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# Nuclear Magnetic Resonance Studies of Potential Environmental Contaminants Interacting with DNA

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## Introduction

Several studies have shown that certain sunscreen ingredients, some hair dyes, and plastic nanoparticles are environmentally harmful to the ecosystem. Our research in NMR explains how these environmentally damaging molecules interact with DNA. We used Nuclear Overhauser Effect Spectroscopy (NOESY) NMR and molecular mechanics calculations to determine the binding geometry between several molecules and DNA. Similar methods have been used by others to study molecules interacting with DNA [1].

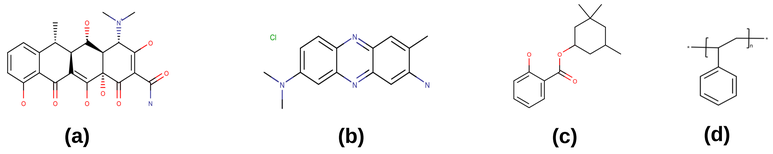



***Figure 1.*** Molecules considered in this study (a) doxycycline (b) homosalate (c) neutral red (d) polystyrene.

## Methods

- DNA (Drew-Dickerson dodecamer with sequence CGCGAATTCGCG) was purchased from Sigma-Aldrich and Integrated DNA Technologies.
- Samples were prepared in 200 mM phosphate buffer, pH 7
- All NMR measurements were made on a Bruker NEO 500 MHz spectrometer with Prodigy cryoprobe at 298K. NOESY spectra were collected with a mixing time of 0.4s, 32 scans, 128 t1-increments, recycle delay of 1.5 s, and spectral width of 10 ppm. Spectra were assigned using the reference [2].
- Energy minimizations were done using the molecular mechanics (MM) methods in Gaussian 16 [3]. The UFF force field [4] was used, atom typing was done automatically, and charges were calculated using the QEq formalism [5]. Na+ counterions were added to make the overall charge on the DNA neutral, and partial single bonds were added to maintain hydrogen bonds across the DNA base pairs.

## Results

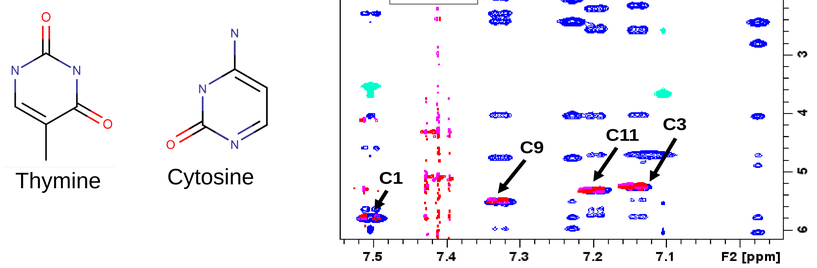



***Figure 2.*** NOESY (blue) and COSY (red) spectra of 2.5 mM DNA.

## DNA + Neutral Red

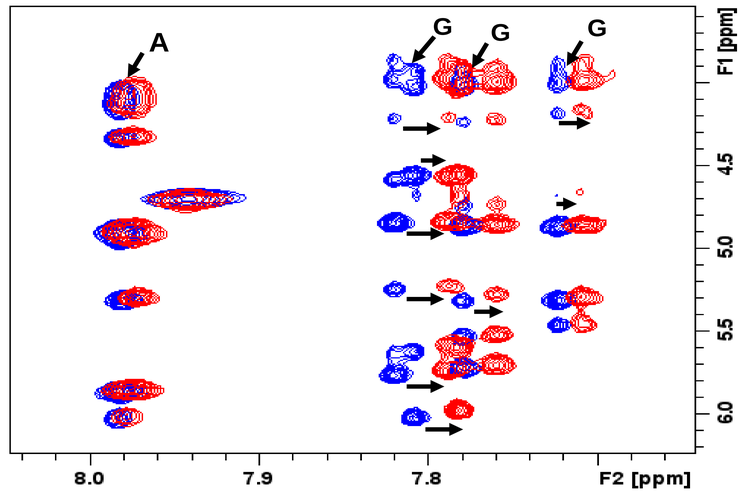
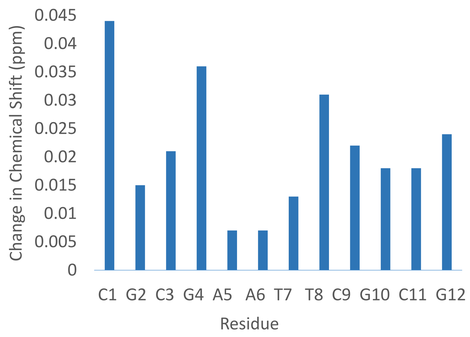



***Figure 3.*** Portions of NOESY spectra of 2.5 mM DNA (blue) and 2.5 mM DNA with 0.5 mM neutral red (red).

***Figure 4.*** Chemical shift changes of aromatic protons as a function of residue number for DNA interacting with neutral red.




## Other Molecules

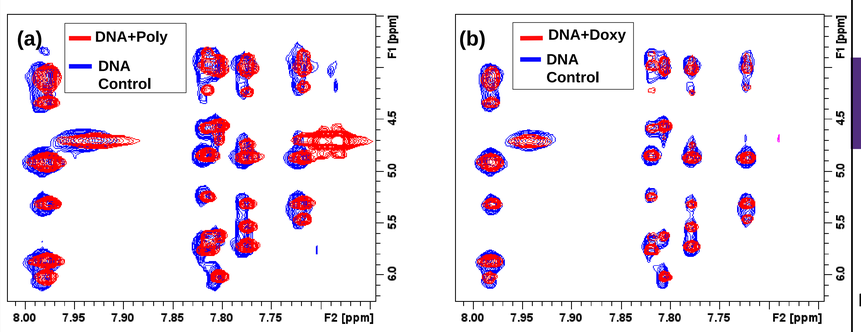



***Figure 5.*** (a) Portions of NOESY spectra of 2.5 mM DNA (blue) and 2.5 mM DNA with polystyrene nanoparticles (red) (b) Portions of NOESY spectra of 2.5 mM DNA (blue) and 2.5 mM DNA with 0.5 mM doxycycline hyclate (red).

## Computational Modeling

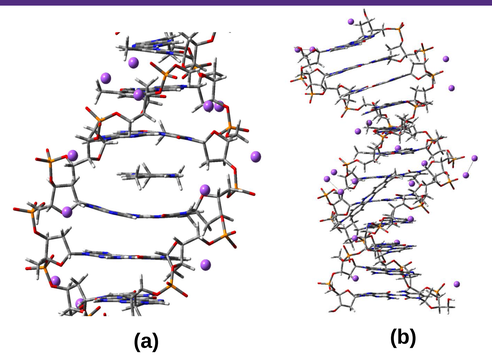



***Figure 6.*** Energy-minimized structures of neutral red interacting with DNA. (a) a representative "intercalating" structure (b) a representative "minor groove" structure. The modeling structures were based on the references [6].

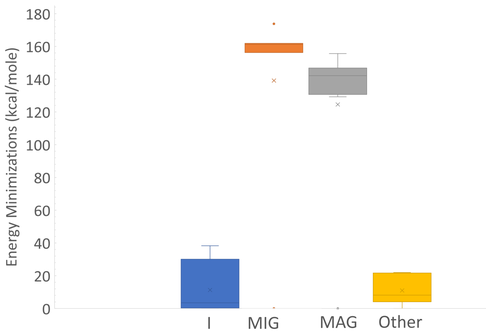



***Figure 7.*** Box-and-whiskers plot showing energy of low-energy structures. I = intercalated. MIG = minor groove. MAG = major groove. Other = any of the structures that did not fit any of the criteria.

## Conclusions

- In the NOESY and COSY spectrums, the neutral red interacted with the C's and G's more than the A's and T's in the DNA. However, the molecular mechanics calculations show something different. More research will need to be done in order to understand this interaction.
- Another molecule, toluene-2,5 diamine sulfate, is found in many hair dyes. It would be intriguing to see the interaction between this molecule and DNA.
- This project is beneficial in our world today because it helps people become aware of what products are pollutants to humans and the environment.

## Grants and References

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(1) B. Khairutdinov, et al. *Journal of Molecular Structure*, vol. 1074, no. 4, Sept. 2014, pp. 126–133, https://doi.org/10.1016/j.molstruc.2014.05.073. ; A. Myari, et al. *JBIC Journal of Biological Inorganic Chemistry*, vol. 12, no. 3, 7 Nov. 2006, pp. 279–292, https://doi.org/10.1007/s00775-006-0184-8. ; L. Wu, et al. *Chemistry (Weinheim an Der Bergstrasse, Germany)*, vol. 19, no. 17, 22 Apr. 2013, pp. 5401–5410, https://doi.org/10.1002/chem.201203175.

(2) P. Fagan, *Nucleic Acids Research*, vol. 24, no. 8, 15 Apr. 1996, pp. 1566–1573, https://doi.org/10.1093/nar/24.8.1566. ; N.A. Froeystein, et al. *Journal of the American Chemical Society*, vol. 116, no. 8, 1 Apr. 1994, pp. 3240–3250, https://doi.org/10.1021/ja00087a009.

(3) Gaussian 16, Revision C.02, M. J. Frisch, et al. Inc., Wallingford CT, 2016.

(4) A. K. Rappé, et al. *J. Am. Chem. Soc.*,114 (1992) 10024-35.

(5) A. K. Rappé, et al. "APT: A next-generation QM-based reactive force field model," *Mol. Phys.* 105 (2007).

(6) H.M. Sobell, et al. *J.MOL.BIOL.*, vol. 114, no. 3, 1 Aug. 1977, pp. 333–365, https://doi.org/10.1016/0022-2836(77)90254-6.