Structural Determination of the Active Anti Cancer Molecule in Achyranthes Aspera

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Introduction

Pancreatic cancer (**PaCa**) is very aggressive and fatal. In India, *Achyranthes aspera* (Family: *Amaranthacea*) is used as an anticancer drug. However, due to lack of clear documentation, its clinical efficacy remains unknown. In a first systematic effort, we demonstrated the anticancer properties of *A. aspera* leaves (Leaf Extract; LE) on human cancer cells *in vitro* and *in vivo* (Subbarayan et al., 2010; Subbarayan et al., 2012). We found that LE was comparatively more toxic to PaCa cells. Importantly LE was nontoxic to mice and significantly inhibited PaCa tumor growth in mice (Subbarayan et al., 2012). The active anticancer compound (PE3) has been purified from LE. The purpose of this project was to structurally characterize PE3 using NMR.

Experimental

We have used the 1.5-mm ^{13}C probe as well as a commercial Bruker cryoprobe, both operating at 600 MHz, to collect the following datasets on PE3: ^{1}H 1D, ^{13}C 1D, ^{13}C-DEPT, dqf-COSY, TOCSY, ROESY, NOESY, ^{13}C-HSQC, and ^{13}C-HMBC. Using these spectra, we were able to determine that this compound was a porphoryin ring and the fragments of this compound.

Results and Discussion

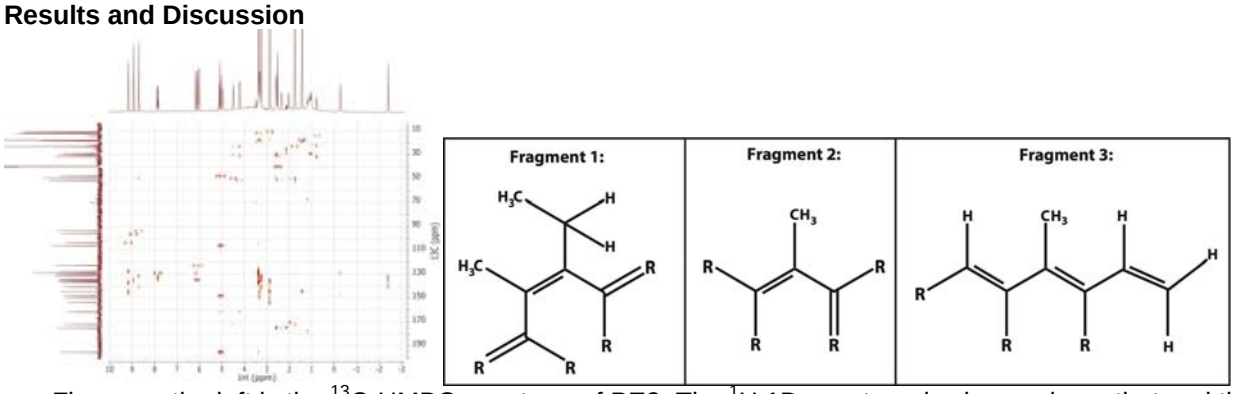

Figure on the left is the ^{13}C-HMBC spectrum of PE3. The ^{1}H 1D spectrum is shown above that and the ^{13}C 1D spectrum is to the left. Datasets such as those on the left were instrumental in identifying many of the fragments such as those in the figure above on right. An unusual 1H at -2.4 ppm, in addition to the MS^{n} fragmentation pattern, is consistent with a porphyrin ring (lacking a coordinated metal) with at least three side chains other than methyl groups. ^{15}N experiments will provide further evidence of this structure and experiments like INADEQUATE or 1, 1 ADEQUATE will help to resolve additional fragments and assign quaternary carbons. Fragments were later connected via X-ray crystallography, which confirmed the suspected structure attained form NMR.

Conclusions

Using NMR and X-ray crystallography, we have identified PE3 as the active anticancer molecule in *Achyranthes aspera*. We have now resolved the structure of PE3 using a combination of NMR and X-ray crystallography techniques.

Acknowledgements

A portion of this work was performed at the National High Magnetic Field Laboratory, which is supported by National Science Foundation Cooperative Agreement No. DMR-1157490, the State of Florida, and the U.S. Department of Energy.

References

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