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IR-spectrum of blood and redox: diagnostics of cancer

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Early studies of blood or serum thin layer on glass plate by IR spectrometer as a potential cancer diagnostic tool were initially received with considerable excitement. But many intrinsic specimen characteristics may have no relationship with the disease in question. Therefore a diagnosis was confirmed only in 60% cases. It has been known for a few decades that a relationship exists between cancer and level of –SH group in patient blood. Some insights into the value of SH group in blood have been obtained from studies performed in experimental animals and in vitro. These studies provided some of the first evidence for differential value IR parameters (band Amid I) between exsiccate thin layer of blood or serum of intact animals and tumor animals ($p < 0.005$). IR parameters correlate highly with level of SH group protein of serum or pure albumin ($r = 0.97$). Human serum albumin (HSA) is the most abundant protein in the circulatory system and it has a number of functions. One of a number functional roles of this molecule seems to be the maintenance of the redox potential in the extracellular fluid, because it is a mixture of mercaptalbumin (reduced form; in humans, HMA) and non-mercaptalbumin (oxidized form; in humans, HNA), i.e., a major part of the redox couple in plasma. Our studies indicate that state of SH group of cysteine of albumin can be related to indicator of redox in term of IR parameters. Based on the structure of a molecule of albumin we developed the theoretical norm of redox for IR data. This method offers important advantages for quantitative estimation as common number oxidized/reduced form as individually one. Use selection IR data (1700cm⁻¹–1600cm⁻¹) on level of SH group without proteomic profiling we obtained nevertheless more exact diagnosis of women with breast cancer. This method offers also important advantages for monitoring of treatment. However, additional control experiments are essential, such as using control subjects with other types of inflammatory disease, or even different cancers.

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Purification and characterization of a novel monocot lectin with mitogenic and in vitro anti-proliferative activity from *Arisaema curvatum*

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A lectin with in vitro anti-proliferative activity and potent mitogenic and has been purified from tubers of a wild monocot plant *Arisaema curvatum* Schott by affinity chromatography on asialofetuin linked amino-activated silica. A single band of pure lectin corresponding to subunit Mr 13.0 kDa was observed in SDS-PAGE at pH 8.3. The native molecular mass as determined by gel filtration chromatography was 52 kDa, suggesting a homotetrameric structure. ACL gave multiple bands in isoelectric focusing and in native PAGE at pH 8.3 like other monocot lectins. ACL was inhibited by N-acetyl-D-lactosamine (LacNAc), a disaccharide and asialofetuin, a complex desialylated serum glycoprotein. When treated with denaturing agents, the lectin was stable in the presence of urea (4 M), thiourea (3 M) and guanidine HCl (4 M). The lectin had no requirement for divalent metal ions. ACL was a glycoprotein with a carbohydrate content of 1.5%. Amino acid analysis revealed high content of

aspartic acid, glutamic acid, glycine and threonine and a very low amount of methionine but complete absence of cysteine. Amino acid modification studies of ACL revealed the involvement of tryptophan and tyrosine residues involved in lectin-sugar interaction. The lectin showed potent mitogenic response towards human lymphocytes. The mitogenic activity of ACL was even more than that of Con A, a standard well-known plant mitogen. ACL exhibited a fluorescence emission maximum (λ_{max}) at 341 nm upon excitation at 295 nm. Using Far UV CD spectra the estimated secondary structure was 38% α -helix, 27% β -sheet and 35% random contributions. In vitro anti-proliferative activity of ACL was tested on eleven different human cancer cell lines viz. MCF-7 (Breast), SK-N-SH (CNS), 502713 (Colon), Colo-205 (Colon), HCT-15 (Colon), HT-29 (Colon), SW-620 (Colon), Hep-2 (Liver), IMR-32 (Neuroblastoma), DU-145 (Prostate) and PC-3 (Prostate). The concentrations of ACL which produced 50% inhibition (IC₅₀) of cancer cell lines viz. Colo-205, SK-N-SH, HCT-15, IMR-32, SW-620, Hep-2 and HT-29 was 28, 33, 42, 44, 45, 48 and 81 μ g/ml respectively. ACL is specific for LacNAc, which is one of important cancer markers. The inhibitory effect of ACL was not associated with toxicity to the cell lines. Future research is focused find in vivo effect of lectins on appropriate animal models of human diseases and other agents (e.g., synthetic compounds) to explore the synergistic effect of these agents to prevent cancer.

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Impression cytology of ocular surface diseases of the bulbar conjunctiva as non-invasive technology for diagnostic and monitoring for cancer prevention

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In recent years, impression cytology has been used in diagnostic various ocular surface diseases.

Aim: To evaluate the accuracy of impression cytology employing a Biopore membrane device in the differentiating benign, premalignant and malignans conjunctival and corneal tumors.

Methods: Impression cytology was used to study of conjunctival and corneal tumors in 45 individuals (19–65 years). From the 45 patients studied 29 had a tumors of melanocytic origin of the conjunctiva, 16 had ocular surface squamous neoplasia (OSSN). Cytological and histological diagnoses were compared in 25 cases. All cytological and histological specimens were examination by two independent investigators. The cytology specimens were obtained using the Biopore membrane device (Biopore Millicell®-CM 0.4mm, PICM01250 MILLIPORE, USA). The specimens were fixed in 95% alcohol, stained with of Schiff's Feulgen and Papanicolaou stains and mounted on the slides for interpretation. The squamous metaplasia cytology was graded according to the classification of Nelson. Cytopathological and histopathological changes were estimated by the differential criteria of atypia cells classified them according to the UICC.

Results: Cytological diagnosis were verified: from tumors of the melanocytic system, benign nevus, 20 cases, proliferation nevus, 3 cases, melanosis, 2 cases, malignant melanoma, 4 cases; from ocular surface squamous neoplasia (OSSN), squamous cell hyperplasia, 5 cases, squamous cell papilloma, 3 cases, dysplasia (mild-grade), 3 cases, dysplasia (high grade), 2 cases, squamous cell carcinoma, 2 cases. In 80 % (20/25) of cases cytological