MS-based approaches for the elucidation of nucleic acid higher-order structure and dynamics

The discovery of ribozymes and riboswitches has keenly reasserted the critical role played by higher-order structure in determining the function of nucleic acid sequences that do not code for actual proteins. Mass spectrometry-based approaches can provide valuable information on base-pairing and long-range interactions, which define the secondary, tertiary, and quaternary structure of nucleic acids. We have been developing strategies based on high-resolution and ion mobility spectrometry (IMS) mass spectrometry to investigate the structure-function relationships in non-coding viral RNA. We have demonstrated that the concerted application of footprinting and crosslinking methods can provide valid spatial constraints for modeling operations, leading to the solution of actual 3D structures. Top-down strategies can provide direct information about the position and strength of base-pairing interactions that stabilize higher-order structure. Putative structures are corroborated by IMS determinations that reveal the global topology of target RNA. These approaches constitute a valuable alternative for the investigation of systems that, owing to their large size and flexibility, are not directly amenable to classic high-resolution techniques employed in structural biology. Their implementation has been providing new insights into the processes of genome recognition, dimerization, and packaging of HIV-1 and other retroviruses, which have the potential of leading to the development of novel therapeutic strategies.