Microcalorimetric studies on the adsorption of DNA by soil colloidal particles

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Abstract

This study applied TAM air isothermal calorimeter to measure the adsorption enthalpies of DNA on eight colloidal fractions from permanent-charge and variable-charge soils. The adsorption of DNA on soil colloids was also examined by equilibrium adsorption analysis. The data evaluated from isotherms fitted by Langmuirean model revealed that the affinity of DNA for variable-charge soil colloids was higher than that for permanent-charge soil colloids. More tightly bound DNA molecules were observed on coarse clays and inorganic clays than on fine clays and organic clays, respectively. The adsorption enthalpies of DNA on permanent-charge soil colloids were higher than those on variable-charge soil colloids. DNA adsorption on organic clays is endothermic, whereas that on inorganic clays is exothermic. Dehydration and electrostatic repulsion were considered to cause the higher adsorption enthalpies of DNA with organic clays, while hydrogen bonding, ligand exchange and electrostatic attraction result in the lower DNA adsorption enthalpies on inorganic clays. The thermodynamic parameters presented in this study have important implication for providing further insight into mechanisms of the adsorption of DNA on soil particles.

1. Introduction

DNA is the genetic material of organisms. In soil environments, DNA molecules are liberated by excretion from microorganisms, plants, and animals or by lysis of dying cells [1]. These extracellular DNA molecules can be adsorbed by clay minerals, sand, humic acids, montmorillonite-humic acids-aluminum or iron hydroxypolymers and soils and are partially protected against degradation by nuclease [2–8]. Moreover, these adsorbed DNA molecules have been shown to retain the capability to transform competent cells in the absence of a susceptible host [1–9]. Therefore, DNA molecules in soils have an important role in biological activity and diversity, and the adsorption of DNA on soil particles is also one of the key processes affecting the horizontal transfer of extracellular genes in soil ecosystems. Knowledge of the extent and mechanisms of DNA adsorption on soil particles would aid in our understanding the fate of DNA in the soil environment.

The mechanisms of DNA adsorption on soil active particles have been studied by several soil biologists over the past years. Adsorption of DNA on montmorillonite and kaolinite increased with the decrease of pH from 9.0 to 3.0 [2,10]. Cations such as Mg^{2+}, Na^+ and NH_4^+ can form bridges between phosphate groups of DNA molecules and the negatively charged sites of clays and sand [11–13]. The length and shape of DNA molecules play an important role in DNA interactions with soil particles [6,14]. X-ray diffraction analysis showed that the basal spacing of montmorillonite and kaolinite were not altered after the adsorption of DNA from *Bacillus subtilis*, indicating that this DNA did not significantly intercalate the clays. Scanning and transmission electron microscopy presented that the binding of DNA from *B. subtilis* was primarily on the edges of montmorillonite and kaolinite [15]. A fourier-transform infrared (FT-IR) analysis revealed that DNA from *Bacillus thuringiensis* subsp. *kurstaki* changes its conformation from B I-to-B II as a consequence of its adsorption on humic acids and that the sugar-phosphate backbone and the bases are involved [16].

Although some attempts have been made to elucidate the mechanism of DNA adsorption on soil active particles, the interactions between soil colloids and DNA are still not totally

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understood. The information relating to thermodynamics of DNA adsorption on soil colloids is quite lacking, particularly in terms of directly measured data. The previous microcalorimetric studies regarding interactions between proteins and the biomaterials such as immobilized metal ion affinity chromatography (IMAC), hydrophobic interaction chromatography (HIC), ion-exchange chromatography (IEC), and molecularly imprinting polymer (MIP) have been demonstrated as a useful way to discuss the binding mechanism, and the variation of enthalpy change values mainly resulted from the involvement of different interaction forces between proteins and adsorbents [17–19]. In addition, for the mechanism of DNA adsorption on soil particles, very little is known about the effects of soil types, organic matter and particle size of soil colloids on DNA adsorption. The main purpose of the present work was to study the mechanism of DNA adsorption on various soil colloids by the adsorption enthalpy measurement. Discussion is also made on the difference of enthalpy change of DNA with different soil clays. The thermodynamic data presented in this study can provide a thorough understanding on the adsorption mechanism between DNA molecules and soil colloidal particles.

2. Materials and methods

2.1. DNA

Salmon sperm DNA was purchased from Sigma Chemical Co., St. Louis, MO.

2.2. Soil colloids

Brown soil (Alfisol) was sampled from the 0–17 cm layer of a forest land in Tianwai village, Taishan, Shandong province, China. Red soil (Ultisol) was collected from the 11–40 cm layer of a cultivated land in Xianning, Hubei province, China. The basic characteristics of soils are listed in Table 1. After removal of organic residue the soil was rinsed in deionized distilled water (ddH$_2$O) and dispersed by adding 0.01 M NaOH solution drop-

Table 1
Basic properties of the soils tested

Soil	Texture	pH (H$_2$O)	OM (g kg^{-1})	CEC (cmol kg^{-1})
Brown soil	Clay loam	6.36	42.40	15.30
Red soil	Clay	5.24	7.21	7.36

OM and CEC stand for organic matter and cation exchange capacity, respectively.

wise to pH 7–8 together with sonication. Two types of soil colloidal components i.e. fine clay (<0.2 μm) and coarse clay (0.2–2 μm) were separated by sedimentation. Part of fine and coarse clays was oxidized by H$_2$O$_2$ to remove organic matter. After flocculation by the addition of CaCl$_2$ solution, the colloidal suspension was washed to be free of Cl$^-$ ions by ddH$_2$O and ethanol, and then air-dried. Soil colloidal particles prepared were ground to pass a 100 mesh sieve (150 μm). Some properties of the clay fractions are listed in Tables 2 and 3.

2.3. Characterization of soil colloids

Texture and pH of Brown soil and Red soil were measured by pipette and potential meter (H$_2$O: soil = 2.5:1). Organic matter (OM) and cation exchange capacity (CEC) were determined by K$_2$Cr$_2$O$_7$ digestion and extraction with NH$_4$C$_2$H$_3$O$_2$, respectively [20]. The content of clay mineral was determined by X-ray diffraction analysis. Specific surface area (SSA) was measured by the method of N$_2$ adsorption (Beijing Analytical Instrument Company).

2.4. Equilibrium adsorption isotherms

Two grams of soil colloid was suspended with suitable volumes of ddH$_2$O in conical flask and adjusted to pH 7.0 for Brown soil colloids and 5.5 for Red soil colloids using 0.1 mM HCl or NaOH. After equilibrium, the suspension was diluted to 100 ml with ddH$_2$O. In a centrifuge tube 0.5 ml of soil colloid suspension was mixed with 2.0 ml of buffer containing 25–350 μg of DNA. The adsorption of DNA was conducted in 10 mM Tris

Table 2
Selected properties of Brown soil colloids

Brown soil colloid	OM (g kg^{-1})	SSA (m^2 g^{-1})	CEC (cmol kg^{-1})	Clay mineral composition
Coarse organic clay	67.6	31.7	46.2	Hydromica (45%), vermiculite (35%), kaolinite (20%)
Coarse inorganic clay	5.9	37.5	41.2	Hydromica (45%), vermiculite (35%), kaolinite (20%)
Fine organic clay	74.4	97.1	68.9	Hydromica (60%), vermiculite (30%), kaolinite (10%)
Fine inorganic clay	13.3	116.5	54.5	Hydromica (60%), vermiculite (30%), kaolinite (10%)

OM, SSA and CEC refer to organic matter, specific surface area and cation exchange capacity, respectively.

Table 3
Selected properties of Red soil colloids studied

Red soil colloid	OM (g kg^{-1})	SSA (m^2 g^{-1})	CEC (cmol kg^{-1})	Clay mineral composition
Coarse organic clay	28.4	48.5	21.2	Hydromica (20%), 1.4 nm mineral (35%), kaolinite (45%)
Coarse inorganic clay	7.1	52.7	17.5	Hydromica (20%), 1.4 nm mineral (35%), kaolinite (45%)
Fine organic clay	32.2	146.2	35.6	Hydromica (45%), 1.4 nm mineral (25%), kaolinite (30%)
Fine inorganic clay	9.7	168.3	28.4	Hydromica (45%), 1.4 nm mineral (25%), kaolinite (30%)

OM, SSA and CEC represent organic matter, specific surface area and cation exchange capacity, respectively.