Unraveling the Interaction Between poly(CUG)RNA And the MBNL1 Protein And Inhibition of Complex Formation by Small molecules

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Trinucleotide repeat expansions are the genetic cause of numerous human diseases, including Huntington disease, fragile X mental retardation, and mytonic dystrophy type 1. Myotonic Dystrophy (DM1) is an autosomal dominant neuromuscular disorder associated with a $(CTG)_n$ expansion in the 3'- untranslated region of the DM1 protein kinase (DMPK) gene. The disease is characterized by a waning of the muscles (muscular dystrophy), eye-lens opacity and myotonia. In the case of DM1 the toxic poly(CUG)RNA binds to and sequester key proteins MBNL1 (muscleblind-like protein 1), preventing them from mediating proper splicing of two pre-mRNAs, cardiac troponin T (cTNT) and insulin receptor (IR). The severity of DM1 correlates with the length of the CTG repeat tract in peripheral blood. Individuals with minimal expansions of 50-100 repeats generally have mild, late-onset symptoms, where as those with 1000 or more repeats usually have severe disease in infancy. Expansions of 1000-4000 repeats affect skeletal muscle, heart, ocular lens and brain tissues. We are studying the inhibition of the poly(CUG)-MBNL1 complex by small molecules using gel-shift assays and fluorescence anisotropy. We are also investigating the binding site requirements, stoichiometry, and cooperativity of complex formation between MBNL1 protein and poly(CUG)RNA. These studies were guide us in designing better small molecule binders for inhibiting the poly(CUG)RNA-MBNL1 complex. The inhibition of $(CUG)_4$ and $(CUG)_{12}$RNA-MBNL1 complexes by a small molecule has been shown by gel-shift assays. The experiments for inhibition studies of complexes formed between MBNL1 and higher repeats of poly(CUG)RNA are under progress.