William A. Gibson
L2
LSU Health Sciences Center, New Orleans, LA

Arnold H. Zea, PhD
LSUHSC; Department of Microbiology, Immunology, & Parasitology

"Decreased L-Arginine Availability Blocks the Induction of NO Synthesis in Renal Cell Carcinoma Cells (Renca) as a Mechanism of Defense in Response to Interferon Gamma"

L-arginine is the only endogenous substrate of NO synthase (NOS), and it thus governs the production of NO during different disease states including cancer. L-arginine metabolism has been shown to be an important pathway in the immune escape mechanism in cancer, including renal cell carcinoma. L-arginine can be metabolized in two primary pathways: conversion of L-arginine into L-ornithine and urea by arginase (ARG) and conversion of L-arginine into citrulline and nitric oxide (NO) by inducible nitric oxide synthase (NOS2). The presence of ARG has been shown to lead to increased tumor growth by the conversion of L-ornithine into polyamines that are essential for cell growth. In contrast, the production of NO by NOS2 has been shown to limit tumor growth due to the cytostatic effects of NO in IFNγ-stimulated tumor cells.

In the murine renal cell carcinoma cell line Renca, the expected production of NO was not seen in response to IFNγ. It has been shown by Western blotting that the NOS2 protein was completely absent in these cells when L-arginine availability was decreased, whereas NOS2 mRNA expression was not inhibited under these conditions. It has also been demonstrated that ARG activity is higher than baseline in Renca cells. We believe that the overactive ARG disrupts NOS2 function by depleting L-arginine levels and causing what is known as the "arginine paradox." This paradox occurs when extracellular levels of L-arginine are depleted (regardless of the intracellular level), leading to decreased levels of NOS2.

We demonstrated that Renca cells produce decreased amounts of NO compared to another murine renal cell carcinoma line, CL-19, in the absence and in the presence of IFNγ. It was also shown that Renca cells are able to produce some level of NO when cultured in media with increased L-arginine concentrations (2000 μM as compared to 1000 μM). This would support the arginine paradox hypothesis. In this case, the arginine paradox is being overcome by adding more extracellular L-arginine than can be metabolized into L-ornithine and polyamines through the ARG pathway, leaving enough L-arginine for production of NOS2. The exact mechanism behind this has yet to be elucidated. Determining this mechanism, as well as investigating the potential role of glutamine in the polyamine pathway, are future goals of this project.