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Adsorptive removal of Methylene blue and Methyl orange from aqueous media by carboxylated diaminoethane sporopollenin: On the usability of an aminocarboxilic acid functionality-bearing solid-stationary phase in column techniques

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Abstract

The adsorption phenomena of Methylene blue (MB) and Methyl orange (MO) on a carboxylated diaminoethane sporopollenin (CDAE-S) solid phase were investigated in a column arrangement by using breakthrough technique. The adsorption phenomena were evaluated using some common adsorption isotherm models and Scatchard plot analysis, and obtained results were interpreted for evaluating the usability of CDAE-S for removal, recovery and preconcentration of the studied dyes both at the laboratory and industrial scales. On the basis of Scatchard plot analysis, the interaction types between the CDAE-S and the studied dyes were criticized in terms of affinity phenomena. Thus, the usability of a biomacromolecule-derived material, CDAE-S, as a cheap, environmentally-friendly and effective solid-stationary phase exhibiting both cation-exchange and anion-exchange characteristics at the same time, is discussed through the present study. Besides, from the obtained results, the protonated CDAE-S, which functionally resembles an amino acid structure, are presented as a two-in-one solid-stationary phase, and its adaptability to common processes performed under column conditions is also drawn in detail.

Keywords: Breakthrough curve; Dye; HPLC; Ion-exchange; Scatchard plot analysis; Sorption; SPE

1. Introduction

The colored materials and dyes pollute the environment, being released as effluents through versatile industrial processes such as textile, food and plastic. The nonbiodegradable nature of most dyes in the environment makes their removal from effluents necessary. Various physicochemical processes/methods are used for removal of dyes from aqueous wastes and among them the adsorption is one of the most effective one [1–4].

The studies involve the investigation of novel materials (adsorbents) to be used in adsorption process is promising, and this is, surely, an esteemed struggle. As known, there exist a great deal of materials being used in adsorption processes for removal, recovery and pre concentration of target compounds. However, the adsorbents suitable for particular operations are more general than that exhibit almost universal character for every purpose [5].

Activated carbon has been widely used as an adsorbent for the removal of various pollutants due to its high adsorption capacity. However, it exhibits relatively high operation costs, regeneration trouble, and it is difficult to separate the treated activated carbon from the bulk solutions after use. Usually, the effectiveness of any adsorption process largely depends on the physicochemical properties of the adsorbent used. Thus, the regerenability, availability and operational cost are becoming of prime importance aspects in adsorption process.

The literature search indicated that the adsorption behaviors of dyes on different adsorbents had been investigated by using especially batch method [6–12]. In fact, from the industrial point of view, processing the removal of dyes and/or colored compounds by using the adsorption techniques carried out in column

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systems is thought to be very useful because of their adaptability to versatile processes, low reagent handling, and accordingly low cost coming out.

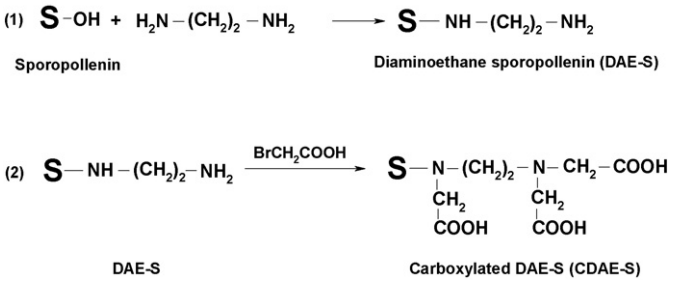
Sporopollenin is a natural biomacromolecule obtained from *Lycopodium clavatum*, which is highly resistant to chemicals, and has a high-capacity, stable, constant chemical structure; exhibits molecular structure stable to mineral acids and alkalines. The sporopollenin exhibits interaction ability with various chemical species, and occurs naturally as a component of spore walls [13–15]. The effectiveness of sporopollenin in adsorption process can be improved by functionalizing the main network with functional groups suitable to adsorption of target compounds, and thus multiple interaction abilities can be gained in addition to simple dispersive interactions. On this account, the adsorption phenomena of various species in functionalized (modified) sporopollenin mediated fixed-bed column systems has been reported elsewhere [16–20]. The adsorption of some chlorinated anilines on Co(II)-loaded carboxylated diaminoethane sporopollenin (CDAE-S) had been previously studied, and applicability of Langmuir model had been proposed for adsorption of mentioned species [16]. Furthermore, the adsorption kinetics of some chlorinated anilines on Co(II)-loaded CDAE-S under the column conditions had been reported in detail [20]. However, according to our knowledge, the studies intensify on adsorption behavior of dyes on the CDAE-S are very few [21].

The present study deals with the potential usability of the CDAE-S as a solid phase for removal, recovery and preconcentration of both cationic and anionic dyes from aqueous media. In the study, a continuous column mode has been employed, and the recorded breakthrough profiles have been utilized to derive equilibrium adsorption data. The adsorption characteristics have been discussed in view of Langmuir and Freundlich isotherm models, and the affinity phenomena have been evaluated by using Scatchard plot analysis. Finally, the interaction types between the studied dyes and CDAE-S have been comprehensively evaluated, and feasible application ranges of the CDAE-S as a solid-stationary phase have also been discussed in detail.

2. Experimental

2.1. Reagents

All solutions were prepared from reagent-grade purity chemicals without further purifications. Doubly-distilled water was used to prepare all solutions. Aqueous Methylene blue (MB) and Methyl orange (MO) solutions were prepared from their respective chloride and sodium salts (Merck), respectively, without further purifications. The concentrations of MB and MO solutions were within the ranges of 5.0×10^{-6} to 2.5×10^{-5} mol l^{-1} and 1.0×10^{-5} to 4.5×10^{-5} mol l^{-1}, respectively, and the solutions were studied without any pH adjustment. During the experimental course, an aqueous 0.5 mol l^{-1} HCl solution and methanol (Merck) were used as stripping agents. All the solutions were filtrated through 0.45 μm Nylon filter before use.



Scheme 1. The main steps for the modification of sporopollenin.

2.2. Modification of sporopollenin

Sporopollenin with a particle size of 20 μm, which was purchased from BDH Chemicals, was chosen as a support material for this study and converted into CDAE-S through the procedure described elsewhere [17]. The main steps of sporopollenin modification are pictured in Scheme 1.

2.3. Insolubility of CDAE-S

The solubility tendency and structural stability of the CDAE-S in aqueous media (dependent on pH) were investigated spectrophotometrically according to the method previously reported for characterization of (DAE-S) [22]. For this, a definite amount of the CDAE-S was added to a tube that included doubly-distilled water at a defined pH and agitated vigorously a few minutes. Then, the suspension was centrifuged at 8000 rpm for 5 min, and the supernatant and the (dried) solid phases were subjected to UV–vis and IR spectrophotometric analyses, respectively. The obtained IR spectrums of the treated CDAE-S were compared with the spectrum of the untreated CDAE-S to check if there was any structural changing. The UV–vis spectrums were recorded within the range of 800–200 nm and compared with the spectrum of doubly-distilled water to check if there was any changing in the spectrum that can be attributed to the adsorbent dissolution. The UV–vis and IR spectroscopy analyses were recorded on a Shimadzu Model 160 A UV–vis and a JASCO FT/IR 300 E (as KBr pellets) spectrophotometers, respectively.

2.4. Apparatus and methods

All materials were analytical grade. A glass mini column of 12 mm in length and 4 mm internal diameter with Teflon fittings was used. Firstly, the empty column was washed thoroughly with appropriate chemicals to remove all impurities adsorbed to the internal surface of the column. Packing of the CDAE-S to the column was done as per the slurry method, so the CDAE-S was filled to the column as its aqueous suspension, and by means of gravity the solid CDAE-S was packed to the column entirely. The height and volume of the bed embedded in water were 10 mm and 0.13 ml, respectively. Then, the column was conditioned for 48 h by pumping doubly-distilled water via an Alitea S2 peristaltic pump. Dye solutions were loaded to the column at average flow-rates of 0.21 ml min^{-1}

(∼97 BV h^{-1}) and 0.65 ml min^{-1} (∼300 BV h^{-1}) for MB and MO, respectively, and the effluents were passed through a continuous flow-through cell adapted Shimadzu Model 160 A UV–vis spectrophotometer, and monitored continuously. The loading breakthrough profiles recorded on the detector were used in evaluations and quantifications. All experimental courses were performed at a constant temperature, 25 °C. The limit of detection, LOD, for MB (657 nm) and MO (468 nm) was found as 7.6×10^{-7} mol l^{-1} and 3.1×10^{-6} mol l^{-1}, respectively.

The solid-phase extraction (SPE) operations were performed according to the methodology reported in [21,22]. The following summarizes the employed methodology:

i. Conditioning: this step includes subsequent pumping of (i) doubly-distilled water, (ii) aqueous 0.5 mol l^{-1} HCl solution, and (iii) doubly-distilled water through the column. Within 30 min time period, the conditioning of the column was reached when a stable base-line was recorded on the detector.
ii. Loading: dye solution was loaded to the column and at a defined wavelength (MB, 657 nm; MO, 468 nm) the efflux was monitored by the detector. The plot of detector response versus time yielded an S-shaped profile that is called breakthrough curve. When a particular breakthrough profiles being recorded on the detector has reached a maximum plateau, the loading step was terminated. The recorded breakthrough profiles were used to derive amount of the analyte adsorbed (q) by the CDAE-S.
iii. Washing: unsorbed dye molecules were removed from the column by pumping of doubly-distilled water until a stable and minimum signal was recorded on the detector, and then the washing step was terminated.
iv. Stripping–regenerating: the stripping–regenerating operations were executed according to the following methodology:
   – pumping an aqueous 0.5 mol l^{-1} HCl solution until end of the peak recorded on the detector (∼20 min);
   – pumping doubly-distilled water (∼10 min);
   – pumping 99% methanol until end of the peak recorded on the detector (∼20 min);
   – executing the conditioning step i (pumping doubly-distilled water–0.5 mol l^{-1} HCl–doubly-distilled water; ∼30 min).

In this way, most of the SPE steps were monitored continuously, and the solid phase (CDAE-S) was done prepared for loading step of subsequent SPE run.

2.5. Reproducibility of the breakthrough curve data

To evaluate the precision with the data, further replicate runs were performed as described in [21]. Through a statistical evaluation, the precision between days was found as 7.1% (R.S.D.).

2.6. Adsorption studies

The adsorption behaviors of MB and MO were investigated in a continuous column mode where the breakthrough technique was utilized as deriving equilibrium adsorption data. In the trials, adsorption behavior of a cationic (MB) and an anionic (MO) dye was studied. Because one of the main objectives of the study was to investigate inherent adsorption of the studied dyes on the CDAE-S, the pH of the influents was not adjusted. Therefore, the unique studied parameter was the influent concentration (C_0) that ranged between 5.0×10^{-6} and 2.5×10^{-5} mol l^{-1} and 1.0×10^{-5} and 4.5×10^{-5} mol l^{-1} for MB and MO, respectively. From the obtained breakthrough profiles, amount of analyte adsorbed (q) was calculated using breakthrough technique. The data obtained through breakthrough technique were used in evaluating adsorption phenomena and usability of the CDAE-S in common column operations as a solid-stationary phase. Also, it was observed that through the employed column technique it was possible to obtain recoveries higher than 97% for each studied dye. This may indicate suitability of the CDAE-S in adsorption process of the studied dyes as a reusable adsorbent. The reusability–regenerability of the CDAE-S was also confirmed by supplementary IR spectroscopic analyses.

3. Results and discussion

3.1. Insolubility of CDAE-S

In investigating the adsorption phenomena at liquid/solid interfaces, the adsorbents having an insoluble character (under studied experimental conditions) are sought for. Because of this, the insolubility characteristics of the CDAE-S were investigated for different medium pH values. The trials were executed in aqueous media, and the results of both UV–vis and IR spectroscopy analyses revealed that the CDAE-S was insoluble between pHs 0.0 and 14.0. Such a wide pH range makes the CDAE-S a very useful material for various processes to be executed in aqueous media. Thus, it was deduced that there was no hesitation of adsorbent dissolution under the studied experimental conditions.

3.2. Adsorption studies

The aqueous wastes being released through some industrial applications contain dyes, and nonbiodegradable nature of this kind of compounds necessitates their removal and/or recovery before the release of related industrial wastes to the environment. As achieving this target, the adsorption process can provide an inexpensive manner, and actually the effectiveness of this process is usually dependent on the properties of adsorbent used. In this sense, the present study aimed at discussing the adsorption phenomena of two common dyes, MB and MO, on a sporopollenin-derived material, CDAE-S. The molecular formula and some physicochemical properties of MB and MO are given in Table 1.

In order to evaluate the adsorption phenomena of MB and MO on the CDAE-S, the effect of loading solution concentration (influent concentration) was investigated for single analyte solutions. Amount of the analyte adsorbed at the equilibrium (q) has been calculated by using breakthrough technique, which is

Table 1
Some physicochemical properties of MB and MO[a]

Physicochemical property	Methylene blue	Methyl orange
Chemical formula	$C_{16}H_{18}ClN_3S{\cdot}xH_2O$ ($x=2-3$)	$C_{14}H_{14}N_3NaO_3S$
Solubility in water	50 g/l (20 °C)	5 g/l (20 °C)
pH value	~3.0 (10 g/l, H_2O, 20 °C)	~6.5 (5 g/l, H_2O, 20 °C)

[a] The data were obtained from the Merck chemical databases.

formulated as follows:

$$q = \frac{C_0 V}{W} \quad \frac{A_U}{A_U + A_D} \tag{1}$$

where C_0 denotes the influent concentration for the analyte in the aqueous phase, V is the volume of loaded analyte solution, and W is the dry mass of the adsorbent. The areas (A_U and A_D) on the upper and subjacent of the breakthrough curves are proportional to the amounts of adsorbed and released analyte, respectively [18,21,22].

The q values obtained through breakthrough technique are graphically pictured in Fig. 1 a and b, and it is clear from the figures that for each studied dye, amount of the analyte adsorbed

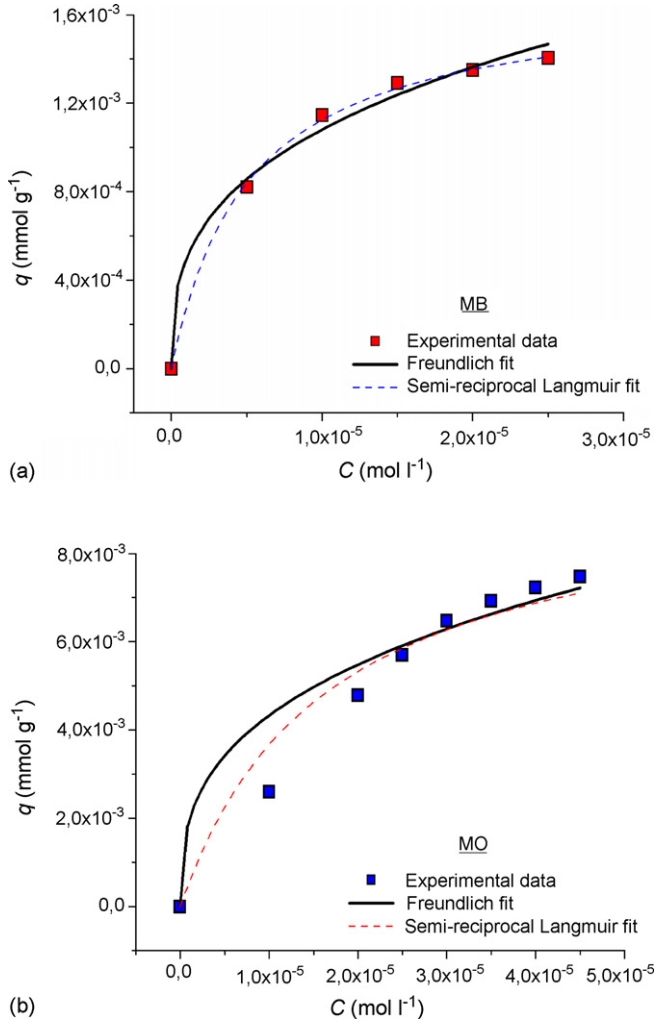


Fig. 1. (a) Binding curves for MB; (b) binding curves for MO.

by the CDAE-S increases with increase in influent concentration. This situation is attributed to the increased concentration gradient between fluid and solid phases with increasing influent concentration. Furthermore, from the statistical point of view, it is reasonable to say that the number of analyte molecules, which are available for functional groups of the CDAE-S, and accordingly the probability of analyte binding increases with increasing influent concentration.

3.3. Adsorption isotherms

The characteristic of a particular adsorption process is usually evaluated in terms of adsorption isotherms, and among the models being employed, those conceptualized by Langmuir [23] and Freundlich [24] are two most widely used. The monolayer Langmuir and the empirical Freundlich isotherms are usually represented in the following linearized forms:

$$\frac{C}{q} = \frac{1}{q_m K_b} + \frac{C}{q_m} \quad \text{(semi-reciprocal transformation of the classical Langmuir equation)} \tag{2}$$

$$\ln q = \ln k + \frac{1}{n} \ln C \quad \text{(a linearized form of the Freudlich isotherm equation)} \tag{3}$$

where q and C are the equilibrium analyte adsorption capacity of adsorbent and the equilibrium analyte concentration in the aqueous solution, respectively; q_m, K_b, k, and n are the adsorption isotherm parameters. From the slopes and intercepts of the C/q versus C and $\ln q$ versus $\ln C$ plots, the isotherm parameters can easily be calculated.

The Fig. 1a and b comparatively represent both (i) experimentally obtained and (ii) model-predicted binding data for MB and MO, respectively, and the isotherm parameters calculated from the applied models are listed in Table 2.

The fit of a model to the experimental data are usually evaluated in terms of linear regression analysis where the r^2 value is used as an indication for the goodness of model fit. With respect to r^2 values (Table 2), the adsorption of MO on the CDAE-S can be evaluated as a process that mainly follows the Freundlich model. Considering the adsorption of MB on the CDAE-S, the Langmuir model seems representing the equilibrium adsorption data better with r^2 value of 0.999.

In order to achieve more comprehensive information about the affinity of binding sites toward analyte and to analyze the results of employed adsorption isotherms, the equilibrium adsorption data were analyzed in terms of the Scatchard plot analysis.

3.4. Scatchard plot analysis

As evaluating the nature of a particular adsorption process suitably, the equilibrium adsorption data are represented in various graphical manners by employing the coordinates related to q and C. Hence, several mathematical transformations of the classical Langmuir equation are reported in the literature and

Table 2
Some parameters calculated from the adsorption isotherms and from Scatchard plots[a]

Analyte	Freundlich fit		Semi-reciprocal Langmuir fit		Scatchard plot analysis	
	k	n	q_m (μmol g^{-1})	K_b (l mol^{-1})	q_m (μmol g^{-1})	K_b (l mol^{-1})
MB	0.051 $r^2 = 0.952$	2.989	1.70 $r^2 = 0.999$	199121.62	1.74 $r^2 = 0.986$	182867.79
MO	11.436 $r^2 = 0.972$	1.381	16.09 $r^2 = 0.944$	20779.65	H; 10.88 L; 26.12	H; 49388.33 L; 11104.31

[a] H and L represent the "high-affinity" and "low-affinity" binding sites, respectively.

the Scatchard [25] transformation is one of them. The technique based on the Scatchard equation Eq. (4) is called "Scatchard plot analysis" and as described in the literature [21,22,26–30], it offers an intuition about the affinity phenomena in a simple manner. The Scatchard equation is represented as follows:

$$\frac{q}{C} = q_m K_b - qK_b \quad (4)$$

where q and C are the equilibrium analyte adsorption capacity of the adsorbent and the equilibrium analyte concentration in the aqueous solution, respectively, and q_m and K_b are the adsorption isotherm parameters. The terms q_m and K_b are respectively related to number of binding sites and affinity of the adsorbent toward analyte.

The shape of a Scatchard plot is related to the number of interaction types between analyte and adsorbent. The presence of a deviation from linearity on a plot based on Scatchard analysis usually points out the presence of more than one type of binding sites, whilst the linearity of the Scatchard plot indicates that the binding sites are identical and independent. So, if the Scatchard plot is linear with a negative slope, it is related to interaction between the analyte and the binding sites that follows the Langmuir model [16]. Namely, compared to other mathematical transformation of the classical Langmuir equation, more informative intuitions about the affinity of binding sites toward analyte can be acquired from a Scatchard plot.

The Scatchard plots derived for MB and MO are given in Fig. 2a and b, and from the shape of the plots it can deduced that in the adsorption of MB on the CDAE-S, the binding sites exhibit the same affinity toward MB, whereas the adsorption of MO mainly realizes on two types of the binding sites having different affinities toward MO. Because MB is a cationic dye, its adsorption on the CDAE-S is mainly attributed to the carboxyl groups, and on the basis of Scatchard plot (Fig. 2a) it can be said that the adsorption of MB on the CDAE-S exhibits a Langmuirean-type character for the studied concentration range (0.5×10^{-5} to 2.5×10^{-5} mol l^{-1}). However, the Scatchard plot derived for MO (Fig. 2b) exhibits a curvilinear shape for whole studied concentration range (1.0×10^{-5} to 4.5×10^{-5} mol l^{-1}), and this may indicate a deviation in the adsorption character from the Langmuirean behavior for the concentration range of 1.0×10^{-5} to 4.5×10^{-5} mol l^{-1}. Hence, it is possible to suggest that for the given concentration range the equilibrium adsorption data of MO does not show a good fit to the Langmuir model. On the other hand, when the Fig. 2b is investigated well, it can be seen that the data points form two separate arrangements that yields two separate linear combinations. The linear combinations of the data points (Fig. 2b, dashed lines) can be attributed to two different populations of the binding sites that exhibit different affinities toward MO. Therefore, the adsorption of MO on the CDAE-S fits to the Langmuirean behavior for the individual concentration ranges of (i) 1.0×10^{-5} to 3.0×10^{-5} mol l^{-1} and (ii) 3.0×10^{-5} to 4.5×10^{-5} mol l^{-1}. Considering the case (i), the binding sites exhibit relatively low affinity but high capacity toward MO, and so the binding sites taking role in the adsorp-

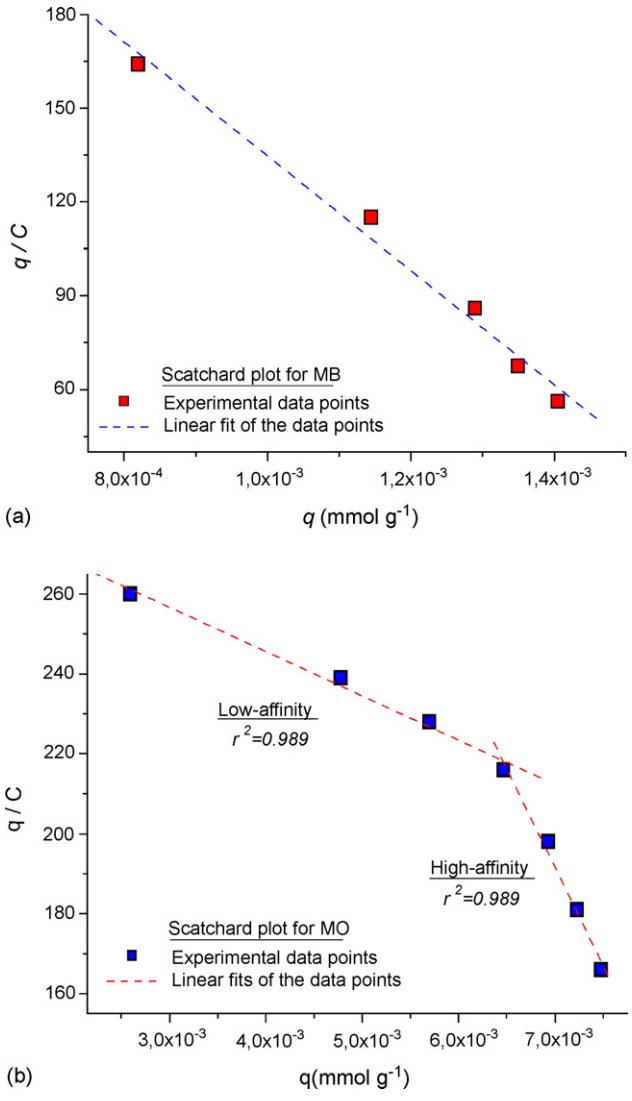


Fig. 2. (a) Scatchard plot for MB; (b) scatchard plot for MO.

tion of MO can be defined as "low-affinity binding sites (L)". In the case (ii), the adsorption of MO on the CDAE-S mainly realizes on the binding sites exhibiting relatively high affinity but low capacity toward MO, and so this type of the binding sites has been defined as "high-affinity binding sites (H)". Conclusively, dependent on the influent concentration, the affinity of the binding sites toward MO can vary and this property can be useful in design of effective strategies in adsorption process of MO by the CDAE-S. It should be noticed that the adsorption of MO (an anionic dye) on the CDAE-S is mainly attributed to the protonated amino groups that exhibit anion-exchanger character.

Some parameters calculated from the Scatchard plots (affinity constant (K_b) and theoretical saturation capacity (q_m)) are tabulated in Table 2. Considering the q_m and K_b values calculated from the $C/q - C$ and Scatchard ($q/C - q$) plots of MB, it can be seen that the isotherm parameters are individually very close to each other, and this may reveal that the adsorption of MB on the CDAE-S is mainly characterized by the Langmuir model. Taking the plots of MO, the isotherm parameters calculated for proposed high-affinity binding sites are close to those calculated from the $C/q - C$ plot of MO and this may indicate that the adsorption of MO on the CDAE-S is a process mainly based on high-affinity binding sites rather than low-affinity binding sites.

3.5. Argumentation: innovative aspects

The breakthrough curves are very useful tools for monitoring the distribution of analyte concentration between fluid and solid phases. A breakthrough curve can give considerable information about adsorption, adsorption kinetics and chromatographic retention time phenomena [31–33]. Sometimes, the differentiation in the shape of breakthrough profiles can be useful for estimating the adsorption tendency of analytes on a same adsorbent, and so comparative breakthrough profiles of MB and MO are given in Fig. 3. It is well known that the breakthrough time is highly correlated with the chromatographic retention time of analytes on the same stationary phase [33]. Therefore, the chromatographic separation of MB and MO can be discussed on the basis of breakthrough profiles. In the light of this fact, the breakthrough profiles were analyzed, and the breakthrough volumes/times of MB and MO were found to be very close to each other for most cases. So, at first, the chromatographic separation of studied dyes on the CDAE-S had been thought to be difficult for given experimental conditions. However, after a collective evaluation of whole SPE process and the results of the Scatchard plot analysis, more reasonable inferences about the usability of the CDAE-S as a stationary phase in liquid chromatographic analyses of both anionic and cationic dyes were gathered.

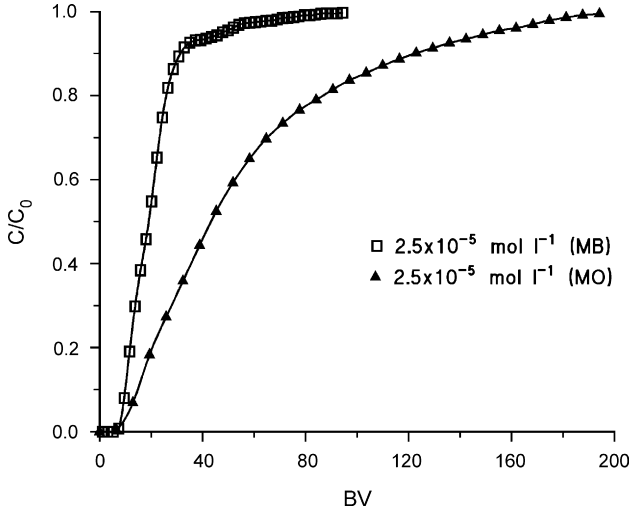


Fig. 3. Comparative breakthrough profiles for MB and MO. Influent concentration, C, 2.5×10^{-5} mol l^{-1}; for MB: flow rate, 0.21 ml min^{-1}; detection wavelength, 657 nm; LOD, 7.6×10^{-7} mol l^{-1}; for MO: flow rate, 0.65 ml min^{-1}; detection wavelength, 468 nm; LOD, 3.1×10^{-6} mol l^{-1}.

The Scatchard plot analysis showed that the CDAE-S could exhibit different affinities toward MB and MO, and the main interactions between the CDAE-S and the studied dyes were attributed to (i) carboxyl and (ii) protonated amino functionalities for MB and MO, respectively. The carboxyl groups of the CDAE-S are thought to have important role in the adsorption of MB (a cationic dye). However, in the case of MO (an anionic dye), the carboxyl groups are thought not to be attractive, and on the contrary to be repulsive toward anionic dyes because of their cation-exchanger nature. For this reason, the binding of MO on the CDAE-S are attributed to the remaining functionality of the CDAE-S, diaminoethane. It was observed that the highest observable capacities of the CDAE-S toward MB and MO are close to each other, and thus it is believed that there must be a close relation between the chemistry of the studied dyes and the functionalities (carboxyl and protonated amino groups) of the CDAE-S. In order to conceive this situation well, it will be beneficial to logically analyze the phenomena taking place in the column system during the SPE steps, especially during the conditioning step. Hence, the following part discusses the issue, "how may carboxyl and amino functionalities work in the adsorption of anionic and cationic species (?)".

The results showed that the protonated CDAE-S can interact with both anionic and cationic dyes at varying degrees. For the studied experimental conditions, the observable adsorption capacity of the CDAE-S for MB and MO was found to vary in the ranges of 0.82–1.41 μmol g^{-1} and 2.60–7.48 μmol g^{-1}, respectively. The observable capacities of the CDAE-S for MB (a cationic dye) and MO (an anionic dye) were found being close to each other, indicating the suitability of the CDAE-S as a solid phase both for anion- and cation-exchange processes at the same time. Hence, in the present study we tended to discuss the feasible application ranges of the CDAE-S as a solid-stationary phase in anion- and cation-exchange processes at the same time.

The multifunctional characteristics of the CDAE-S have previously been reported by Gezici et al. [21] where the adsorption behavior of Crystal violet (CV; a cationic dye) on the CDAE-S has been discussed in view of pH point of zero charge (pH_{pzc}) measurements. In the mentioned study, the surface charge characteristics of the DAE-S and CDAE-S solid phases are comprehensively discussed, and pH_{pzc} values are reported as 8.02 and 4.30, respectively. The carboxyl and amino functionalities have been observed to have important effect on the surface charge characteristics of the CDAE-S. The mentioned study suggests that between the medium pH values of 4.30 and

8.02, the HCl treated-CDAE-S exhibits a zwitterionic character that can make it possible to perform both anion- and cation-exchange processes on the same solid phase. Below the pH 4.30, the CDAE-S is believed to almost lose its cation-exchange ability, whereas its anion-exchange ability remains considerably high under that condition. Besides, above the pH 8.02, the CDAE-S almost losses its anion-exchanger character due to the dissociation of the protonated amino groups. It should be noticed that in order to take the benefit of the mentioned multifunctional characteristics of the CDAE-S, it is required to interact the CDAE-S with a suitable acidic aqueous solution (viz. 0.5 mol l^{-1} HCl) during the conditioning step. Because of this, in the present study, during the conditioning step, an aqueous 0.5 mol l^{-1} HCl solution was pumped through the column as converting the CDAE-S in a protonated form. Briefly, carboxyl and protonated amino groups are respectively believed to attract cationic and anionic species through ion-exchange mechanism. Therefore, the adsorption of MB and MO on the previously protonated CDAE-S is respectively attributed to carboxyl and amino functionalities of the CDAE-S. Due to the fact that the amino groups exhibit relatively weak basic character, they can be protonated in acidic media, and in a sense, can form binding sites that can attract anionic species through ion-exchange mechanism. In view of these approaches it can be suggested that the chromatographic separation of cationic and anionic species can be achieved on the CDAE-S by determining the mobile phase composition suitably. It seems reasonable to say that with increasing mobile phase pH, the previously protonated amino groups are gradually converted into their respective unprotonated forms, and accordingly the retention of MO on the CDAE-S diminishes, whilst that of MB rises. Hence, a selective separation of cationic dyes from anionic dyes can be achieved in the CDAE-S-mediated column systems by determining appropriate chromatographic conditions, and these approaches can be interpreted for potential usability of the CDAE-S as a stationary phase in routine liquid chromatographic analyses of dyes. As a result, by careful determination of mobile phase/loading solution pH several opportunities can be achieved in column operations while the CDAE-S is solid-stationary phase.

3.5.1. CDAE-S works as a cation-exchanger

Because the studied CDAE-S has a pH_{pzc} value of 4.30, it is reasonable to say that the CDAE-S can easily bind the cationic species through cation-exchange mechanism when medium pH is higher than 4.30. Besides, similar to ethylenediaminetetraacetic acid (EDTA), the CDAE-S is thought to exhibit different acidic dissociation constants (pK_a) between 3.0 and 10.0 in aqueous media, and because of this, the CDAE-S can act as a cation-exchanger and bind the cationic species through cation-exchange mechanism.

3.5.2. CDAE-S works as an anion-exchanger

The diaminoethane functionality of the CDAE-S is thought to be useful in anion-exchange process, if amino groups are protonated previously or simultaneously. As previously reported by Gezici et al. [21], the DAE-S has a pH_{pzc} value of 8.02. So, it can be said that when medium pH value is roughly below 8.02, the CDAE-S exhibits an anion-exchanger character via protonated amino groups. As mentioned before, unexpected high and strong adsorption of MO on the CDAE-S is attributed to the effect of protonated amino groups. Hence, from the experimental results it is deduced that the CDAE-S can also act as an anion-exchanger after being interacted with a strong acid solution (e.g. HCl). So, the medium pH is believed to be a very important experimental variable in controlling the effectiveness of separation processes where the CDAE-S is used as a solid-stationary phase. For example, it seems reasonable to say that with increasing medium pH, the dissociation of protonated amino groups and carboxyls increases and this favors the cation-exchange process rather than anion-exchange. In order to support these approaches by the experimental results and to well understand the effect of diaminoethane functionality, supplementary SPE experiments were carried out while DAE-S, the un-carboxylated form, was the solid phase. During the supplementary SPE experiments, all of the experimental parameters were fixed to those in the case of the CDAE-S, unless being stated otherwise, and the trials were performed in two manners:

- The stripping step was performed by pumping of 0.5 mol l^{-1} NH_3 aqueous solution (instead of 0.5 mol l^{-1} HCl) and followed by doubly-distilled water, methanol, and doubly-distilled water again. After the stripping step, the conditioning step was performed that involved pumping of (i) doubly-distilled water, (ii) aqueous 0.5 mol l^{-1} HCl solution, and followed by (iii) doubly-distilled water through the column. In this way, the DAE-S was converted into an anion-exchanger form through the protonation of amino group(s). Afterwards, the dye solutions were separately pumped to the column. As expected, amount of the MB (a cationic dye) adsorbed on the DAE-S was found to be considerably lower than that obtained on the CDAE-S, whereas amount of the MO (an anionic dye) adsorbed on the DAE-S was slightly higher than that obtained on the CDAE-S for most cases. The obtained results are compatible with theoretical approaches, and effect of the protonated diaminoethane functionality in the cases of the CDAE-S and DAE-S is clear. The slight increment observed in the amount of the MO adsorbed on the DAE-S in comparison to that on the CDAE-S is attributed to decrease in steric effects and elimination of repulsive effects may rise from carboxyl groups of the CADE-S which complicates a suitable binding of MO on protonated diaminoethane groups.
- After the stripping step (0.5 mol l^{-1} NH_3–doubly-distilled water–methanol–doubly-distilled water), the conditioning step was performed. The conditioning step consisted of pumping the (i) doubly-distilled water, (ii) aqueous 0.5 mol l^{-1} NH_3 solution (instead of 0.5 mol l^{-1} HCl), and followed by (iii) doubly-distilled water through the column. So, during the conditioning step, the DAE-S was not interacted with 0.5 mol l^{-1} HCl solution in order to well conceive the effect of amino groups on the unprotonated DAE-S. Hence, the single dye solutions were separately pumped through the column that involved the unprotonated DAE-S. The results revealed a considerably lower adsorption of each dye on the unprotonated

DAE-S for most cases in comparison to those on the CDAE-S and the protonated DAE-S, and the observed low bindings were attributed to the simple dispersive interactions between the DAE-S and dyes.

The obtained results are compatible with the results previously obtained on the CDAE-S [21] and DAE-S [22] for (i) CV and (ii) MB and MO, respectively. All the findings reveal the role of carboxyl and diaminoethane groups as well as experimental conditions in the adsorption of studied dyes, and support the usability of the CDAE-S as a two-in-one solid–stationary phase in common column operations of ionic and/or ionizable species.

4. Conclusion

As a result, the CDAE-S seems to be a very useful material in SPE and chromatographic separation of dyes as well as ionic and ionizable species, acting as a "two-in-one" solid–stationary phase. The aminocarboxilic acid functionality with the CDAE-S was found useful both in cation-exchange and anion-exchange processes at the same time. Such a feature can provide various application and design possibilities in column processes where the CDAE-S is used as a solid–stationary phase. Furthermore, in view of pH_{pzc} phenomenon, a number of combinations can be derived for SPE and chromatographic separations of ionic and ionizable chemical species on the CDAE-S by careful determination of medium pH. The adsorption isotherms revealed that the adsorption of MB on the CDAE-S mainly fitted to Langmuirean-type bindings, whereas the adsorption of MO on the CDAE-S was found followed mainly Freundlich model. Through the Scatchard plot analysis it was proposed that in the adsorption of MO on the CDAE-S, two binding types (high- and low-affinity bindings) took role, whilst the adsorption of MB on the CDAE-S was observed to be mainly based on a one-type binding for the studied experimental conditions. The obtained findings support the utility and multifunctionality of the CDAE-S in a variety of processes, and are believed to be improved through further investigations. This point of view can be an important step for development of novel solid-stationary phases, and selective interactions that similar to those between enzyme and substrate can be achieved between amino acids and the CDAE-S by evaluating the isoelectric point matter, comprehensively.

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References

[1] D. Ghosh, K.G. Bhattacharyya, Appl. Clay Sci. 20 (2002) 295.
[2] J. Avom, J.K. Mbadcam, C. Noubactep, P. Germain, Carbon 35 (1997) 365.
[3] N. Kannan, M.M. Sundaram, Dyes Pigments 51 (2001) 25.
[4] H.G. Linge, Fuel 68 (1989) 111.
[5] I. Liška, J. Chromatogr. A 885 (2000) 3.
[6] G. Atun, G. Hisarli, W.S. Sheldrick, M. Muhler, J. Colloid Interface Sci. 261 (2003) 32.
[7] A. Gürses, S. Karaca, Ç. Doğar, R. Bayrak, M. Açıkyıldız, M. Yalçın, J. Colloid Interface Sci. 269 (2004) 310.
[8] S. Senthilkumaar, P.R. Varadarajan, K. Porkodi, C.V. Subbhuraam, J. Colloid Interface Sci. 284 (2005) 78.
[9] K.G. Bhattacharyya, A. Sharma, Dyes Pigments 65 (2005) 51.
[10] S. Chakrabarti, B.K. Dutta, J. Colloid Interface Sci. 286 (2005) 807.
[11] Y.-L. Ma, Z.-R. Xu, T. Guo, P. You, J. Colloid Interface Sci. 280 (2004) 283.
[12] C.-H. Weng, Y.-F. Pan, Colloids Surf. A 274 (2006) 154.
[13] G. Shaw, in: J.B. Harbone (Ed.), Sporopollenin in Phyto Chemical Phylogency, Academic Press, London, 1970 (Chapter 3).
[14] J. Brooks, G. Shaw, Nature 220 (1968) 678.
[15] H. Bubert, J. Lambert, S. Steuernagel, F. Ahlers, R. Wiermann, Z. Naturforsch 57c (2002) 1035.
[16] M. Uçan, A. Ayar, Colloids Surf. A 207 (2002) 41.
[17] A. Ayar, S. Yildiz, E. Pehlivan, Sep. Sci. Technol. 30 (1995) 3081.
[18] A. Ayar, S. Yildiz, Colloids Surf. A 201 (2002) 291.
[19] A. Ayar, A.I. Pekacar, B. Mercimek, Colloids Surf. A 212 (2003) 51.
[20] M. Uçan, A. Gürten, A. Ayar, Colloids Surf. A 219 (2003) 193.
[21] O. Gezici, M. Küçükosmanoğlu, A. Ayar, J. Colloid Interface Sci. 304 (2006) 307.
[22] M. Küçükosmanoğlu, O. Gezici, A. Ayar, Sep. Purif. Technol. 52 (2006) 280.
[23] I. Langmuir, J. Am. Chem. Soc. 40 (1918) 1361.
[24] N. Freundlich, Colloid and Capillary Chemistry, Methven, London, 1926.
[25] G. Scatchard, Ann. N.Y. Acad. Sci. 51 (1949) 660.
[26] V.D. Noto, L.D. Via, P. Zatta, Coord. Chem. Rev. 228 (2002) 343.
[27] H. Chaouk, M.T.W. Hearn, J. Biochem. Biophys. Methods 39 (1999) 161.
[28] J. Zhou, X. He, Y. Li, Anal. Chim. Acta 394 (1999) 353.
[29] H. Chaouk, M.T.W. Hearn, J. Chromatogr. A 852 (1999) 105.
[30] J. Matsui, Y. Miyoshi, O. Doblhoff-Dier, T. Takeuchi, Anal. Chem. 67 (1995) 4404.
[31] O. Gezici, H. Kara, M. Ersöz, Y. Abali, J. Colloid Interface Sci. 292 (2005) 381.
[32] A.A. Gürten, S. Uçan, M.A. Özler, A. Ayar, J. Hazard. Mater. B120 (2005) 81.
[33] M.-C. Hennion, J. Chromatogr. A 856 (1999) 3.