dodecane) have been determined in solution in undecane. The experiments were performed with slight stream splitting (1:100) for a dose of 0.015–8.0 μl. The efficiency, expressed in terms of the number of theoretical plates, fell with increase of the dose by a factor of 2–4 for the light components. The explanation is that the sorbate band falls within a field with a negative liquid phase distribution gradient (section II, Fig.6). As a result of the arrest of the rear edge of the band and the acceleration of the leading edge, a reverse SE, leading to the distortion

of the peaks, is manifested. For the heavy component (durene), an increase of the NTP by a factor of 9 was observed.[96] This effect has also been discussed by Yang et al.[97] and Brötell et al.,[98] who obtained anomalously high efficiencies with respect to C_8 and C_{10} n-alkanes (4390 and 14400 plates m^{-1} respectively). The formal application of the familiar expression for the NTP (Fig.7).

$$n = \frac{5.54}{L}\left(\frac{t_R}{\mu}\right)^2$$

leads to unduly high values of this quantity. In the determination of the true retention times of the components for the correct calculation of the thermodynamic characteristics of the substances and their identification, it is essential also to take into account the shift of the retention times. The use of a heavier solvent results in an increase of the elution time of the impurity components. This effect is attained on a lighter solvent (pentane, hexane) with increase of the dose.

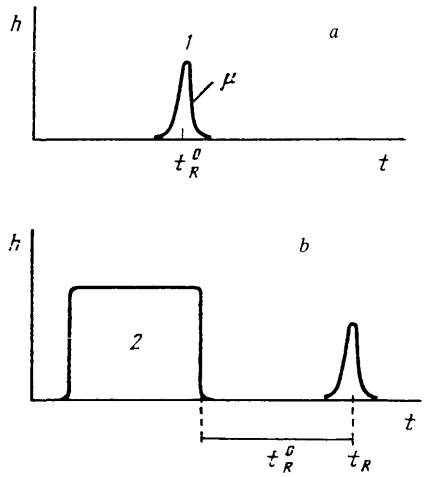


Figure 7. Schematic explanation of the calculation of the NTP (n) in the injection of a sample without solvent (a) and with solvent (b): 1) impurity; 2) solvent; t_R = retention time; μ = peak width.[96]

Factors leading to the distortion of the peaks have been examined in relation to the determination of chloroform impurities in solvents.[99] The authors claim that the distortion of the peak owing to the partial trapping of the impurity cannot be eliminated with the aid of the subsequent process on the solvent-saturated SP. Chloroform peaks with ideal shape were obtained only when water, acetone, and diethyl ether were used as solvents combined with a polar SP. In the first two cases the successful result can be accounted for by the formation of azeotropes in the systems indicated, which lead (as mentioned above) to the rapid separation of the impurity from the main component.

Studies have been made[100,101] on the additional spreading of the peaks due to substances emerging before the solvent. In experiments with model mixtures of C_5–C_{12} n-alkanes in octane and ethanol, it has been shown that "effectively" trapped components emerge in narrow peaks and "poorly" trapped components emerge in the form of broad steps. It is noteworthy that the boundary between the "poorly" and "effectively" trapped impurities is indefinite and is associated, according to the authors,[100] with distillation processes. In conformity with the classification of impurities into light and heavy presented above,[49] one must distinguish impurities capable of forming solutions having a vapour pressure greater or smaller than that of the solvent. Thus when ethanol is used as the main component, all the alkanes up to n-heptane form azeotropic mixtures[83] and are evolved at the leading edge of the solvent. Higher-boiling n-alkanes, which do not form azeotropes, should emerge after the more volatile solvent. The recommended increase of the initial column temperature in order to reduce the solvent injection time can lead only to a partial narrowing of the peaks.[100] A more radical means of compressing the peaks of light impurities involves the creation of a negative temperature gradient (see Section V).

The questions examined in this Section show that isothermal elution constitutes the initial stage of the CD separation and in many instances can be used directly to concentrate and determine impurities.

IV. LIMITING CHROMADISTILLATION

In the previous Section it was shown that the IE of a mixture entails extensive spreading of the peaks of both light and heavy impurity components. The broadening of the sorbate bands as well as the losses of substances associated with the evolution of the impurity component together with the solvent zone can be eliminated with the aid of limiting CD.[2,8,9]

Limiting CD consists in a procedure where a dose of the pure substance, whose vapour pressure is greater than or equal to the vapour pressures of the mixture components is deposited beforehand, before the injection of the mixture to be separated in a column filled with an inert carrier. The main mixture component (the solvent) can be used as a compound of this kind, referred to as the limiter, when the determination of the lighter components is not required.

The mechanism of the action of the limiter on a column without a SP is explained in Fig.8. In the initial instant the limiter *3* is deposited on section II of the column and the mixture (*1*, *2*) is deposited on section I (Fig.8*a*). In the first elution stage (Fig.8*b*), the following processes take place. The frontal IE of the mixture, described previously (Fig.3) and leading to the evolution of the pure less volatile component *1*, takes place at the rear edge. At the leading edge of the *1*, *2* zone the saturated vapour is transferred by the stream of carrier gas to section II and is condensed at the rear edge of the limiter. In conformity with distillation laws,[80] the formation of the zone comprising the mixture having the initial composition should be accompanied by the simultaneous liberation of the pure more volatile component *2*, which forms its own zone intermediate between the zones of the mixture and the solvent. It follows from the theory of limiting CD[21] that the rate of migration of the rear edge of the mixture zone (u'_2) (Fig.8*c*) is greater than the rate of migration of the leading edge (u''_2):

$$u'_2 = \frac{\alpha c_2^0}{q_{1,2}} > u''_2 = \frac{\alpha c_1^0}{q_{1,2}}, \qquad (13)$$

where $q_{1,2} = q^{II}/(N_2 + \beta_{2,1}N_1)$ is the amount of the liquid mixture *1*, *2* per unit volume of the column on section II, as a result of which the zone of the *1*, *2* mixture on the bed disappears and ultimately the mixture components are fully

separated (Fig.8*d*). A typical CDG for this case is illustrated in Fig.8*e*. Thus the separation in limiting CD is a result of numerous frontal separation processes at the rear and leading edges of the liquid. A relation has been derived[6,8] between the amount of limiter Q_{lim} and the dose Q_{mix} (of a mixture of known composition) which ensures complete mixture separation:

$$\frac{Q_{lim}}{Q_{mix}} > \frac{N_2 + \beta_{2,1} N_1}{\beta_{2,3}(\beta_{2,1} - 1)}, \qquad (14)$$

where $\beta_{2,1} = c_2^0/c_1^0$ and $\beta_{2,3} = c_2^0/c_3^0$. It follows from this relation that a definite length of the bed is required along which the rear boundary of the *1*, *2* zone formed (Fig.3*c*) should be able to catch up with the leading boundary before it emerges from the column. In essence the main mixture separation process takes place and is completed on section II of the limiter. Special cases for two problems in the determination of impurities follow from Eqn.(14).

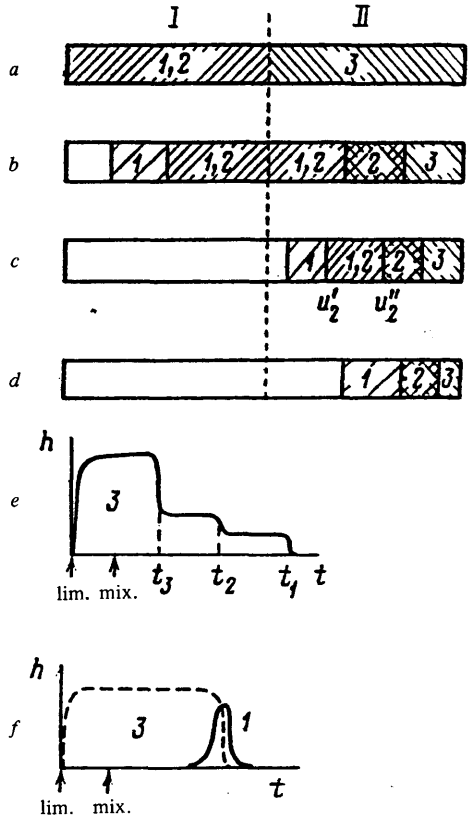


Figure 8. Schematic illustration of limiting CD.[21]

$N_1 \to 0$—determination of a heavy impurity. For this purpose it is useful to employ the pure solvent as the limiter. Then $\beta_{2,3} = 1$ and we obtain the relation

$$Q_{lim} \geqslant Q_{mix}(\beta_{2,1} - 1). \qquad (15)$$

In contrast to the CDG of the impurity component obtained in IE (Fig.3*g*), in this case a narrow peak of the impurity (without a low-slope stepped section) is observed on the elution curve and is located at the rear edge of the solvent (Fig.8*f*).

$N_1 \to 1$—determination of a light impurity. Under these conditions, Eqn.(14) assumes the form

$$Q_{lim} \geqslant Q_{mix}\beta_{3,1}/(\beta_{2,1} - 1). \qquad (16)$$

The impurity will be concentrated at the boundary between the limiter and the main mixture component.

It follows from Eqn.(14) and its corollaries that in most practical cases ($\beta_{2,1} < 2$) the complete separation of the mixture and the removal of the impurity require that the volume of the limiter should as a rule exceed the dose of the mixture to be separated. Large amounts of the limiter are required both in the separation of the mixture components with similar vapour pressures and in the isolation of the more volatile mixture component and when a more volatile limiter is employed.

Questions concerning the theory of the impurity isolation process, which in fact constitutes limiting CD, have also been examined by Pretorius et al.[76] A relation was obtained for the solvent and sample doses which ensure the complete isolation of the heavy impurity component in the form of a single peak on the chromatogram. It is identical with our Eqn.(15). The derivation was based on the consideration of the elution of the impurity component zone in terms of the more volatile solvent. The authors determined the rate of migration of the leading edge u_2'' in accordance with the partition coefficient, which is in the general case incorrect because, it follows from Eqn.(13), that at high sorbate concentrations the rate u_2'' depends on the mixture composition.

We carried out an experimental test of the principal relations of limiting CD on a packed column[6,8] and showed that the use of the limiter leads to the concentration of the substances. Thus, if the concentration of the test substance in the gas phase is less than the vapour pressure and the substance enters the column filled with the more volatile solvent together with the stream, then the given component will accumulate at the rear edge of the solvent and, as a result of the CD separation, can form a zone of the pure liquid at the concentration of the saturated vapour in the gas phase. This accumulation principle has been realised[43,44] in the determination of heavy impurities in a solvent and it has been shown that, after a single injection of the limiter dose, it is possible to inject repeatedly the mixture doses and obtain a proportional accumulation of the impurities.

Analysis of light impurities in solvents, even when the impurity components are sparingly soluble, is frequently hindered by the slowness of their evolution. Thus, in the direct analysis of alcohols in water, the impurity zone emerges from the preliminary CD column in the course of several minutes in the form of a peak with an extended "tail". The introduction of the limiter (ethanol) makes it possible to collect the entire test substance in the form of a narrow step at the closing front of the limiter. In studies[48,51] on the analysis of impurities as fermentation products (wine-making materials, brandies, etc.), it was noted that the accumulation of impurities at the boundary between two solvent zones (ethanol–water) leads to additional compression of the impurity peaks. A method was developed for the determination of impurities in wine-making materials, which are evolved at the leading front of ethanol, at the boundary between ethanol and water and at the rear edge of water.[48]

In studies by non-Soviet workers, the principle of limiting CD has been used to determine impurities using direct injection.[72,73]

The preliminary injection of the solvent to ensure the complete isolation of the impurity was first used by Deans.[72] Thus, in order to determine Carbitol to an aqueous 20 μl sample, 20 μl of pure water was introduced into the column beforehand. The author points out in this connection the necessity to select the appropriate solvent dose in order to attain the complete isolation of the impurity component.

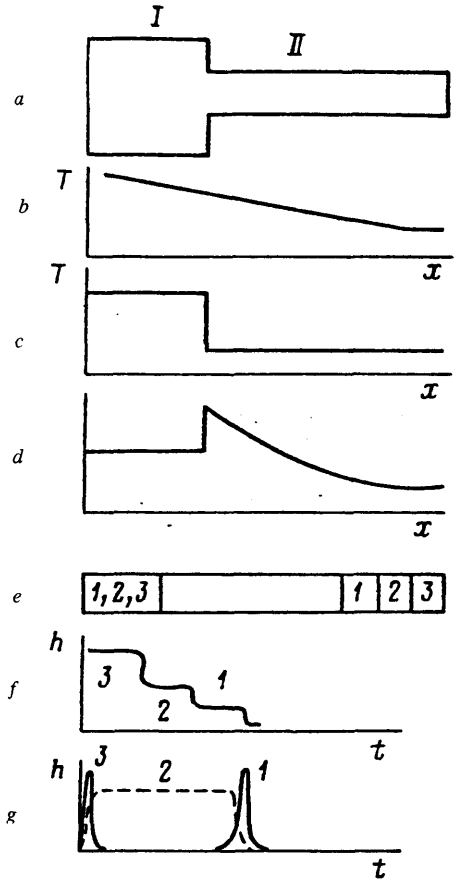


Figure 9. Schematic illustration of thermal CD:[7] *a*) junction between the injector (I) and the column (II); *b*) temperature field in thermal CD; *c*) ditto with evaporating injector; *d*) ditto along CD column; *e*) distribution of components I, II, and III along the bed; *f*) CDG of the mixture; *g*) CDG of impurities *1* and *3* in the solvent *2*.

A method for the injection of large gaseous samples (in vapour phase analysis), which is a development of the direct injection method with SE, has been considered.[73] Samples of mixtures of octane and air were injected by means of the usual injector, permitting the splitting of the stream. These experiments must be regarded as the version of limiting CD in which the condensation of large gaseous samples in the CC is achieved with the aid of the preliminary introduction of the solvent into the column. The unduly low heights of the octane peaks observed in experiments following an increase of the interval between the injection of the liquid and gaseous samples are due to the fact that the main bulk of octane failed to "catch up" with the rear edge of the limiter (Fig. 8c). The conditions under which the SE is manifested were investigated quantitatively in the above study. The simultaneous injection of the solvent and the gaseous sample naturally led to a negative result—the bulk of the octane passed through the CC without being retained on hexane. After the replacement of the solvent by pentane and the increase of the dose from 2.5 to 3.5 μl, the limiting effect was not observed at a CC temperature of 35 °C (apparently pentane did not condense). At room temperature a moderate impurity retention effect was noted.

The limiting CD process examined in this Section constitutes an effective means for isothermal concentration and isolation of impurities from solvents.

V. THERMAL CHROMADISTILLATION

As already mentioned above, the IE of a mixture involves only partial separation into individual components (Fig. 3). In order to achieve complete separation of the mixture, it is necessary to ensure conditions for the condensation of the saturated vapours formed at the leading edge of the band. The simplest method whereby condensation conditions can be created involves the use of a temperature field with a negative gradient (the temperature should decrease monotonically from the inlet to the outlet from the column—Fig. 9*a* and 9*b*). Under the conditions of thermal CD,[7] the repetition of the condensation and evaporation processes is achieved as a result of the movement of the entire mixture along the bed, which leads to its separation into the pure substances at the outlet from the column (Fig. 9*e*). The elution curve (the CDG) has a stepped form (Fig. 9*f*), where the height of the step corresponds to the vapour pressure and its length to the amount of each component. In CD the impurities are concentrated at the leading or rear edge of the main component (depending on the type of solution formed).[49] As a rule, the concentration of the impurity substance in the sample is insufficient to form a separate zone and for this reason the impurities are isolated in the form of narrow bands (Fig. 9*g*).

The temperature gradient is the most effective means of separation in CD. In contrast to chromatography, a stationary temperature field in CD results in the concentration of the substances, since repeated mixture condensation steps take place continuously along the bed, each leading to the liberation, in conformity with distillation laws,[80] of the lighter component and to the separation of the mixture as a whole. It is noteworthy that a longitudinal temperature gradient also arises in fractionation, but its character is wholly determined by the composition of the mixtures formed in each section of the column. In the continuous separation of a mixture under adiabatic conditions, a stationary distribution of concentrations arises along the packing. The heat supplied to the lower part of the column leads to the boiling of the liquid in all the sections; the temperature can then vary only in the range of the t_b of the components to be separated. In contrast to fractionation, the temperature gradient in CD is specified from outside. The process is based neither on boiling nor on evaporation. The discrete nature of the injection, the absence of a countercurrent flow, and movement along the bed, as in chromatography, lead to a simpler way of achieving multiple separation steps, which

makes it possible to increase the efficiency of the separation. In contrast to chromatography, under conditions where components are separated by thermal CD (Fig. 9e) stationary fronts are observed[19] and there is no progressive spreading of the bands. In thermal CD the separation proceeds not only via frontal stages at the rear and leading liquid edges (as in limiting CD) but also along the entire layer of the mixture, since condensation takes place in all the sections of the column. Visual studies of thermal CD have shown that each component of a mixture boiling over a wide range occupies a definite section of the column within the limits of which a uniform distribution of the liquid on the bed is maintained (q = 0.2 ml of the liquid in 1 cm^3 of the column). Both boundaries of a strongly wetted section (leading and rear) migrate along the bed in such a way that the zone width remains unchanged. This made it possible to develop a simple visual method for the analysis of high-boiling substances.[22]

In contrast to IE (section III) and vapour phase analysis,[81,82] thermal CD leads to the concentration of light impurities. Thus it has been shown[50] that, in the analysis of a butanol–water mixture on an isothermal column, butanol emerges in the form of a peak with a highly extended base despite the presence of positive deviations from Raoult's law (γ_2 = 45). The creation of a longitudinal temperature gradient compresses the alcohol band and enriches it by a factor of 250. As mentioned above, in effective thermal CD, the impurities are evolved at both edges of the principal component. There is therefore a possibility of different versions of the group separation of impurities. In relation to the isolation of butanol and acetic acid from water, it has been shown[15] that, by varying the dose, it is possible to regulate the distance between the components indicated.

All the principal results in the analysis of impurities have been obtained under the conditions of thermal CD. The methods described were developed mainly for the determination of impurities in aqueous systems.[49] Two versions of analysis are proposed: the direct version where the aqueous mixture to be analysed is injected directly into the CD column and the indirect version where the substances to be determined are first extracted with a solvent. The degree of the additional enrichment as a result of extraction was (depending on the ratio of the volumes of the aqueous sample and the solvent) between 40 and 200. On the basis of the first version of analysis, the following procedures have been developed:[45,51] determination of hydrocarbons dissolved in water, isolation of the petrol fraction and its separation by group distillation, the determination of light alcohols (C_1–C_5) which are readily soluble in water, and the separate determination of the sparingly soluble fractions of high-boiling alcohols (C_7–C_{13}). A method for the determination of impurities, which are evolved before ethanol, at the ethanol–water boundary, and after water, has been developed for the analysis of fermentation products containing two main components (water and ethanol). The relative standard deviation in the determination of alcohols at the level of 10–100 mg $litre^{-1}$ was 0.2–0.5 for a fiducial probability of 0.95. When measurements were made at the level of the limiting permissible concentrations (LPC) of alcohols in drinking water (0.1 mg $litre^{-1}$), a 20–30% change in the composition of the solutions was observed after several hours. The use of a solvent in the indirect methods (hexane or carbon tetrachloride) as well as an additional solvent in the direct methods (ethanol) permits the determination of components which are sparingly soluble in water and which form emulsions with the latter. The sensitivity of the indirect analytical methods for the petroleum products with t_b corresponding to the C_7–C_{20} n-alkanes was 0.014 mg $litre^{-1}$ and was limited by the purity of the solvents employed.[45,93]

The characteristics of CD, which arise when a moving temperature field with a negative gradient is employed—the so-called chromathermodistillation (an analogue of chromathermography), have been considered.[15,105,106] This method ensures the concentration of impurities less volatile than the solvent as a result of their evolution at higher temperatures and also makes it possible to shorten the duration of the analysis.

We shall now consider studies in which the SE was ensured by a negative temperature gradient along the column. In the initial version of the direct injection method,[53,55] the usual injectors were employed for the CC in which the outlet for discarding the carrier gas was closed during the injection period (30 s). A sharp temperature gradient was then employed (Fig. 9c)—the sample was evaporated at a high temperature (210 °C) and was condensed on the CC at room temperature. In the first studies the compression of the impurity band was observed. In order to investigate the compression effect, the sample was injected over different periods (τ_{inj}) and the width of the sorbate band on the elution curve (τ_{el}) was measured. The ratio of these quantities characterised the degree of enrichment, which gradually increased in the series of cycloalkanes from C_4 to C_8 and amounted to ~10. A method was also developed for analysis in which the solvent is discarded. A section of a capillary 1.6 m long was connected to the injector, the working flow rate was established, the sample was injected into the evaporator (at 210 °C), and it was condensed on the capillary at 25 °C over a period of 30 s. The gas flow was then stopped, the outlet end of the capillary was connected by means of a Teflon tube to the main CC, and the experiment with programmed temperature was begun. It was then shown that the losses of light components (C_4–C_5), emerging after the solvent, are 60–80% and those of the heavy C_6 component are 10%. The experiments in which the inlet capillary was cut into three sections (40, 40, and 80 cm) and the contents of the latter were subsequently analysed on the CC confirmed that volatile impurity components accumulate at the closing solvent front in CD. The use of an unduly short (albeit with a high gradient) temperature field (Fig. 9c) for a small difference between the vapour pressures of the solvent and the substances analysed leads to a decrease of the efficiency of the CD separation. The incomplete extraction from the solvent of even heavy impurities shows that a single condensation step in the thermal version of CD is insufficient.

It has been noted[79] that the difference between the temperatures of the evaporator and the CC can be used both in the vapour phase analysis in the absence of a solvent and in the analysis of a sample with a solvent. When the column is moved in the thermostat, a large gradient is established which improves the shape of the peaks.[86]

It was not clear from previous studies of the SE to what extent this effect determines the impurity concentration process, because one could also postulate that it is associated with the low-temperature extraction. The conditions making it possible to differentiate these processes have been investigated[94] using an injector, for the direct injection of the sample into the CC.[67]

Certain theoretical questions associated with the distortion of the sorbate band in direct injection into a cooled column have been examined.[102] The authors assumed that direct injection leads mainly to a distribution of the liquid with decreasing amount of the latter along the carrier gas stream. For this reason, one should have observed a distortion due to the reverse SE (Fig. 6, section II).[96] When the initial

section of the CC is unduly cooled, a temperature gradient, increasing along the CC, arises, which also enhances the "reverse SE". It follows from the examination of the retention characteristics adopted in GLC[89] that this entails the broadening of the band in the temperature field. The authors[102] suggest that the peaks be compressed by establishing a negative temperature gradient, i.e. in essence by intoducing the elements of thermal CD into the experimental technique. For this purpose, in the devices described by Galli and co-workers[103,104] it is necessary to supply cooling air in the direction opposite to the flow of the carrier gas from the side of the warmer column thermostat.

In order to assess the possibilities of the proposed method and also for comparison of the expected results with the data actually obtained we shall carry out the following calculation. We shall assume that the concentration of the impurity substance is measured by a flame-ionisation detector whose sensitivity is the same in all the methods compared and amounts to $c_{fid} \cong 10^{-4}$ (vol.%). With the aid of a linear detector, such a concentration can be recorded on the 10^{-11}–10^{-12} A electrometer scales. The minimum concentration which can be determined in the initial mixture (injected into the chromatograph) is related to the experimental parameters by the following formula:

$$c_{min} = \frac{c_{fid} v_0 t_\mu M}{2.24 V_{ds}} \text{ (mg litre}^{-1}\text{)},$$

where v_0 is the carrier gas flow rate (ml min^{-1}), t_μ the peak width on the chromatogram (min), M the molecular weight of the substance analysed, assumed to be 100, and V_{ds} the volume of the sample injected (ml). We shall examine the numerical data for two principal cases.

1. Analysis on a filled chromatographic column with injection of the sample into the CD pre-column. We shall assume that $v_0 = 20$ ml min^{-1}, $t_\mu = 0.5$ min, and $V_{ds} = 1$ ml. In this case $c_{min} = 5 \times 10^{-2}$ mg litre^{-1}, while the concentration actually determined[45] was 5×10^{-3} mg litre^{-1}.

2. Analysis on a CC using the direct sample injection methods. If $v_0 = 2$ ml min^{-1}, $t_\mu = 2$ s, and $V_{ds} = 10$ µl, then $c_{min} = 3 \times 10^{-2}$ mg litre^{-1}. The results of the analysis of solutions containing 1–20 p.p.m. of the test substance have been quoted in studies on the SE. In both cases presented here, c_{min} is of the same order of magnitude despite the difference in the dose by a factor of 100. This can be explained by the higher efficiency of the CC, working at a lower flow rate, which leads to the isolation of the impurity in a small volume and hence to a greater sensitivity of the measurements on the CC. It follows from the data presented that the use of the CD pre-column in combination with a CC can increase the sensitivity by a further two orders of magnitude, whilst retaining the efficiency of the CC, which makes it possible to attain a level of measurable concentrations of about 0.1 p.p.m.

Studies on thermal CD have shown that the temperature gradient influences the mixture separation to the greatest extent. In order to enhance the concentrating effect, one can combine the limiting and thermal CD processes.[74]

VI. CHROMATOGRAPHY OF NEARLY SATURATED VAPOURS

This Section deals with studies in which the stationary phase is greatly overloaded by the solvent during the sample injection period. The isolation of the impurity components along the initial section of the column is accompanied by the interactions of the sorbates not only with the SP and the solvent but in many cases also with one another. These phenomena are described by the chromatography of nearly saturated vapours[9,15,17] in which one considers processes at high concentrations corresponding to the concave sections of the sorption isotherms (Fig.10*a*).

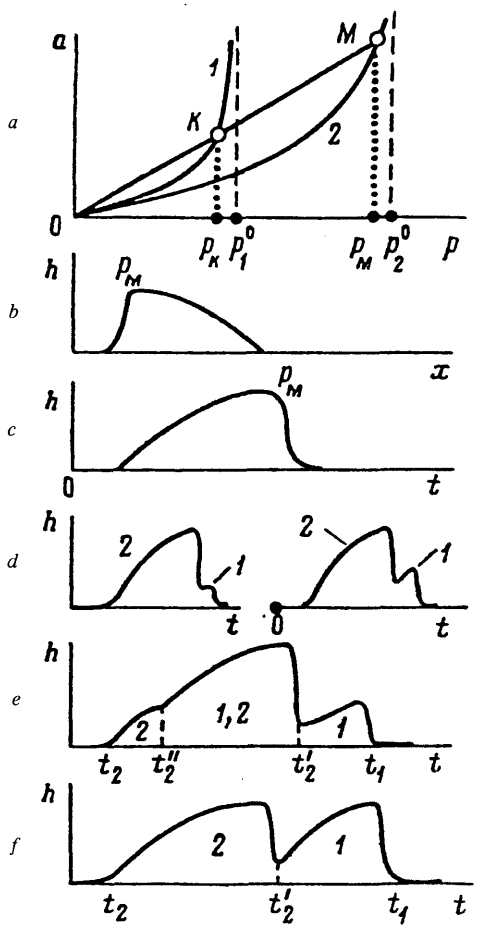


Figure 10. Schematic illustration of the CNSV process:[15] *a*) sorption isotherms for components *1* and *2* to be separated; *b*) distribution of the component along the bed; *c*) CDG for a single component; *d*) the possible CDG in the enrichment of the impurity; *e*) CDG for partial separation of the mixture; *f*) CDG for complete separation of the mixture.

It is known from the general theory of chromatography[56,88] that, under the conditions of concave sorption isotherms, the band of the substance analysed should have a greatly diffuse leading front and a cut-off rear edge (Fig.10*b*). A diffuse "tongue" and an abruptly terminated "tail" are recorded on the elution curve (Fig.10*c*). Chromatograms of this kind are observed in gas–liquid and gas–adsorption chromatography at concentrations close to saturation. The injection of large samples into the chromatographic column leads to concave sorption isotherms both for the solvent *2* and for the impurity *1*. The rate of migration of the points in the leading diffuse front along the bed is described by the derivative of Eqn.(1), while the rate of migration of

the curtailed rear edge along the bed is determined, in conformity with the material balance, by the slope of the straight line *OM* (Fig.10*a*) drawn from the origin of coordinates *O* through the point *M* corresponding to the maximum concentration at the front. The straight line *OM*, drawn for the rear edge of the more volatile component *2*, intersects the isotherm of component *1* at the characteristic point *K*. All the points in the leading front of component *1*, for which the derivative is less than the slope of the straight line *OM*, move along the bed faster than the rear edge of component *2* and can therefore catch up with it. This leads to the "limiting effect"—component *1*, dissolving at the rear edge of the limiter *2*, is retarded and concentrated. According to the material balance at the zone boundary, the following equation should hold:

$$a_2/c_{2M}=a_1/c_{1K}, \qquad (17)$$

where c_{1K} and c_{2M} are the concentrations in the gas phase corresponding to the points *K* and *M* and a_1 and a_2 are the degrees of adsorption at these concentrations. It must be emphasised that the final result of the enrichment is determined by the isotherms of the individual components and is independent of the character of the solutions formed in the intermediate zone between the components. The chromatogram of the less volatile impurity component *1* can have either the stepped form, when the initial concentration was smaller than at the point *K*, or can have a growing leading front in the opposite case (Fig.10*d*). However, in both cases Eqn.(17) should hold for the concentrations at the rear solvent boundary.

The advantages of the injection of large samples into a column with a SP for the separation of mixtures have been demonstrated.[5,15,17] It is known[56,57] that an increase of the dose in traditional chromatography leads to the overloading of the column and to the impairment of the separation owing to the overlap of the peaks. However, further increase of the dose makes it possible to attain operating conditions where CD effects exert an influence. The influence of the interaction of sorbate molecules not only with the SP but also with one another is then manifested. In contrast to CD, in the CNSV there is a possibility of using the sorbent selectivity for the improvement of the separation. The temperature gradient and the limiter promote the occurrence of distillation processes. The action of all the remaining auxiliary separation factors has been illustrated by means of examples.[5,15] Thus, after the injection of 0.1 ml of a liquid heptane–cyclohexane mixture into an isothermal column (at 10 °C) with tricresyl phosphate, a chromatogram similar to that illustrated in Fig.10*e* is obtained. The frontal processes at the leading and rear edges of the band lead to the partial isolation of the components, while the intermediate zone contains the unseparated components *1* and *2*. The use of a negative temperature gradient (from 75° to 10 °C) makes it possible to attain complete mixture separation (Fig.10*f*). From the standpoint of traditional chromatography, this result is paradoxical, since an increase of column temperature should impair the selectivity of the separation. The results obtained are associated with the CD processes on the SP. An analogous effect is attained by employing a limiter. In contrast to CD, in the CNSV there is a possibility of the separation of azeotropes or systems which are otherwise difficult to separate. This has been demonstrated[5,15] in the separation of the heptane–isooctane and benzene–cyclohexane mixtures on α-iodonaphthalene as the SP. Whereas the components are not separated under isothermal conditions (0 °C) for a dose of 0.04 ml, the application of a gradient (85–0 °C) leads to complete mixture separation.

CNSV processes are manifested and are used in the concentration and determination of impurities. A short (20 cm) chromatographic column with a thermal pulse has been used[44] at the concluding stage after repeated CD accumulation of C_9–C_{11} alkane impurities in heptane. The lowest concentration which could be determined with the aid of a katharometer was 10^{-3} vol.%.

In order to transfer the impurity from the preliminary CD column to the next, separating CC, it is necessary to create favourable conditions for the occurrence of the CNSV. In the analysis of the butanol impurity in water[51] without a limiter (ethanol) or under conditions which rule out the condensation of the alcohol (CC heated above the t_b of ethanol), highly diffuse butanol peaks appear on the chromatogram. In this case the butanol band passes from the CD column to the CC in a large volume of gas (20 ml). If a temperature difference is maintained between the columns (from 70° to 20 °C), ensuring the condensation of ethanol in the initial section of the CC, then the width of the butanol peak on the chromatogram diminishes by a factor of 7. The possibility of combining a CD pre-column with the CC in order to increase the size of the injected sample at least by an order of magnitude was demonstrated in the same study.

The role of the SP in the concentration of impurities from a large sample has been discussed briefly.[72] Harris[107] considered the influence of large samples injected into a column with a SP on the retention of the component under the conditions of marked deviations from Raoult's law. Taking into account the dependence of the activity coefficients on the mole fraction of the component in solution, the author determined qualitatively the character of the change in the retention volume of the impurity with change in the size of the dose. In fact Harris[107] stated the necessity to take into account the intermolecular interaction in the complex system comprising the components of the mixture to be analysed and the SP and the influence of this interaction on the results of the separation. It has been shown[77] that the presence or absence of the SP on the initial section of the CC does not affect the shape of the peaks when the latter spread. This is associated with the presence of a large amount of the solvent, which predominates along the section and forms an unusually large film. However, for comparable amounts of the SP and the solvent, their mutual influence on the isolation of the impurities is manifested.

A suitable solvent was selected[79] for the injection of the sample using the SE and it was established that there must be a definite correspondence between the polarities of the SP, the solvent, and the sorbate.

Jenkins[108] made a special study of the influence of solvent polarity on the shape of the peaks for the direct injection of the sample into the CC. A new method was developed involving the preliminary injection of an additional solvent:[73] several solvent samples were introduced by a single syringe using an air gap to separate different solvents. It was established[108] that the use of non-polar sorbates (alkanes) for injection in a polar solvent (methanol) leads to symmetrical peaks on silicone phases (of the type of SE 30 and 52). The use of a weakly polar secondary solvent (cyclohexane), which is injected before and after the main sample and forms an azeotrope with the first solvent, makes it possible to isolate alkanes in the form of narrow peaks. The solvent evaporation processes occurring are similar to azeotropic distillation.

A number of methods for "correcting" the shape of the peaks, corresponding to our ideas about the CNSV process, have been proposed.[102,108] It has been noted that polar compounds in a polar solvent have normal peaks, but the

shape of some of them is distorted. This is associated with the fact that the condensed solvent prevents the penetration of non-polar substances to the SP on the surface of the CC. In order to improve the process, it is recommended that one should use a mixture of solvents forming an azeotrope, which is equally permeable to both polar and non-polar substances. The prospects for the employment of two solvents simultaneously in order to elucidate whether the split peak corresponds to one substance or several substances have been discussed.[102]

In the course of several years, starting in 1976, a number of studies on the effects occurring in gas-liquid preparative chromatography when samples with a large content of the main component are injected have been published.[109-112] A number of characteristic features of the CNSV considered above and observed in the isolation of impurities from *m*-xylene (dose volume up to 10 μl) on a 640 × 1.4 cm column filled with Spherochrome with the PEGA 25% phase have been noted: for concave *m*-xylene sorption isotherms, an extended leading front of the main component is observed, while the rear edge is sharply cut off, which promotes a clear-cut isolation of the impurity; an increase in sample size shifts the retention times of the impurity components; the observed decrease of the width of the impurity peak following the main component, compared with the injection of an equal amount of an individual impurity component, has been observed.

The last two factors leads to a sharp increase of the formal HETP (by a factor of 10–20), which was also noted by Berezkin[113] and was discussed in Section III of the present review. This treatment of the mechanism of the phenomena occurring is close to that put forward by Jennings[60,89] and Grob[52,79] and is based on the consideration of the elution of the impurity band against the background of a binary solution with a concentration gradient of the main component in the SP.[114] It has been noted[109] that the transfer of the impurity in the field of a variable sorption capacity is analogous to the processes occurring in non-stationary chromathermography. Considering the processes from the standpoint of GLC, Mar'yakhin and Vigdergauz[109] clearly formulated the idea that the main component can play the role of the "sorbate-concentrator" (in other words the limiter). The sooner the impurity is eluted after the main component, the greater the increase in concentration under these conditions. It has also been noted that the main component can be introduced after injecting the mixture containing the components to be concentrated. It is convenient to use a main component which is not recorded by the detector. These features of the CD methods for the concentration of impurities (in the absence of a SP on an enrichment column) were used in our later investigations.[43-45]

The mechanism of the formation of a narrow zone of the impurity component under CNSV conditions, determined by the occurrence of frontal-sorption processes on the SP, has been confirmed in a series of studies published in 1983.[70,71,99-101] The authors investigated experimentally the band forming processes on two sections of the initial part of the CC. On section I (Fig.5), the SP is saturated by the solvent vapour. As noted above and demonstrated in a number of studies,[10,21,49,72] after the elution of the sample from the section I, not all the impurity substance is contained on the rear edge of the solvent. The complete isolation of the impurity component in the form of a single peak is attained by employing a limiter. However, in a number of experiments also without the limiter a complete isolation of the impurity in the form of a sharp peak has been observed on the chromatogram—this occurs in the subsequent separation of the injected sample on the SP in the CC. In order to investigate the process mechanism, a detailed experiment has been performed in which the distribution of the substance along the bed was determined.[70] A 20 m column consisted of a short etched capillary (1 m) and 15 sections on which a SP (OV-1) was deposited and which were connected by Teflon tubes. Some time after the injection, the column was taken apart and the sorbate contained in the part of the CC located between the injector and the disassembly point was blown out into the atmosphere. The CC was then reassembled and the chromatographic process was completed. The large number of disassembly points in the first half of the CC and experiments carried out with a shift of 2–15 s made it possible to obtain a detailed picture of the change in the distribution of the sorbate along the bed as a function of time. The results obtained can be represented by means of the series of diagrams in Fig.11. On the elution curve for the impurity extracted from the first section, a low "base" (110 s long) is observed, while at the end of the zone there is a narrow "tower" (5 s long), whose area corresponds to 70% of the injected octane. This result is fully consistent with the above description of the elution curves obtained on an inert carrier.[10,72] The theory of this process was examined by ourselves in a study of the isothermal CD elution of a mixture[21] (see above).

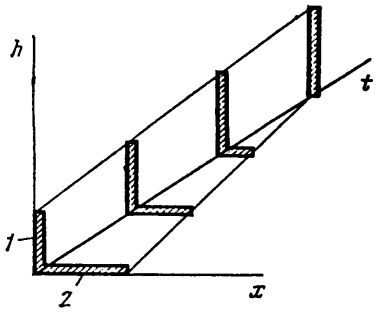


Figure 11. Distribution of the sorbate along the section of the column with SP at different times;[70] h = height of band on the bed; x = width of band: 1) "tower component"; 2) "base".

The experiments[70] in which the movement of the sorbate along the SP in a CC was investigated have shown, over the first 0.8 m, that the band consisting of a planar "base" and a narrow "tower" migrates almost without change. Then, over a section of the CC between 2 and 5 m from the origin, the "base" is rapidly shortened as a consequence of the increase of the area of the "tower" part of the peak and towards 6 m the entire substance is concentrated in a single narrow peak. This result has been confirmed by data obtained by comparing the rates of migration of the leading edge of the "base" of the octane band and of its rear "tower" component. Over the first 5 m, the "base" migrates at a rate of 2.4 cm s^{-1}, which is associated with its presence within the limits of a zone with a high content of the solvent. At a distance of 5–6 m from the start of the CC with a SP, the rate of migration of the leading octane front increases to 8.5 cm s^{-1}, corresponding to a linear rate of migration of octane along the SP in the complete absence

of the solvent. The "tower" component of the band passes slowly only over the first 2 m and then, at a distance of 2–3 m from the start of the CC, it acquires the maximum rate of 9 cm s^{-1}, which enables both fronts to be united after 3.3 min in the vicinity of the 6 m marker. The passage of the sorbate band through a narrow rear edge of the solvent leads, according to the authors,[70] to a concentrating effect. Initially the solvent band migrates more slowly than the sorbate band, which adjoins, via its "tower" component, the rear edge of the solvent over a long period. This can be seen from plots of the migration (Fig.12). Over the section from 2 to 5 min, the rear edge of the solvent begins to overtake by 2–3 s the rear edge of the impurity. During this period, both rear boundaries (of the solvent and impurity) are accelerated and in the vicinity of 5 m they combine with the leading boundary of the impurity, which migrates slowly within the limits of the solvent-saturated SP. Next, the rear solvent edge overtakes both boundaries of the impurity band and continues to migrate along the column. In short columns (5–10 m) an impurity peak 0.6–1.1 s wide was obtained directly behind the solvent zone after 3–5 min.

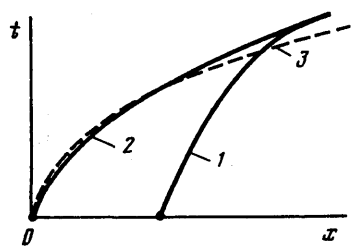


Figure 12. Graphical representation of the movement of the sorbate and solvent edges:[70] 1) and 2) leading and rear sorbate edges; 3) rear solvent edge.

It has been emphasised[71] that the concentrating effect on a solvent-saturated SP is associated with the difference between the rates of migration of the solvent and sorbate edges. The reduced rate of migration of the rear solvent edge accompanied by a high rate of migration of the sorbate along the solvent-free SP in fact leads to the compression of the band from 120 s to 1 s. The authors calculated the retention coefficient K_{ret} defined as the ratio of the rates of migration of the sorbate along the SP in the presence and absence of the solvent. In experiments with repeated successive injection of the sample and additional "supply" by the carrier gas saturated by the solvent vapour, a study was made of the dependence of K_{ret} on the nature of the SP, the solvent, and the sorbate. When solutions of decane in pentane ($1:10^3$) were injected at 3 min intervals, individual decane peaks, moving in succession at intervals of ~60 s ($K_{ret} \cong 3.5$) were observed. Analogous results were obtained in differential CD[16] with solutions of volatile components which were used instead of SP. It is noteworthy that the injection of a dose of the pure solvent makes it possible to select experimental conditions such that the impurity can be collected in the form of a single peak from several successive injections of the sample.[44]

We reached previously[2,9,15] the conclusion that the zone boundaries should adjoin on the basis of the study of the concave sorption isotherms of the components. The mechanism of the concentrating effect can be accounted for by the occurrence of a limiting process in CNSV. As the solvent band moves, it spreads and the concentration at the maximum on the curve falls. The increase of the rate of the sharp rear edge of the band is associated with the decrease of the slope of the *OM* secant on the plot of the sorption isotherm (Fig.10*a*). The solvent *2* therefore initially functions as a limiter until the secant drawn from the point *M* on the solvent sorption isotherm (corresponding to the maximum concentration) intersects the sorbate (impurity) sorption isotherm *1*. This period is in fact responsible for the effect involving a sharp increase of the concentration. It follows from this explanation that the impurity concentrations in the gas and liquid phases should increase. As the solvent migrates along the layer of SP, its concentration at the maximum falls and it moves ahead as the lighter component, while the concentrated impurities migrate in narrow bands further along the column in accordance with their partition coefficients in the SP.

The study of the mechanism of the concentrating process on the basis of the sorption isotherm makes it possible to predict the final result of the enrichment of the impurity at the rear solvent edge. This does not rule out the approach associated with allowance for chromatography on the SP with a gradient distribution of the latter. However, in this case it is essential to consider the movement of the SP along the bed. Certain quantitative relations describing the migration of the sorbate fronts at high concentrations have been published.[15]

VII. DEVICES FOR THE INJECTION OF LARGE SAMPLES AND THE RESULTS OF THEIR APPLICATION

The direct injection methods examined above require the creation of new designs of injectors. In the present Section we shall consider only the injectors in which the sample injection is accompanied by the formation of a large amount of liquid in the initial section of the column where the CD processes are manifested. The injection system and the method of injecting substances in many respects determine the quality of the separation. When a highly effective injection system is used, the following requirements must be met:[97] the absence of the overlap of the solvent peak with the peaks of the components to be separated; the elimination of the fractionation of the sample components; the reproducibility of the injection times with a relative standard deviation less than 0.1%; the reproduction of the normalised peak areas with a relative standard deviation less than 1%; the reduction to a minimum of the thermal decomposition and sorption processes as well as reactions; the constancy of the column efficiency; quantitative and representative extraction of both impurity and main sample components; independence of the injection processes of the operating conditions of the column.

In the studies on the CD determination of impurities[43–51] the sample was injected into the column through the usual injectors used in commercial chromatographs (Tsvet-100, LKhM-8MD, etc.). The initial section of the column (about 10 cm) was located in the evaporator of the injector and the sample was injected by means of a syringe directly onto the bed. Glass and metallic U-shaped and spiral CD columns, up to 3 m long and 2–4 mm in diameter, filled with metallic or etched glass beads (0.25–0.5 mm fraction) were employed.[44,45,49,50] In experiments designed to

isolate impurities under the conditions of thermal CD, a temperature field with a negative gradient was created along a section up to 1 m long. The gradient was established by employing a furnace with a variable-pitch nichrome spiral winding. The temperature along the field was varied from t_b for the solvent to room temperature. The temperature in the evaporator was kept somewhat below t_b of the solvent (Fig. 9d) in order to prevent the entry of the sample into the inlet connectors, since the injection of large amounts of liquid into a strongly heated column entails an increase of pressure. The use of a preliminary CD column made it possible (with the aid of a switching tap) to inject samples with volumes up to several millilitres.

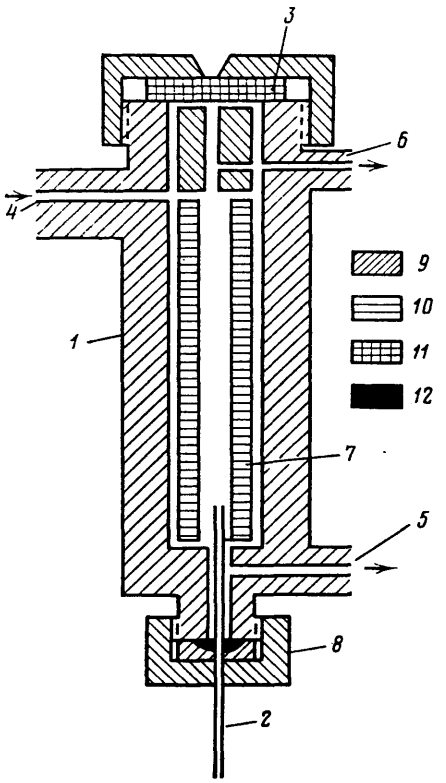


Figure 13. Schematic illustration of the evaporating injector for the splitless introduction of the sample:[52] 1) evaporator; 2) CC; 3) septum; 4) and 5) entry and exit of carrier gas; 6) flushing of the septum; 7) insert; 8) attachment of the CC; 9) metal; 10) glass; 11) rubber; 12) graphite.

Special injectors have been developed for the direct splitless injection of samples into a CC. The sample was initially[52] injected from the usual injectors (Fig. 13) in which the carrier gas disposal line 5 was initially closed during the transfer of the sample from the evaporator to the CC. In order to eliminate the influence of the septum 3 of the injector, a special device with a controlled leakage of the gas has been used.[5] The Carlo Erba Company later designed an injector[52,55] where the septum was in contact with a separate stream 6. The advantages of splitless injection have been demonstrated in analyses of an admixture of benzene and pentane (0.01 p.p.m.), steroids, and the components of tobacco scent and tobacco smoke. Injection without stream splitting has been used successfully for the analysis of natural scents and specimens of the ambient medium.[79] The Hewlett Packard Company has published manuals for the use of commercial injectors in routine analyses.[89]

Having analysed the system for injection into CC existing at the time, Yang et al.[97] concluded that the best injection system is one without stream splitting.

Data for the reproducibility of the injection method without stream splitting have been obtained[97] using the automatic injector of the Varian-3700 apparatus in relation to the analysis of a mixture of C_{13}–C_{32} n-alkanes with the temperature programmed in the range from 120° to 280 °C. The solvent was n-decane. The relative standard deviation of the retention time based on the results of 11 measurements was on average 0.05% and that for peak areas was 1%. The optimum parameters were chosen[97] using a simplex experimental design method. It has been suggested that one should be guided by the following rules for the selection of the parameters: sample size from 0.1 to 10 μl; rate of sample injection about 1 μl s^{-1}; sample injection time more than 20 s; time during which the blower is switched on after injection more than 40 s; effective dynamic range of sample dilution 10^5: 1; evaporator temperature not more than 230 °C (for solvents with t_b < 100 °C); the solvent should have t_b smaller by 25 °C than the t_b of the lightest component to be determined, since otherwise it is lost together with the eluted solvent; the initial constant temperature should be lower by 15–30 °C than the t_b of the solvent; the components evolved before the solvent can be determined quantitatively only when their relative capacity is sufficiently different from that of the solvent.

The splitless conditions ensuring the entry of 95% of the sample into the CC have been investigated.[115] The injector produced by the Carlo Erba Company for models 2900 and 4160 was used.

The injection of large samples (up to 50 μl) of solutions of C_{11}–C_{16} n-alkanes containing pesticides into large-diameter (0.77 mm) CC using an electron capture detector has been described.[98] The injection system, described by Grob and Grob,[52] in which the values for the ejection of the stream and for the flushing of the septum were closed throughout in order to eliminate the possible losses and to ensure the constancy of the flow rate through the column, was used in the above study on the basis of the Varian-1400 apparatus. Since the injection of large samples distorts the peak retention times, the results of the analyses were characterised by the separation factor K_1 for the DDT pesticide vapour. The relative standard deviation in the determination of K_1 varies from 1.5 to 5.5% depending on the solvent selected. The maximum value of K_1 was obtained when a 2–4 μl sample was injected. On further increase of sample volume, no significant decrease of K_1 up to 100 μl was observed.

In a number of instances where a SP is used, it is possible to achieve injection with stream splitting.[68] This permits a decrease of the stream splitting coefficient and hence an increase of the amount of sample injected. By virtue of the decrease of the volume of the vapour on condensation, the additional amount of substance is as it were "sucked"

onto the column. Under such conditions, the stream splitting coefficient is more stable and does not change during the injection period.

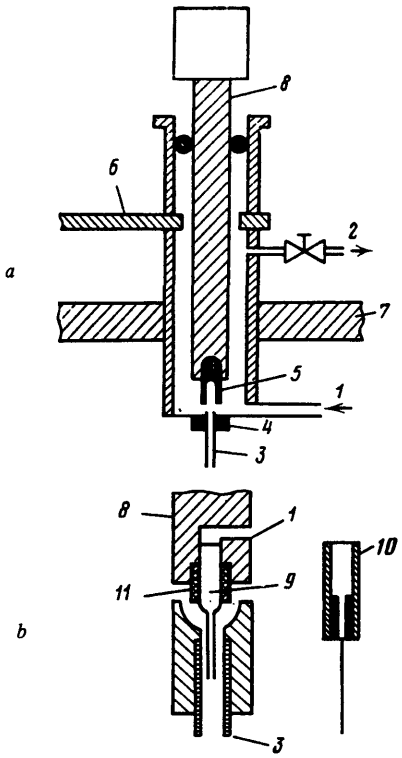


Figure 14. Schematic illustration of the injector for the direct introduction of the sample into the CC:[117] *a*) "macro"-version; 1) and 2) entry and exit of carrier gas; 3) CC; 4) seal; 5) crucible; 6) valve; 7) insulation of the thermostat; 8) valve shaft; *b*) "micro"-version: 9) and 10) micropipette; 11) silicone rubber.

Schomburg et al.[116–118] were the first to point out that the splitless method employing an evaporator is irrational. This is because ultimately one has to inject the initial liquid sample directly into the CC; for this reason the intermediate operations involving evaporation and condensation are superfluous in splitless injection. The disadvantages characteristic of the usual injection systems with stream splitting can then have an effect. Schomburg was the first to employ the direct injection of liquid into CC of normal size (0.3 mm) and described two designs for injectors:[117,118] the "macro"- and "micro"-versions. In the first case (Fig.14*a*) a 5–10 μl sample is placed in a small glass crucible *5* attached with the open end downwards to the shaft *8* of a sliding valve *6*. The shaft is exposed to the carrier gas which flows along line *1* in the CC *3*. In order to inject the sample, the liquid in the crucible *5* is brought into contact with the end of the CC. Under the influence of capillary forces, the liquid to be injected enters the CC. In the "micro"-version of the direct method (Fig.14*b*), the extended end of a micropipette (with a volume of several nanolitres) is inserted into the CC with the aid of the same valve *6*. The carrier gas blows the liquid into the CC. Special care must be taken to ensure that the CC and the pipetter are coaxial. The characteristics of the three injection methods (with and without stream splitting and also the direct method described) have been compared. The relative standard deviation (reproducibility error) in the determination of the composition of a mixture of C_{18}–C_{34} n-alkanes was not worse than 1% for all the methods. Two completely identical chromatograms for the analysis of the products of the Fischer-Tropsch reaction with a dose of 1 μl have been published. In the direct injection, the sample was diluted by a factor of 100 with octane and in injection with stream splitting the splitting coefficient was 1:100. Direct injection has the following advantages:[118] the separate evaporator unit with all its disadvantages (superfluous volume and surface, selection of flushing conditions, and the creation of a uniform gas mixture) is eliminated, there is no rubber septum, there is no need for a syringe, and there is no stream splitting. The disadvantages of the direct injection include the contamination of the inlet section of the CC by involatile sample components, the washing off of the SP, and the difficulty of replacing the injected volume.

The methods for sample injection with stream splitting have been criticised[67] and conclusions similar to those of Schomburg and co-workers[116–118] have been reached. In a general assessment of the methods of injection with stream splitting, it has been concluded[63] that the reproducibility is excellent but the results are poor.

Another version of the direct injection of the sample into the CC (Fig.15), differing from Schomburg's method by the fact that the injection is via the usual microsyringe through a special valve[119] without a septum, has been achieved.[63] Initially the needle of the syringe, whose outer diameter is smaller by 0.05 mm than the inner diameter of the CC (0.3 mm), is inserted in the channel *5* of the injector up to the shaft of the valve *4* sealing hermetically the channel. The shaft is then moved back and a small controlled ejection into the atmosphere is established in the gaps between the walls. The syringe is lowered and the needle is inserted into the CC via the guiding channels *3*. Samples with a volume of 0.1–0.2 μl are injected rapidly and those with larger volumes are injected at a controlled rate. The injector is placed in an insulating beaker with air cooling. The volume of the sample injected can be measured with the aid of such a device and the rate of supply of the sample, the cleanness of the surface, etc. can be monitored. The continuous operation of the injector described in the course of a year confirmed its advantages and led to an insignificant modification:[94] the lock nut was replaced by a two-position Teflon tap (open-shut) and cooling was achieved by means of a radiator exposed to air from the thermostat ventilator.

The results of the analysis of dilute solutions of polynuclear aromatic compounds in methylene chloride showed[67] that the relative standard deviation in the determination of the content of substances with the aid of the injector developed is 3–5%, which is smaller by a factor of 4–5 than the value obtained with the aid of an evaporating injector. The results of the use of the above system for the analysis of thermally unstable compounds—mustard oil and wide-boiling fractions of the methyl esters of C_8–C_{22} aliphatic acids—are quoted in the above study. The

relative standard deviation was 1.2–1.7% and was respectively smaller by factors of 3 and 5 than in injection with and without stream splitting.

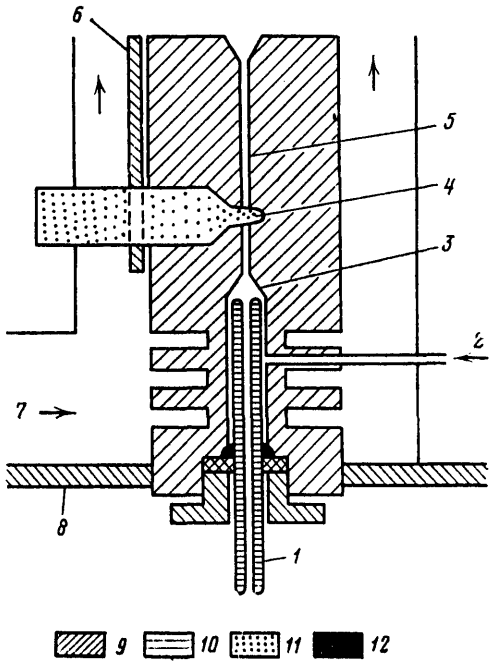


Figure 15. Schematic illustration of the injector for the direct introduction of the sample into the CC:[94] 1) CC; 2) entry of carrier gas; 3) centreing cone; 4) closing valve; 5) membrane replacement channel; 6) valve spring; 7) entry of cooling air; 8) thermostat insulation; 9) metal; 10) glass; 11) Teflon; 12) graphite.

The direct injection method[120] has been used as a reference procedure for the estimation of the accuracy of injection by other methods employing stream splitting after the evaporation of the sample. The technique of sample injection from a syringe was varied (cold and hot needle, air or solvent plug). In most cases concordant results were obtained for C_9–C_{20} alkanes, while for high-molecular-weight hydrocarbons close to C_{44} the usual method leads to losses of the test substance amounting to 25–75%. The authors believe that the components of a mixture with a wide boiling range are fractionated on the hot needle of the syringe. When the needle is heated to a temperature exceeding the t_b of the solvent, the high-boiling components are concentrated on the liquid meniscus at the piston and do not reach the CC. This required a change in the technique of direct injection onto the column[67] (also using a syringe) in the part concerned with the improvement of the cooling system.[103] In the "Fractovap" model 4160 injector,[121] provision is made for additional channels to allow the passage of cooling air and a heat exchanging insert is placed in the connecting ring, which makes it possible to cool the CC from the inlet to a distance 10 mm below the level of the column thermostat.

The accuracy and reproducibility of the injection by three methods (direct injection onto a cooled column and injection with and without stream splitting) were compared in the analysis of samples containing extremely high-boiling compounds—triglycerides.[122] The samples were prepared in hexane with added pentadecane as the standard. Direct injection onto the cooled column (modified version of the injector[103]) ensured the best results—the relative standard deviation in the determination of the peak area did not exceed 1–3%. However, the losses calculated taking into account the sensitivity of the flame-ionisation detector ranged from 3 to 10%. When the injection technique without stream splitting, described by Grob and Grob,[123] was used with evaporation of the sample at 400 °C, the losses of the test substance were even greater (14–20%) and the reproducibility of the analyses was to within 9–13%. The usual injection method with stream splitting yielded unsatisfactory results. The losses of the test substance increased to 25–50% and the error in the reproducibility rose to 15–30%. The causes of the errors in the method involving direct injection onto a cooled column have been examined.[104,124] Attention has been concentrated on the stage involving the transfer of the liquid from the syringe needle onto the inner surface of the CC wall. It is emphasised that this must be a purely mechanical process without evaporation. The partial loss of the components is ruled out when the syringe plunger is moved rapidly (several metres per second). Injection of 1 μl samples (from a 10 μl syringe) showed that appreciable losses of C_{20} and C_{28} n-alkanes (in hexane and pentane) are observed for an injection time reaching 1 s. Use of 0.5 μl syringes proved unsuccessful. Direct injection onto a cooled column permits an extensive variation of the sample volumes. The upper limit is determined by the risk of washing off the stationary phase and the lower by the possibility of the incomplete transfer of the sample from the syringe. An increase of column temperature diminishes the extent of the washing off of the stationary phase but, as the t_b of the solvent is approached, it induces the reverse flow of the test substance from the CC. In order to avoid contamination of the column by the stationary phase, the phase is first removed from a large section of the column. A reasonable maximum dose for CC is 10 μl. When pentane and an initial temperature of 30 °C were used, a relative standard deviation of 0.3–1.5% was obtained without an appreciable loss of the component. The minimum permissible dose was 0.2 μl and satisfactory results were obtained with large 5 and 10 μl syringes. When the column temperature (140 °C) exceeds the t_b of the sovent (hexane), the sample is appreciably condensed on the outer walls of the needle even under the conditions of rapid injection (the losses reached 20%); if the time during which the needle remains in the injector is 5 s, then the losses are doubled. The possible operating conditions involve a temperature of the CC slightly exceeding t_b—by 7 °C for pentane and hexane.

The test of the operation of the usual injectors with heated septums showed that, when syringes (with any injection technique) and systems in which the sample is evaporated are used, it is impossible to avoid a partial loss of the components (C_{20} and higher alkanes).[125] The only reasonable alternative involves direct injection onto a cooled column.

Galli and Trestianu[104] paid particular attention to the selection of operating conditions preventing the "ejection" of part of the sample from the CC in the direction opposite to that of the flow. Knauss et al.[126] considered the

mechanism of the distribution of liquid along the initial section of the CC under the conditions of a temperature gradient. Badings et al.[127] and Chaudhan and Darbre[128] described designs for glass direct injection devices. A comparative study has been made[129] of various types of injections: direct injection with a crucible—the "macro"-version (Fig.14); direct injection using a syringe and an injector (Fig.15) with additional cooling; injection with stream splitting and injection without stream splitting using a standard injector (Fig.13). Both direct injection methods yield satisfactory results. The version with the crucible is technically simpler, but leads to a greater error—the relative standard deviations in the determination of the area are respectively 2.5 and 0.7. When these methods were used to determine the sensitivity coefficient of methyl octanoate (in relation to n-dodecane) the relative standard deviations were 0.25 and 0.14% respectively. Schomburg et al.[130] estimated the level of losses of C_{10}–C_{40} alkanes when a stream splitter was used. The application of a dodecane instead of an octane sample as the solvent and reduction of the evaporator temperature from 310° to 210 °C made it possible to increase the C_{40}/C_{13} peak area ratio from 0.13 to 1.02.

Comparison of the direct injection methods with and without stream splitting demonstrated a statistically significant difference between the reproducibilities and accuracies of the analysis of 10 aliphatic acids (from C_{16} to C_{22}) in rape seed oil.[131] The relative error was on average 2.3% for the direct method and 6.4 and 6.0 respectively for injection without and with stream splitting.

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A review of studies on injection methods for the determination of impurities shows that the mechanism of the processes occurring is in fact related to the new separation method introduced by the present author,[1-9,15-51] namely chromadistillation (CD), which takes place along the initial section of the column. All the existing methodological versions of CD have found an application: isothermal elution, limiting chromatography, CD, the chromatography of nearly saturated vapours, and thermal CD.

Analysis of a fairly large number of publications on the "solvent effect" has shown that their authors actually arrived at the conclusion that the CD principles must be employed. Studies on the SE constitute an important step forward in the field of injection. It has become possible to introduce samples up to 10 μl into capillary columns, whereby the accuracy of the results has been improved. A further significant (by two orders of magnitude) increase of the sample injected (and hence of the sensitivity of the measurements) is possible as a result of the employment of special CD pre-columns or injectors. The CD theory helps in the formulation of the principal requirements ensuring the optimum operating conditions of the injection system: the sample must be introduced directly (without preliminary evaporation) into the column under the same conditions as the mixture to be analysed and the injecting syringe, and the site of injection must have a sufficiently large volume (0.2–1 ml) to permit the distribution of the sample; along the sample distribution section, it is essential to create conditions for the repeated condensation of the mixture components—a temperature gradient must be set up (with a decrease of temperature from the closest possible value to the t_b of the solvent) or an additional solvent must be introduced. The above description makes it possible to improve the design of the injector for capillary and packed columns.

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