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## TITLE

### Use of the Single Cell Gel Electrophoresis (Comet assay) for Comparing Apoptotic Effect of Conventional Antibodies versus Nanobodies

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The large molecular size of antibodies is considered one major factor preventing them from becoming more efficient therapeutically. It is well established that all camelids have unique antibodies circulating in their blood called heavy-chain antibodies (HcAbs). Unlike antibodies from other species, these HcAbs contain a single variable domain (VHH) and two constant domains (CH2 & CH3). HcAbs are a novel type of immunoglobulin-like, antigen binding protein with beneficial pharmacokinetic properties that are ideally suited to targeting cellular antigens for molecular imaging or therapeutic purposes. Since the antigen-binding site of dromedary HcAb is comprised in one single domain, it was referred to as nanobody. In the present work, the different IgG subclasses from immunized camel (*Camelus dromedarius*) were purified employing their different affinity for protein A column (PA) and protein G column (PG). Characterization of IgG subclasses was done by using 12% SDS-PAGE under reducing conditions. Protein bands were visualized after staining with CBB, showing two bands at 50kDa and 30kDa in case of IgG1 while IgG2 and IgG3 produce only one band at 46kDa and 43kDa respectively. The induction of apoptosis by either conventional or nanobodies was evaluated on two different cell lines, Colon and Hepatic cancer cell (HCT116 & HepG2), using the comet assay. Induced apoptosis were confirmed by visualizing DNA fragmentation bands on 2% agarose gel, and the gel was photographed under UV light. This study demonstrates the successful targeting of human cancer colon cell lines by nanobodies in vitro. It may open perspectives for their future use as tumor target vehicle, due to their small size, soluble behavior and they are interact with epitopes that are less antigenic for conventional antibodies.

#### Biography

Professor Ghada Shaker has Egyptian Mission, as a Ph. D. Student in the National research Centre for Biotechnology (GBF), Braunschweig, Germany. She is the member of American Nano Society. She has published more than 23 papers.