synthase with Q211E substitution. Using the crystal violet staining assay, we found that susceptibility of the cells transfected with either vector was ten times higher than susceptibility of the untransfected cells. In the thin layer chromatography overlay of glycolipids derived from transfected 2102Ep cells, Shiga toxins were found to bind only to Gb3. TLC overlay of glycolipids from red blood cells also revealed the binding to Gb3, but in NOR-positive erythrocytes an additional band was detected, migrating similarly to P1 glycolipid. Thus, our results show that Shiga toxins VT1 and VT2 do not bind to NOR antigens suggesting that the penultimate galactose residue of the Shiga toxin receptor cannot be replaced by *N*-acetylgalactosamine.

SW06.S28–18
X-ray structure of a stable protease-resistant glectin-9 with short linker

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Human Galectin-9 (hG9) belongs to a lectin family and has two different carbohydrate recognition domains (N-terminal and C-terminal CRDs) which are specific to beta-galactosides. The physiological function of hG9 is miscellaneous and known to be an eosinophil chemoattractant, T-cell immunoglobulin domain and mucin domain 3 ligand, and inducer of T helper 1 cell apoptosis. There are isoforms depending on the length of the linker region between N- and C- CRDs. Two isoforms, hG9L (355 a.a.) and hG9M (323 a.a.), are protease sensitive due to their long linker with ca. 30–60 a.a. To date, hG9Null has been known as protease resistant hG9 mutant with short linker (2 a.a.) and retaining the biological function of native hG9, eosinophil chemoattractant activity, apoptosis induction of activated T-cells and pleiotropic immune response [1]. The structure of each CRDs has been already available, however the whole structure of hG9 with two CRDs is not reported and is intriguing for understanding of interaction of N- and C- CRDs. In this study, we determined the crystal structure of a stable hG9Null with a mutation of sugar binding site of C-CRD. Each CRD structure has no big difference with the previously reported structure as known as beta sandwich structure. Although the linker region including N-terminal region of C-CRD (10 a.a.) was not visible in the electron density maps, N- and C- CRD interact with each other via a metal ion, making a rigid stable structure of hG9Null mutant. Recently, Nishi *et al*. constructed a highly soluble and stable hG9 mutant with a truncated and optimized linker region (284 a.a.) [2]. Interestingly, the optimized truncated linker region corresponds to the invisible linker region in our crystal structure of hG9Null mutant. It is still unknown that N- and C- CRD of hG9 can interact with each other without depending on the length of linker region. Although it might be occasional, our crystal structure of hG9Null mutant showed the possibility of their interaction.

Reference

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A new lectin from coral *Gerardia savaglia:* purification, physico-chemical characterization and thermodynamics of saccharide binding

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A new lectin has been purified from Mediterranean coral *G. savaglia* by affinity chromatography on mannose-sepharose. Two-dimension electrophoresis showed a single spot at pI 8.9, while MALDI TOF/TOF mass spectrometry analysis revealed four molecular variations with molecular weights 16 803, 16 769, 16 617 and 16 583 Da. Identical N-terminal amino acid sequence confirmed similarity between these four molecules. The hemagglutination activity of the lectin was analyzed at different temperatures, pH and in the presence of divalent cations. Far UV circular dichroism (CD) showed unusual spectra with high content of β-structures. Thermal stability was investigated by differential scanning calorimetry (DSC) and CD spectroscopy. The DSC yielded a curve with endothermic peak above 72°C with midpoint at 78.9°C, accompanied with loss of structure confirmed by CD. Both methods showed that the thermal unfolding process is irreversible. Lectin-D-mannose interactions were studied by CD and isothermal titration calorimetry (ITC). No changes in far UV CD spectra have been observed upon addition of mannose even in the presence of divalent cations. On the other hand ITC studies showed binding of mannose is accompanied by large negative enthalpy change (due to high number of hydrogen bonds) and unfavorable entropy contribution (due to solvent rearrangement or loss of ligand conformational flexibility). Obtained affinity for D-mannose is in millimolar range, which is in accordance with results reported previously for specific lectin – saccharide interactions.

Biological functions of lectins in corals are poorly understood. There are suggestions that they are important for symbiosis with algae and pathogen defense. Corals are endangered species and one of the reasons for their extinction is connected with the loss of symbiosis with algae. This study presents a novel type of lectin with unusual mannose specific structure and provides basis for understanding role of lectins in coral.

SW06.S28–20
Effects of astragalus, *lemon balm, clove,* fenugreek and cinnamon on blood glucose level after oral glucose loading in rats

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Background and aim: Herbs like astragalus, *lemon balm, clove,* fenugreek and cinnamon are consumed in various ways (dietary supplement, herbal tea etc.) all over the world. Consumption of these herbs (along with other health beneficial effects) has been mentioned to be helpful in decreasing blood glucose levels of dia-