pathophysiology of aging. In the recent years, PPAR-gamma, activated by J2 series cyclopentenol prostoglandin (cyPGs), was observed to have anti-proliferative, apoptotic, differentiation and anti-inflammatory effects on various cancer cells. The aim of this study is to investigate the cytotoxic and apoptotic effects of CAPE in CCRF-CEM cell line.
**Material and Methods:** The cytotoxicity of CAPE evaluated by Trypan blue dye exclusion test and XTT methods and apoptosis was examined with Acridine orange/Ethidium Bromide dye, Cell death detection ELISA and JC-1 mitochondrial membrane potential assay kit.
**Results:** CAPE had dose and time dependent cytotoxicity and had 10 μM $IC_{50}$ in CCRF-CEM cell line. ELISA and Acridine Orange/Ethidium Bromide dye and JC-1 methods revealed that CAPE induced apoptosis in the cell line. The effect associated with PPAR-gamma and hemeoxygenase is evaluated with Western Blotting. According to the flow-cytometric analysis of JC-1 stain, cell viability is detected as 38.13% for control, 29.78% for $24^{th}$ hour, 26.76% for $48^{th}$ hour, 25.82% for $72^{nd}$ hour and 25.22% for $96^{th}$ hour. It was detected that 10μM concentration caused an increase till $24^{th}$ hour and in the following period it caused a reduction when the effect of CAPE on PPAR-gamma expression was analyzed.
**Conclusion:** Since CAPE is a chemotherapeutic and anti-tumoral agent with very less toxic effects on normal tissues, these results opened a new horizon for use of CAPE in the treatment of acute lymphoblastic leukemia.

**9208** POSTER

**Activity of CKD-581, Histone Deacetylase Inhibitor, in Cutaneous T-cell Lymphoma Models**

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Cutaneous T-cell lymphomas (CTCLs) are characterized by accumulation of malignant T cells in the skin. Early disease resembles benign skin disorders but during disease progression cutaneous tumours develop, and eventually the malignant T cells can spread to lymph nodes and internal organs. Pan-histone deacetylase (HDAC) inhibitors (HDIs), including depsipeptide, vorinostat, and panobinostat, have demonstrated clinical efficacy in CTCL, and vorinostat has been approved and available as treatment for CTCL.
CKD-581, highly water-soluble HDI, developed in our institute has shown a strong cytotoxicity against several cancer cell lines including HCT-116, PC-3, A549 and H460 in previous study.
In this study we investigated the cellular and molecular effects of CKD-581 using both *in vitro* and *in vivo* models of CTCL.
Three cell lines (MJ, Hut78, and HH) were treated with CKD-581, LBH589 and SAHA, for 72 h and the cell viability was quantified using the MTT assay. In addition, western blotting analysis for ac-H3, ac-H4, ac-tubulin, p21 (WAF-1/CIP-1) and p-ERK and caspase 3/7 and 9 assays were performed to verify the associated molecular mechanisms involved in CKD-581 mediated cell death. After 72 h of incubation, $IC_{50}$s of CKD-581 on cell viability test were noted at 0.68 μM, 0.04 μM and 0.1 μM in MJ, Hut-78 and HH cells, respectively and showed a more potent inhibitory activity against human HDAC1, 2, 3, 6 and 8 enzymes than SAHA at single nMs. CKD-581 treatment caused an accumulation of acetylated histones (H3 and H4) and acetylated tubulin and increase of p21 WAF1 and phospho-ERK, and activation of caspase-3/7 and 9 in HH cell.
In a CTCL xenograft mouse tumour model, CKD-581 treatment resulted in a significant tumour regression compared with other HDI. Treatment of CKD-581 (50 mg/kg, b.i.wk, i.p.) caused 49% reduction in the mean tumour volume without a change of body weight while other HDIs showed weak antitumour activities. There was no change in neutrophil counts, but the number of platelets was slightly decreased in CKD-581 treated mice.
These data provide preclinical support that CKD-581 is a promising therapy for CTCL and its strong antitumour activity warrants further clinical investigations.

**9209** POSTER

**Imbalanced Frequency of Regulatory T Cells in Different Subsets of Chronic Lymphocytic Leukemia**

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**Background:** Recent studies have shown the expansion of different subsets of regulatory T cells (Treg) in a variety of autoimmune and malignant diseases indicating their immune regulatory function. There is little data regarding the frequency and the role of Treg cells in hematopoietic malignancies, particularly chronic lymphocytic leukemia (CLL).
**Materials and Methods:** The frequency of $CD4^+CD25^+FoxP3^+$, $CD8^+CD25^+FoxP3^+$ and $CD8^+FoxP3^+$ cells was analyzed in peripheral T cells isolated from CLL patients and 8 normal subjects by flow cytometry. Patients were broadly classified into either progressive (n = 20) and indolent (n = 20), or immunoglobulin heavy chain variable region (IGHV) mutated (n = 24) and unmutated (n = 16) subsets.
**Results:** The frequency of $CD4^+CD25^+FoxP3^+$ cells was significantly higher in progressive (11±0.8% of total $CD4^+$ cells) compared to indolent CLL patients (5.75±0.7, $p < 0.001$) and normal subjects (2.4±0.5, $p < 0.001$). Other subsets of Treg, $CD8^+CD25^+FoxP3+$ and $CD8^+FoxP3^+$ cells were also significantly increased in progressive (4.3±0.44 and 7.67±0.65) as compared to indolent patients (1.65±0.26 and 4.6±0.81, $p < 0.001$ and $p < 0.001$) and normals (0.62±0.13 and 1.51±0.31, $p < 0.001$ and $p < 0.001$), respectively. This difference was also significant when analyzed between indolent patients and normal subjects ($p = 0.03$). No differences, however, were observed between IGHV mutated and unmutated samples in frequency of all subsets of Treg. Furthermore, the frequency of Treg showed no correlation with the prognostic markers CD38 and ZAP70.
**Conclusion:** Our results indicate that progression of CLL is associated with significant increase in circulating Treg cells implying the immune inhibitory function of these cells with subsequent expansion of leukemic cells and disease progression.

**9210** POSTER

**The Efficacy of Anticoagulant Treatment on the Evolution of Thrombotic Complications in Patients With Polycythemia Vera Syndrome**

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**Background:** Polycythemia vera (PV) is a monoclonal myeloproliferative disorder due to the ability of PV erythroid progenitor cells to proliferate in the absence of erythropoietin. One of the most relevant problem of patients with PV is a haemostatic imbalance, resulting in increased risk for thrombotic events. These events have been attributed to quantitative and qualitative abnormalities of red blood cells and platelets arising from the clonal rearrangement of hematopoietic cells, to reduced levels of physiologic anticoagulants (antithrombin III, proteins C and S), and decreased fibrinolytic activity that in part may be secondary to increased plasma levels of plasminogen activator inhibitor 1 (PAI-1).
**The objective** of this study was to investigate the efficacy of anticoagulant treatment in the prevention of thrombotic events, in patients with PV syndrome with or without cardiovascular disease (CVD), by monitoring specific markers of the coagulation profile.
**Material and Methods:**The study comprises 40 patients divided in 2 groups: 20 patients with PV syndrome (PV) and 20 patients with PV with CVD associated (PV+CVD). The patients were tested by determining three factors of coagulation profile: von Willebrand factor, Protein C and PAI-1, before and after administration of anticoagulant therapy. Warfarin® was administrated as anticoagulant treatment, in doses that were adjusted according to the International Normalized Ratio (INR) values.
**Results:**The level of the three studied parameters were found significantly modified in both groups of patients (PV, PV+CVD) ($p < 0.05$). After the administration of anticoagulant treatment, in the first group (PV), it was observed a direct correlation between the treatment and the values of investigated parameters; in this group, the levels of studied parameters returned close to normal values. This correlation was less evident in the second group of patients (PV+CVD), as the monitored values, although lower than the levels at the begining of the treatement, remained within pathologic levels.
**Conclusions:** In PV syndrome the risk for thrombosis is due to endothelial dysfunction and changes in coagulation and fibrinolysis factors. The anticoagulant treatment prevents only partially the occurrence of thrombotic events, but does not completely stop it. In spite of the anticoagulant therapy, the patients with PV presents a high risk for developing thrombotic complications.