**Statement of Purpose:** According to the United States National Cancer Institute, the overall cancer death rate has declined, and the number of cancer survivors has increased. However, the challenge to overcome systemic toxicity and drug resistance caused by current conventional treatments still remains. Therefore, viable cancer therapy alternatives are highly desired. Immunotherapy is a type of cancer treatment that stimulates the immune system to destroy tumor cells. Even though various cancer immunotherapies have been approved by the FDA to treat different kinds of cancer they may encounter limited success because of tumor-induced immune suppression and evasion mechanisms. Myeloid-derived suppressor cells (MDSC) are a heterogeneous population of immature myeloid cells that are induced by tumor-mediated inflammation. These cells play a major role in anti-tumor immunity by inhibiting effector T cell function. It has been shown that depleting this immune suppressive mechanism can lead to enhanced immunotherapy efficacy in cancer (Gabrilovich DI, 2009). Strategies able to target and deplete MDSC could therefore improve the success of cancer immunotherapies. According to this information, a novel approach was used in this work. MDSC targeting polymeric nanoparticles (NP) were prepared using a MDSC-targeting peptide (Qin H, 2014) on the surface of poly(lactic-co-glycolic acid) (PLGA) nanoparticles (NP) loaded with sunitinib. This antiangiogenic drug has shown ability to decrease MDSC accumulation in tumors, spleens and bone marrow of tumor bearing mice. The objective of this study was to prepare, characterize and test the efficacy of this NP formulation. In the future, the benefits of combining this MDSC targeting therapy with an adenovirus based vaccine in controlling tumor growth in tumor-challenged mice will also be tested.

**Methods:** NP were prepared by the nanoprecipitation method. Briefly, sunitinib and PLGA 75:25 carboxylate were dissolved acetone and added drop wise into a MES buffer solution containing polyvinyl alcohol. Subsequently, 1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide and N-hydroxysuccinimide were added and NP were kept under magnetic stirring. NP were purified and collected by centrifugation at 16,100 x g. The particles were then incubated with streptavidin, purified and finally incubated with a biotinylated MDSC-ligand peptide. Sunitinib solution was prepared using DMSO, tween and PBS. Loading and encapsulation efficiency determined by a high-performance liquid chromatography assay with ultraviolet detection. The in vivo efficacy of the targeted NP were evaluated in tumor-challenged mice. C57BL/6 female mice were injected subcutaneously with 10⁶ EL4 cells into the dorsal right flank on day 0. On day 7, mice were treated intravenously during 4 alternate days with 2 mg/Kg of sunitinib in solution of in NP (n=8 mice/group). On day 15 cells isolated from spleen were stained for the presence of monocytic and granulocytic MDSC (mMDSC/gMDSC) with a combination of anti-CD11b-PE, anti-Ly6C-FITC and anti-Ly6G-APC. Samples were acquired using a LSR violet flow cytometer and analyzed with FlowJo software.

**Results:** Sunitinib-loaded PLGA NP had a uniform size distribution with mean diameter of 295 nm. Drug loading and encapsulation efficiency was 1.72μg/mg and 2.41%, respectively. In vivo results show a reduction of gMDSC levels in spleen of animals treated with targeted sunitinib-loaded NP compared to the untreated animals. No differences of mMDSC levels were observed between treated and untreated animals.

**Conclusions:** The preliminary results obtained indicate that the MDSC-targeted NP formulation prepared is a promising formulation to be used as a MDSC targeting therapy. The antitumor benefits of this formulation in combination with an adenovirus based vaccine in tumor-challenged mice will be evaluated in a following experiment.

**References:**