Combined use of NIR Raman spectroscopy and Baro-Bioreactor to correlate the impact of extreme physico-chemical environments (D₂O solvent, high temperature and high pressure) on the viroids structure and function

Viroids are the smallest non-encapsidated RNA plant pathogens. They are able to infect dramatically a broad range of plants. The Avsunviroidae such as Avocado sunblotch viroid minus strand (ASBVd(-)) is a compact rod-like circular RNA which possessa catalytic hammerhead ribozyme (HHR) motif responsible for crucial cleavage step during viroid replication. To date little is known regarding the structure and conformation of ASBVd viroids, the catalytic role of Mg²⁺ and the ways by which such viroids induces diseases. This prompts us to develop a NIR Raman spectroscopy which is a sensitive technique for monitoring RNAs molecular structure and a Baro-bioreactor designed to allow rapid injections of effectors and sampling out of biomolecular products under pressure for activity studies. ASBVd(-) viroids exhibit a typical A-type RNA conformation with ordered double helical content and a C₃'-endo/anti sugar pucker configuration. Deuteration and temperature perturbed differently the RNA's phosphodiester conformation. Mg²⁺-activated self-cleavage does not significantly alter the secondary RNA's structure but noticeable Raman frequency downshifts were observed suggesting that several phosphodioxy structure, internal loops and hairpins of the cleaved viroids have changed. RNA self-cleavage activity decreased upon deuteration indicating some accessibility of H-bonding network and rigidity of RNAs structure. A pressure-induced decrease of the RNAs self-cleavage pH profile is interpreted as a consequence of some compaction of the structure and a release of catalytic water molecules during catalysis. All these data will constitute the basis for further studies of the interactions of such viroids with therapeutic agents and cell membranes.

Biography

Gaston Hui Bon Hoa has completed his PhD in 1974 from the University of Paris XI and obtained his positions in I.N.S.E.R.M. since 1975. He is an Emeritus Director of Research since 2000 in the Hospital Bicêtre Center, France. His expertise is focus on the studies of cryo-enzymology and enzyme intermediates (1978-1980), cytochrome P450's structure and function (1981-1992), pressure-induced protein's stability, compressibility and dynamics (1992-2015). In 2012, he started the study of viroids structure, conformation and function. He has published more than 150 papers in reputed international journals and has been serving as an Editorial Board Member of repute.

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