

BACKGROUND

The Ku autoantigen was originally isolated as a nuclear protein recognized by the sera of lupus erythematosus patients. It is a heterodimer made of two subunits of 70 (Ku70) and 80 kDa (Ku80) and is endowed with both duplex DNA end-binding capacity and helicase activity (human DNA helicase II). Ku has the ability to bind specifically to the ends of duplex DNA and then slide into the duplex to form a structure similar to beads on a string. Ku is also endowed with an ATP-dependent DNA helicase activity denominated human DNA helicase II (HDH II), probably located on a different moiety of the molecule than the one involved in duplex DNA binding.¹ The helicase activity of the Ku protein resides uniquely in the Ku70 subunit. Whereas the DNA end-binding activity can be found only from the two subunits in the heterodimeric form and is practically absent in the separate subunits. The Ku80 subunit, when refolded in the absence of the Ku70 subunit, forms homodimers unable to unwind DNA and bind duplex ends. The three separate species (heterodimer, Ku70 subunit, and Ku80 subunit homodimer) all have ssDNA-dependent ATPase activity.²

Moreover, it is a tightly associated heterodimer of Ku70 and Ku80 subunits that together with the 470 kDa catalytic subunit, DNA-PKcs (which belongs to the PI-3 kinase family), form the DNA-dependent protein kinase. This enzyme is involved in repairing DNA double-strand breaks (DSBs) caused, for example, by physiological oxidation reactions, V(D)J recombination, ionizing radiation and certain chemotherapeutic drugs. The Ku-dependent repair process, called illegitimate recombination or nonhomologous end joining (NHEJ), appears to be the main DNA DSB repair mechanism in mammalian cells. Ku itself is probably involved in stabilizing broken DNA ends, bringing them together and preparing them for ligation. Ku also recruits DNA-PKcs to the DSB, activating its kinase function. DNA-PK phosphorylates many DNA-binding proteins to regulate their functions in DNA repair, recombination, replication and transcription.³ Ku antigen is also an RNA-binding protein that associates with hnRNP complexes. Moreover, DNA-PK consisting of Ku antigen and a kinase subunit (DNA-PKcs) was able to perform an RNA-dependent phosphorylation of hnRNP proteins and of NDH II. Thus, in addition to its functions in DNA double-strand break repair and V(D)J recombination, DNA-PK may also be involved in RNA metabolism.⁴ Inhibition of DNA Unwinding and ATPase Activities of Human DNA Helicase II by Chemotherapeutic Agents provides a effective tool for cancer treatment.⁵

References:

1. Featherstone, C & Jackson, S.P.: Mutation Res./DNA Repair 434:3-15, 1999
2. Ochem, A.E. et al: J. Biol. Chem. 272:29919-26, 1997
3. Smith, G.C.M. & Jackson, S.P.: Gene Dev. 13:916-34, 1999
4. Zhang, S. et al: Nucleic Acid. Res. 32:1-10, 2004
5. Tutaja, N. et al: Biochem. Biophys. Res. Commun. 236:636-40, 1997

TECHNICAL INFORMATION

Source:

Ku70 antibody is a rabbit antibody raised against a short peptide from human Ku70 sequence.

Specificity and Sensitivity:

This antibody detects endogenous Ku70 proteins without cross-reactivity with other family members.

Storage Buffer: Tris-buffered Saline containing 0.1% BSA containing 0.09% Sodium Azide

Storage:

Store at -20°C for at least one year. Store at 4°C for frequent use. Avoid repeated freeze-thaw cycles.

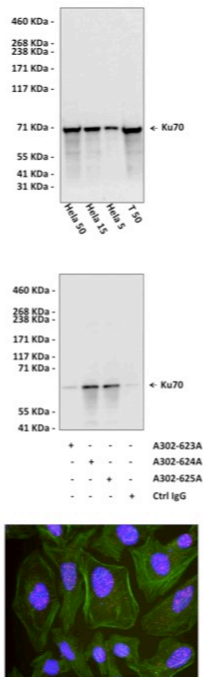
APPLICATIONS

Application:	*Dilution:
WB	1:2,000-10,000
IP	2-5 ug/mg
IHC	1:200-1:1000
ICC	n/d
FACS	n/d
IF	1:100-1:

**Optimal dilutions must be determined by end user.*



QUALITY CONTROL DATA



Top: Affinity purified rabbit anti-Ku70 antibody A302-624A used for WB at 0.04 $\mu\text{g/ml}$. Chemiluminescence with exposure time of 3 seconds.

Middle: Affinity purified rabbit anti-Ku70 antibody A302-624A used for WB at 0.4 $\mu\text{g/ml}$. Chemiluminescence with exposure time of 1 second.

Bottom: This antibody is qualified for the Proximity Ligation Assay (PLA). The image shows representative results for PLA using three color fluorescence, including DAPI stained nuclei (blue), phalloidin stained cytoplasmic F-actin (green), and PLA positive signal (red).

