Anti-neoplatic and immunosuppressive activity of compounds isolated from the leaves of Artocarpus tonkinensis

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The leaves of *Artocarpus tonkinensis* (*At*) are used in Vietnamese traditional medicine for treatment of arthritis, and the compound maesopsin 4-O-β-D-glucoside (TAT-2), isolated from them, inhibits the proliferation of activated T cells. Our goal was to test the anti-proliferative activity of TAT-2 on the T-cell leukemia, Jurkat, and on the acute myeloid leukemia, OCI-AML. TAT-2 inhibited the growth of OCI-AML (and additional acute myeloid leukemia cells, such as U937, KG-1, and HL-60) but not Jurkat cells. Growth inhibition was shown to be due to inhibition of proliferation and, at a lesser extent, to increase in cell death. Analysis of cytokine release showed that TAT-2 stimulated the release of TGF-β, yet TGF-β neutralization did not reverse the maesopsin-dependent effect.

The drugs currently used for the therapy of AML are anthracycline and cytarabine. In order to compare these drugs with TAT-2, we stimulated OCI-AML cells with sub-optimal concentrations of aracytidine (ARA-C), doxorubicine and TAT-2 alone or in combination. TAT-2 given together with either ARA-c or Doxorubicin significantly decreased the OCI-AML cell number to the same level of ARA-C plus doxorubicin given together. Thus, TAT-2 can significantly increase the effectiveness of the drugs used currently in the AML therapy.

Gene expression profiling determined that Maesopsin modulated 19 identifiable genes. Transcription factor CP2 was the gene most significantly modulated. Real-time PCR validated that up-regulation of sulphiredoxin 1 homolog (SRXN1), hemeoxygenase 1 (HMOX1), and breast carcinoma amplified sequence 3 (BCAS3) were consistently modulated. The role of HMOX1 has been analyzed in depth. It is an anti-oxidant protein that, generally, protects cells from cell death. A western blot analysis confirmed that HMOX1 mRNA was translated in its protein and both TAT-2 and the *At* leave decoction up-regulated its expression when compared to the untreated control. Moreover, ARA-C and doxorubicin, the drugs currently used in the therapy of AML, did not up-regulate the HMOX1 protein. Thus, also the western blot analysis indicate that TAT-2 but not aracytidine (ARA-C) or doxorubicin up-regulates HMOX1, confirming the data of microarray and RT-PCR for this particular gene. To see if HMOX1 was responsible for the TAT-2-dependent growth inhibition, OCI-AML cells were transfected with HMOX1 transgene. Results suggest that overexpression of HMOX1 did not decrease but rather significantly increased OCI-AML cell number, suggesting that HMOX1 overexpression was not responsible for TAT-2 dependent inhibition of OCI-AML cell growth.

Decoction of the leaves of *At* has also been tested for its activity in a model of collagen-induced arthritis in mice. In the thymus of these mice, a subclinical form of arthritis determined a block of differentiation of the step that bring to $CD4^+CD8^+$ double positive (DP) from $CD4^-CD8^-$ double negative (DN) thymocytes. The consequence was a dramatic increase in $CD4^-CD8^-$ DN and a parallel decrease of DP thymocytes. The administration of At decoction completely abrogated the differentiation block.