Prima-1 Reactivates Mutant p53 through the Mobilization of its Aggregated Fraction in the Cell

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INTRODUCTION: p53 is a transcription factor involved in cell cycle control, leading to apoptosis or to the arrest of the cell cycle for DNA damage repair. It is mutated in around 50% of all tumors. In general, mutations occur on the DNA-binding domain, leading to its loss of function. It was shown that mutant p53 can form aggregates that are related to cancer development. The reactivation effect of PRIMA-1, a classical drug described to stabilize and reactivate mutant p53 structure and function, has been widely described, but its mechanism of action is still controversial.
OBJECTIVES: Here, our aim is to demonstrate that Prima-1 is capable to inhibit p53 aggregation, to describe the protein interactions involved , and whether amyloid p53 is reactivated by Prima-1.
MATERIAL AND METHODS: Recombinant p53C aggregation was followed by ThT fluorescence at 440nm (exitation) and 482nm (emission). Immunofluorescence, immunoblotting and immunoprecipitation assays were performed using the antibodies A11 (anti-amyloid oligomers) and DO-1 (anti-p53).
RESULTS AND DISCUSSION: 2-methylene-3-quinuclidinone hydrate (MQ), the major PRIMA-1 active metabolite, has been shown to inhibit WT and mutant recombinant p53 central core domain (p53C) aggregation. The WTp53 form has been protected in a lower degree. MQ has also been shown to inhibit the seeding promoted by mutant p53 cellular extract on WTp53C. The same seeding inhibition effect was observed for extracts from cells treated with PRIMA-1. Immunoprecipitation assays showed that the p53 content in the amyloid fraction of cell extracts is reduced after prima-1 treatment, indicating that the amyloid fraction of total cell p53 is the mobilized one after prima-1 treatment.
CONCLUSION: These results might lead to a different understanding on the controversial mechanism of action of PRIMA-1, which may contribute to the development of new drugs targeting p53 aggregation for cancer treatment.

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