By the means of quantitative real-time PCR, we measured the expression levels of 23 genes involved in apoptosis, proliferation and endoplasmic reticulum stress in Jurkat cells RNA, extracted from ionomycin/PMA treated and non-treated cells. The expression data were normalized to the expression levels of four housekeeping genes and cDNA concentration. Our preliminary results show that in the Jurkat cells, in the absence of exogenous SCF (c-kit ligand), ionomycin/PMA treatment down-regulates the expression c-kit receptor and induces the moderate up-regulation of both pro-apoptotic and pro-survival genes. The increased expression of IL-2, NFkB, JNK, ERK, XBP and GADD34 genes, together with the down-regulation of c-kit, show that the ionomycin/PMA treatment induces the proliferation, inflammation and differentiation processes, independently from c-kit activation.
According to our data, the up-regulation of the genes involved in Jurkat cells proliferation and endoplasmic reticulum stress, does not disturb the balance between pro- and anti-apoptotic Bcl-2 family genes upon the ionomycin/PMA treatment.

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**New curcumin analogues show enhanced antitumour activity in malignant melanoma cells**

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**Background**: Malignant Melanoma (MM) is one of the fastest growing cancer in western populations with the incidence having tripled in the last decades. Chemotherapy, immunotherapy and vaccines are still unsatisfactory thus new approaches for MM treatment are urgently needed. Curcumin, a natural spice extracted from the root of *Curcuma longa* L. and largely used in oriental cuisine and medicine, has recently been described as potential anticancer agent. We tested several curcumin-related compounds for their capability to inhibit cell growth on primary MM cell lines.
**Material and Methods**: Viability and antiproliferative assays together with dose and time-response assays have been carried out on MM cell lines to compare antitumour activity of curcumin to that of six related biphenyls. Cultured fibroblasts from healthy donors have been used as controls. DNA fragmentation with ELISA and TUNEL assays have been performed to assess apoptosis triggered by some of the treatments.
**Results**: Curcumin, a natural compound already known for its antitumour activity, showed to be a potent antiproliferative agent on our MM cells. We tested six curcumin-related hydroxylated biphenyls (D2-D7) on MM cells to assess their potential antitumour activity in comparison with that of curcumin: IC50 values established after 5 days of treatments showed the $\alpha,\beta$-unsaturated keton (D6) the most efficient at concentrations around 1−2 $\mu$M, much lower than the IC50 values calculated for curcumin (about 10 $\mu$M). Fibroblasts proliferation rate was not affected in the same conditions. Wash-out experiments further demonstrated that the D6 action was more powerful and rapid in arresting MM cells growth than that of curcumin, giving rise to irreversible effects after only 2−4 hours of co-culture with MM cells. Clonogenic assays were performed to measure long-term effects of D6 on permanent cell growth arrest and cell death, showing a dose-dependent reduction in MM colony formation. ELISA and TUNEL assays on some of the MM cell lines allowed the detection of oligonucleosomes in the cytoplasm and apoptotic bodies in the nucleus, showing involvement of apoptosis in D6 activity.
**Conclusions**: Our results indicate this compound as good lead to develop new therapeutic agents against MM. D6 activity should be further investigated on in vivo melanoma models to assess the real anticancer effectiveness on such tumour.

# Aurora kinase

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**MLN8054, a selective inhibitor of Aurora A kinase: final results of a phase I clinical trial**

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**Background:** MLN8054 is an oral, selective, small-molecule inhibitor of Aurora A kinase. This phase I clinical trial examined the safety, pharmacokinetics (PK), and pharmacodynamics (PD) of MLN8054 administered over two weeks in a 28-day cycle.
**Materials and Methods:** Patients (pts) with advanced solid tumors at 2 centers were enrolled. Cohorts of 3−6 pts received successively increasing doses until dose-limiting toxicity (DLT) was seen in $\geqslant$2 pts. The first 2 cohorts received 10 and 20 mg once daily (QD) on days 1−5 and 8−12. Subsequent cohorts were treated on days 1−14 with 25, 35, 45, 55, 60, 70, and 80 mg/day in four divided doses (QID) with the largest dose at night to mitigate against benzodiazepine-like effects, such as somnolence. Starting at the 45 mg dose level, oral methylphenidate (MP) 5−15 mg was also permitted during daytime dosing. Serial blood samples were collected to estimate PK. Skin and tumor biopsies were obtained before and after dosing to assess accumulation of mitotic cells as a measure of PD effects.
**Results:** Of the 44 pts enrolled, 43 were treated with MLN8054. Pts received a median of 1 cycle (range, 1−10). DLT included reversible Grade 3 benzodiazepine-like effects, primarily somnolence (n = 3), and reversible Grade 3 liver function test (LFT) elevations (n = 2). Dose-escalation was stopped at 80 mg/day because of DLTs of somnolence despite prophylactic therapy with MP (1 pt), and LFT elevation (1 pt). Grade 2 neutropenia and alopecia (1 pt) and mucositis (1 pt) were first observed at the highest dose level of 80 mg. Mean exposure levels were roughly linear with dose. The terminal half-life was 30−40 hours. Among skin biopsies evaluable pre- and post-treatment in 40 pts, there was sporadic evidence of accumulation of mitotic cells in basal epithelium within 24 hours after the first daily dose or at steady-state. Among tumor biopsy specimens evaluable pre- and post-treatment in 14 pts, there was evidence of Aurora A inhibition as measured by multiple mechanistic PD markers, especially at the higher doses.
**Conclusions:** MLN8054 dosing for up to 14 days of a 28-day cycle was feasible. Somnolence and LFT elevation were dose-limiting ahead of clinical anti-proliferative effects. Skin and tumor biopsy findings supported Aurora A kinase inhibition. MLN8054 has been replaced in clinical trials by MLN8237, a more potent second-generation Aurora A kinase inhibitor.

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**Phase I study of the safety, pharmacokinetics (PK), and pharmacodynamics (PD) of MLN8237, a selective Aurora A kinase inhibitor, in the United States**

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**Background:** Preclinical studies suggest the selective Aurora A kinase inhibitor, MLN8237, is more potent than MLN8054 and less likely to cause benzodiazepine-like effects. This ongoing phase I clinical trial examined the safety, PK, and PD of MLN8237.
**Materials and Methods:** MLN8237 was given orally once daily (QD) for 7 days in 21-day cycles. Cohorts of 3 patients (pts) with advanced solid tumors were enrolled to increasing dose cohorts (5, 10, 20, 40, 80, and 150 mg/day) until dose-limiting toxicity (DLT) was seen in $\geqslant$2 of 6 pts. Serial blood samples were collected to estimate PK. PD effects on Aurora A kinase were inferred from accumulation of mitotic cells in the basal epithelial layer of skin biopsies.