## Conference Series LLC Joint International Event on 5<sup>th</sup> European Immunology & Innate Immunity

July 21-23, 2016 Berlin, Germany

## The clathrin assembly protein from *Arabidopsis thaliana* possesses adenylate cyclase activity and is involved responses to biotic stress

Oziniel Ruzvidzo North-West University, South Africa

The need to develop agricultural crops with improved stress resistance has led plant biotechnologists to strategically focus L on those plant molecules involved in the maintenance of homeostasis. One such molecule that is typically involved in signal transduction is the cyclic adenosine 3', 5'-monophosphate (cAMP), which is generated by adenylate cyclases (AC). Even though ACs have previously been experimentally proven to be centrally involved in numerous stress response systems in animals, prokaryotes and lower eukaryotes, their existence and or functional properties in higher plants have until recently been a very serious matter of debate and controversy. To date, only four ACs have since been confirmed in higher plants, specifically in the Arabidopsis thaliana, the Zea mays, the Nicotiana benthamiana and the Hippeastrum hybridum plants. Since it is inconceivable that a single AC per plant can essentially account for all the known and or currently reported cAMPdependent processes in plants, we then set out to practically identify and functionally characterize a second probable AC candidate from A. thaliana in the form of a putative clathrin assembly protein (AtCAP: At1g68110), with a view of elucidating its exact physiological and biological roles in higher plants. Firstly, we conducted a preliminary bioinformatic analysis of this protein, which then showed us that this putative protein candidate is actually a multi-domain, multi-functional molecule with a possible role in AC-dependent stress response and adaptation mechanisms. We then proceeded to clone and recombinantly express its AC-containing fragment domain (AtCAP-AC) in chemically competent E. cloni BL21 (DE3) pLysS DUOs cells and then showed its ability to induce the generation of endogenous cAMP in this prokaryotic expression system. Furthermore, we also demonstrated the complementation of a mutant non-lactose fermenting cyaA SP850 Escherichia coli strain by this recombinant AtCAP-AC protein, to eventually ferment lactose as a result of this protein's ability to generate the most required cAMP for this process. In addition, we also undertook the purification process of this truncated AtCAP-AC protein followed by the functional characterization of its in vitro AC activities, which in turn revealed that this recombinant is indeed a bona fide soluble adenylate cyclase (sAC) physiological and biochemical roles may be mediated by cAMP and possibly, via a calmodulindependent signaling system. Lastly and in order to gain more insights into the possible physiological roles of this AtCAP-AC protein in higher plants, we then bioinformatically analyzed its expressional profiles using the various computational and web-based bioinformatic tools and found out that the protein is centrally involved in responses to biotic stress factors, whose systems are both cAMP and SORLIP1AT core motif-dependent.

## Re-thinking the role of immunity: Lessons from the evolutionary past for the present and the future

Thomas C G Bosch Kiel University, Germany

Characterization of the innate immune repertoire of evolutionary old organisms is of both fundamental and applied interest; it not only provides insights into the basic immunological "tool kit" of the common ancestor of all animals but is also likely to be important in understanding human barrier disorders by describing ancient mechanisms of host/microbial interactions and the resulting evolutionary selection processes. Here, I evaluate our current understanding of the evolution of epithelialbased innate immunity. I highlight growing evidence that the innate immune system with its host-specific antimicrobial peptides and rich repertoire of pattern recognition receptors has evolved in response to the need for controlling resident beneficial microbes rather than to defend against invasive pathogens.