

Behind the Scenes in Pharmacognosy: Team Spirit Leads the Way

In January of 2013, the *Journal of Natural Products* published work from ASP member Dr. Leng Chee Chang and colleagues at the University of Hawai'i, Hilo, Hawai'i entitled, "Biologically Active Withanolides from *Withania coagulans*." Dr. Chang shares her work on the vegetable rennet, as well as her inspiring laboratory motto. This article is among the top read articles in JNP from January to March of this year. Please read the full article in the *Journal of Natural Products*, 2013;76: 22–28. doi.org/10.1021/np300534x.

By Dr. Amy Keller

How did you become interested in working with *Withania coagulans* compounds, and how did you come to focus on their potential anti-cancer activity?

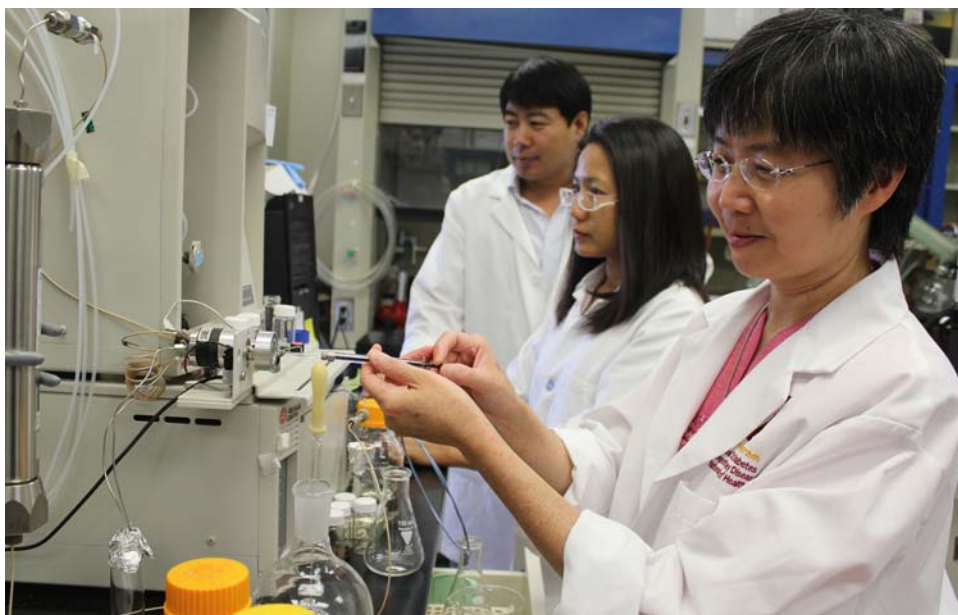
My research interest is focused on naturally occurring cancer chemoprevention and anticancer agents with low toxicity. Therefore, I usually work with edible species. As a part of the exploration of important medicinal plants of Pakistan for cancer treatment, initially six Pakistan plants were chosen for the study based on ethnopharmacological data. Among these plants, *Withania coagulans* showed the highest biological potential. Interestingly, this plant, popularly known as vegetable rennet, is also a highly valued medicinal plant.

Who in your laboratory carried out the research?

The research was made feasible with the arrival of Dr. Ihsan-ul-Haq, a visiting scholar who worked in Dean John Pezzuto's laboratory at The Daniel K. Inouye College of Pharmacy, University of Hawai'i at Hilo, Hawai'i (DKICP-UH Hilo.) Sponsored by the Higher Education Commission Pakistan, Dr. ul-Haq conducted most of the experimental work, along with my postdoctoral associate, Dr. Ui Joung Youn. In addition, our collaborators from Dean Pezzuto's laboratory performed the cancer chemopreventive bioassay testing. The X-ray crystal structure was done by Dr. Charles Simmons, from the Department of Chemistry at UH Hilo.

Could you provide a brief explanation of the work and results in your own words? In what way are the data in your paper new?

The overall aims of this study were to evaluate the cancer chemopreventive potential of withanolides from *W. coagulans* as nuclear factor kappa B (NF- κ B) and inducible nitric oxide synthase



MS. MAGGIE MORRIS

(From right to left) Dr. Leng Chee Chang works in her laboratory at The Daniel K. Inouye College of Pharmacy at the University of Hawai'i at Hilo with Ms. Mayuramas "Jan" Sang-Ngern, a Ph.D student and Dr. Ui Joung Youn, a postdoctoral associate. Dr. Chang is the principal investigator on a project that aims to find a natural product treatment for cancers with fewer side effects and lower toxicity than current therapies.

(iNOS) inhibitors. NF- κ B is an important transcriptional factor that regulates numerous physiological processes including cellular proliferation, development, differentiation, immunity, apoptosis, inflammation, and metabolism. The critical role for persistently-active NF- κ B is evident in many cancers. Aberrant NF- κ B activity dysregulates growth and survival, promotes angiogenesis, migration and invasion of tumor cells, and induces tumor immune tolerance. The structure and absolute stereochemistry of new withanolides (1-3) were determined by NMR, NOESY, CD and X-ray diffraction methods. This is the first report of structure-activity relationship of stereo structures of *W. coagulans* withanolides with inhibition of nitric oxide and NF- κ B activities. They inhibited nitric oxide production in LPS murine macrophage RAW

continued on page 15

Behind the Scenes in Pharmacognosy: Team Spirit Leads the Way

continued from page 14

264.7 cells, and inhibited tumor necrosis factor-alpha induced NF-kB activation, with IC_{50} values in the range of 1.6-38.5 μ M.

What impact does this research have on natural product science and health research in general?

Plants with ethnopharmacological uses have been a primary source for early drug discovery and chronological experiences with these plants as curative tools have helped to isolate and develop single chemical entities in modern medicine. In the present study, six medicinally-important plants selected from different areas of Pakistan and based on extensive information from local healers, were investigated for biological activities to evaluate their cancer chemopreventive and cytotoxic potential. Seven different bioassays were used: inhibition of TNF- α activated NFkB, aromatase inhibition, inhibition of NO production in lipopolysaccharide activated macrophage RAW cells (iNOp), interaction with RXRE, induction of QR1, DPPH free radical scavenging and inhibition of LU-1 lung cancer cell and MDA-MB-231 breast cancer cell proliferation. This investigation established the isolation of biologically active entities from *W. coagulans* that had been used extensively in the folk medicine to treat cancer. The exploration of the cancer chemopreventive and anticancer potential of the plant and the compounds isolated therefore provides a new addition to natural products science and health research. The purified compounds can serve as a promising source for new drug development to combat cancer. Proposed future prospects of the research include:

The isolation of more withanolides from the plants containing them for the establishment of a broader structure-activity relationship (SAR); this will be followed by the development of synthetic approaches for the most active analogues. Also, studies for preclinical trials could be carried out to evaluate the in vivo activity and toxicity of the isolated compounds. The identification of bioactive and less cytotoxic natural products in *W. coagulans* that are edible should promote the use of *W. coagulans* as herbal remedies for anticancer regimens. The low cost of *W. coagulans* should increase access to their use as an alternative treatment for cancer in Pakistan and in United States.

What is a favorite nonscientific activity of your lab?

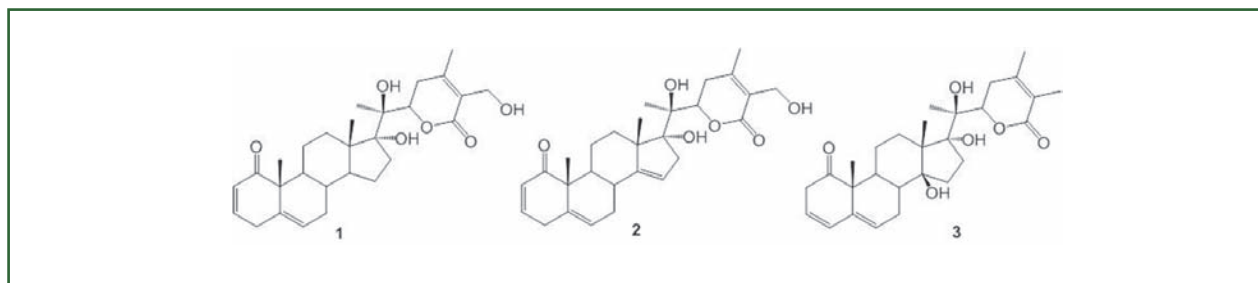
It ranges from exploring the beauty of Hawaii to practicing meditation for inner peace.

What is your lab's motto?

Practice the discipline of perseverance, promote an effective-team spirit with consensus and cooperation; be passionate in what we do, and compassionate in helping others.

What is your greatest extravagance in the laboratory?

Shimadzu high performance liquid chromatography (HPLC) includes evaporate light scattering detector (ELSD) and photo diode array (PDA) detectors. ■



(From left to right) Three new steroidal lactones, withacoagulin G (**1**), withacoagulin H (**2**), and withacoagulin I (**3**)

Practice the discipline of perseverance, promote an effective team spirit with consensus and cooperation; be passionate in what we do, and compassionate in helping others.