Translational inhibition of mRNA with a point mutation by photoresponsive α-haloaldehyde-conjugated oligonucleotides

(*Graduate School of Science and Technology, Kyoto Institute of Technology*) ○Kentaro Kobata, Soichi Tatsumi, Kazuya Matsuo, Tomonori Waku, Akio Kobori

Keywords: Antisense oligonucleotide; Photo-cross-linking reaction; α-haloaldehyde

It has been found that point mutations in GTPase-coding genes are responsible for the transformation of cells. Therefore, the selective regulation of the mRNA transcribed from genes with a point mutation could be effective in cancer treatment. Previously, sequence-selective cross-linking reactions between photo-cross-linking oligonucleotides (ODNs) and a target nucleotide with a point mutation have been reported [1, 2]. These ODNs selectively inhibit translation of KRAS gene having uridine at the mutation site under clinically relevant conditions. Considering potential benefits of photo-cross-linking reagent-nucleotide conjugates, it is important to develop ODNs with a photo-cross-linking group forming covalent bonds with purine nucleobases at a specific position in target nucleotides.

Previously, we developed photo-cross-linking ODNs with a photoresponsive α-haloaldehyde moiety at the 5'-end of the strand (**Figure 1a**) that selectively react with target DNAs or RNAs with an adenosine or a cytosine nucleobase at the frontal position of the photo-cross-linking moiety. However, it is also found that the α-haloaldehyde group introduced at the 5'-end of the strand reacted with the neighboring bases of the mutation site. To solve this problem, we have developed new photo-cross-linking ODNs with a photoresponsive α-haloaldehyde moiety in the middle of ODNs (^{pro}PXA-ODN) as shown in **Figure 1b** [3]. According to cross-linking studies, ^{pro}PXA-ODNs selectively reacted with mutated RNAs in a sequence-specific manner. In this study, we evaluated the selective translational inhibition of mutated mRNA using ^{pro}PXA-ODNs by *in vitro* translation system.

(a) PXA-ODN (b) ^{pro}PXA-ODN

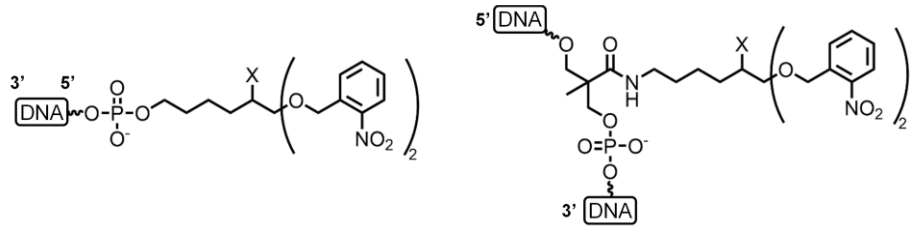

Figure 1. Chemical structures of (a) PXA-ODN (X=Cl: PCA-ODN, X=Br: PBA-ODN) and (b) ^{pro}PXA-ODN (X=Cl: ^{pro}PCA-ODN, X=Br: ^{pro}PBA-ODN).

(1) M. Higuchi, *et al.*, *Oligonucleotides*, **2010**, *20*, 37-44.
(2) T. Sakamoto, *et al.*, *Biomater. Sci.*, **2014**, *2*, 1154-1157.
(3) Y. Sugihara, *et al.*, *J. Org. Chem.*, **2016**, *81*, 981-986.