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The *In Vitro* Adsorption study of Isoniazide and Pyrazinamide on Indigenous Bentonite

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In the research series on the interactions of some drugs with bentonites, having a special interest for the pharmaceutical science we studied the adsorbing capacity of revived and HCl activated bentonites from Valea Chioarului. The adsorbed drugs were isoniazide and pyrazinamide. The adsorbing capacity was expressed in the amount of drug adsorbed per unit mass of adsorbent.

The practical application of the experimental data was made using the liquid phase adsorption isotherm of Freundlich and of Langmuir.

The characteristic parameters of the bentonite–drug system were obtained processing the experimental data by regression analysis. The adsorption coefficient (a) and the adsorption capacity at the equilibrium concentration equivalent to unity (b) were determined. The spreading of the data show obedience to the Langmuir pattern.

The derivatographic study of the bentonite drug system allowed the calculation of the activation energie (E) and of the reaction order (n) characteristic parameters of the dehydration kinetics. For this purpose the differential method of Freeman and Carroll was used.

The electronic spectra of bentonite–drug system in KBr pellets, shows hypsochromic shifts of the 265 nm band characteristic for isoniazide and the 269 nm band characteristic for pyrazinamide.

The IR spectra for bentonite–drug system show significant changes in the 1700–1500 cm^{-1} range characteristic for $\nu_{C=O}$ and δ_{H_2O} modes.

TABLE

System		Freundlich Method	Langmuir Method	n	E (kcal/mol)
Bentonite–Isoniazide	a	0.69	0.298		
	b	0.0075	0.0019	0.7	10.67
	s^2	4.84×10^{-5}	0.87×10^{-5}		
Bentonite–Pyrazinamide	a	0.13	0.87		
	b	2.99	3.01	0.7	6.54
	s^2	0.041	5.39×10^{-5}		

The Nature of the Type 2 Copper Binding in Blue Oxidases

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Of the three different forms of copper usually contained in the blue oxidases, the Type 2 Cu is a divalent paramagnetic form characterized by a 'normal' EPR spectrum, similar to that of tetragonal copper complexes, with $A_{\parallel}$ of 150–200 gauss [1]. The Type 2 Cu site is open to solvent access and binds dissolved monovalent anions, such as azide, cyanide and fluoride [1]. These anions have an inhibitory effect on the oxidase activity much stronger than their affinity for the resting enzyme would predict. For tree laccase the affinity of the Type 2 Cu for azide increases on reduction of the Type 3 Cu [2], a likely event during turnover. All oxidases show in the presence of azide an intense absorption band at 400 nm and a shoulder at about 500 nm. The latter is never observed in mononuclear Cu(II)–azide complexes and is assigned to a charge-transfer from the bridging azide to the Cu(II) in the Cu(II)–N_3–Cu(I) group of azide-bound met-hemocyanine [3]. This suggests that a similar type of binding might be present in the oxidases. The EPR spectra of the Type 2 Cu(II)–N_3 and –CN adducts of tree laccase, measured under reducing conditions, are very similar to those of the corresponding complexes of the Cu(II) substituted carbonic anhydrase, where the Cu(II) is believed to be pentacoordinated in a distorted square pyramidal geometry [4]. They are characterized by low $A_{\parallel}$ in the range 106–137 gauss. The metal ion is bound to three hystidyl residues and one water molecule, besides the anion, in the Cu(II) carbonic anhydrase [4], to at least one water [1] and one hystidyl residue [5] in the tree laccase.

Selective removal of the Type 2 Cu abolishes the oxidase activity. In the tree laccase both the rate of