Abstract:
Ebola virus (EBOV) belongs to the family *filoviridae* and causes a severe hemorrhagic fever with 50-90% human mortality. EBOV entry requires the surface glycoprotein, GP, to initiate attachment and fusion of viral and host membranes. Here we report the crystal structure of a trimeric and prefusion conformation of GP (GP1+GP2) in complex with a neutralizing antibody fragment, KZ52, derived from a human survivor of the 1995 Kikwit outbreak. The metastable, flexible and highly glycosylated nature of EBOV GP makes this molecule highly challenging to crystallize. We constructed over 140 different versions of the protein, grew 50,000 crystals, and out of the 800 crystals harvested, only one diffracted to 3.4 Å resolution. The construct crystallized contains all domains required for attachment, fusion and entry, and leads to productive cellular infection when pseudotyped onto vesicular stomatitis virus. In the structure, three GP1 viral attachment subunits assemble to form a chalice, cradled in a pedestal comprised of the GP2 fusion subunits (Fig. 1), while a novel glycan cap and projected mucin-like domain restricts access to the conserved receptor-binding site sequestered in the chalice bowl. The glycocalyx surrounding EBOV GP is likely central to immune evasion and explains why patients that survive have low to insignificant neutralizing antibody titres. The KZ52 antibody recognizes a protein epitope at the chalice base where it clamps several regions of the prefusion GP2 to the N terminus of GP1 and precludes rearrangements required for fusion. This structure now unravels the mechanism of Ebola virus GP-mediated fusion and provides a template for future immunotherapeutic development.