2.6.1 INTRODUCTION

KN035 is a novel Programmed death-ligand 1(PD-L1) antagonist. As a recombinant fusion protein, KN035 consists of two identical polypeptide chain linked via a pair of disulfide bonds. Each chain contains a human IgG1 Fc fragment and humanize single domain antibody. The single domain antibody (dmAb), which binds to PDL1 and blocks its interaction with PD-1, has been obtained from a focused phage library, derived from PBMC of human PDL1 immunized camel. The dmAb has been humanized there after. Due to two point mutations, the Fc part has muted effector functions, antibody-dependent cell-mediated cytotoxicity (ADCC) and complement-dependent cytotoxicity (CDC).

In vivo and *in vitro* pharmacology studies, clearly demonstrate that KN035 binds to human PD-L1 protein with high affinity (Module 2.6.2 section 2.1) and blocks inhibitory PD-1/PD-L1 interaction (Module 2.6.2 section 2.1). Application of KN035 thus leads to restoration of CTL function, which in turn trigger tumor cell elimination.

The theoretical molecular weight of KN035 is 79.56 kDa. The apparent molecular weight is approximately 82-84 kDa due to glycosylation on Fc. KN035 is formulated to high protein concentration (200 mg/ml) and provided as a transparent liquid solution for subcutaneous injection. Each vial contains 300 mg KN035 in1.5 ml. The investigational product should be stored and shipped at 2-8^{o}C with 9-month stability data available.

The *in vivo* pharmacology studies in animal models suggest the effective dose range for KN035 is between 0.01mg/kg and 10mg/kg (Module 2.6.2 section 2.2). The pharmacokinetic studies suggest KN035 mostly distributed into blood system and tumor tissue with higher affinity and binding specificity. The absolute bioavailability of a single subcutaneous administration of 15mg/kg KN035 was 104.47%. There were no cross-reactions and accumulations in the main organs of test animals.

KN035 did not induce apparent toxicity at the highest dose of 150 mg/kg in the 4-week GLP toxicity study in monkeys. Therefore, 150 mg/kg was considered as the NOAEL in monkeys, it is already 30 times of the clinical dose for most antibody drugs (5mg/kg). Based on FDA guidance for “Estimating the Maximum Safe Starting Dose in Initial Clinical Trials for Therapeutics in Adult Healthy Volunteers”, one tenth of the NOAEL can be used as starting dose for healthy volunteers, it would be 15 mg/kg for KN035. If ICH guideline S9 “Nonclinical Evaluation for Anticancer Pharmaceuticals” is referred, then 1/6 the highest non-severely toxic dose (HNSTD) is used as starting dose, it would be higher than 25 mg/kg for KN035. However, the effective dose in pharmacology study of KN035 was much lower. When both the effective dose and the NOAEL are considered, 1 mg/kg is selected as the safe starting dose for the phase 1 clinical trial. This will be a very safe starting dose. It is more than 150 times lower than the NOAEL in monkeys.