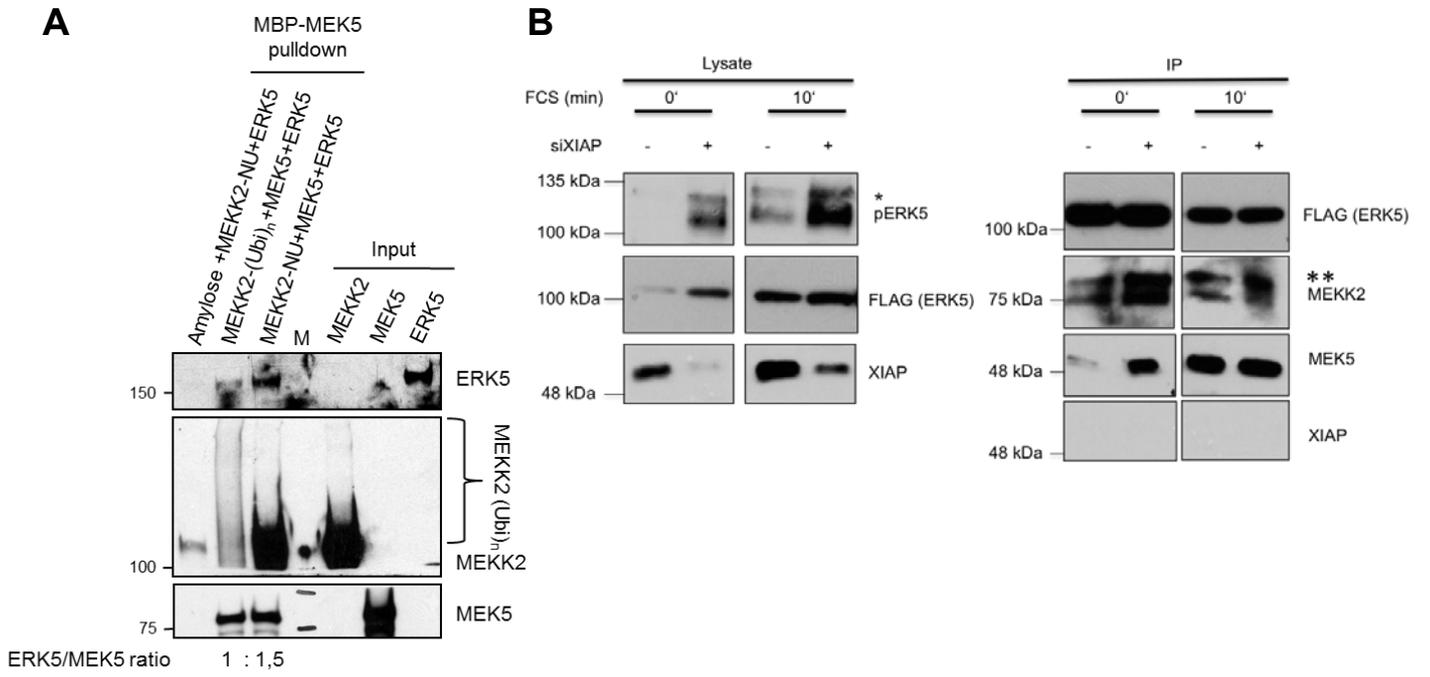


# Supplementary Figure S6



### **Supplementary Figure S6**

(A) Ubiquitination of MEKK2 impairs ERK5 binding to MEK5. The trimeric complex formation between MEKK2-MEK5-ERK5 was analyzed using non-ubiquitinated (-NU) and ubiquitinated  $-(Ubi)_n$  MEKK2. MBP-tagged MEK5 was used for pulling down GST-MEKK2 and GST-ERK5. Background binding of MEKK2 and ERK5 to Amylose resin was tested as a control. (B) HeLa cells stably expressing MEK5-V5 and ERK5-Flag were transfected with Control or XIAP siRNAs. The cells were stimulated with FCS after 15 hours of starvation and the ERK5-Flag fusion protein was isolated with magnetic anti FLAG-beads. The co-precipitating MEK5 and MEKK2 were monitored by immunoblots. The activation of pERK5 and the efficiency of the knock down were verified in the total cell lysates.