# CD39+ Treg Cooperate with a CD73-Expressing Th1/Th17 Subset for Adenosine-Mediated Immunosuppression in Human Breast Tumors

**GOURDIN Nicolas**[1,2,3,4] ; RODRIGUEZ C[1,2,3,4] ; DURANDI [4,6] ; JANDUS C[5] ; FAGET J[1,2,3,4] ; ROMERO P[5] ; CAUX C[1,2,3,4*] ; MENETRIER-CAUX C[1,2,3,4*]

1- Team 11, Inserm U1052, Cancer Research Center of Lyon, F-69000 Lyon, France
2- Université de Lyon, F-69000 Lyon, France
3- Université Lyon 1, ISPB, Lyon, F-69622, France
4- Centre Léon Bérard, Lyon, F-69008, France
5- Ludwig Cancer Research Center, Department of Oncology, Faculty of Biology and Medicine, University of Lausanne, Lausanne, Switzerland
6- Cytometry platform, Inserm U1052, Cancer Research Center of Lyon, F-69000 Lyon, France
* Equal contributors
*Contact: nicolas.gourdin@lyon.unicancer.fr*

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Our group and others have previously reported that Treg infiltrating (Ti-Treg) breast tumors have a negative impact on patients' outcome. We further reported their selective recruitment in the tumor environment through CCL22 secretion and expansion upon ICOS engagement by plasmacytoïd dendritic cells.
Herein we investigate the mechanism of Ti-Treg mediated suppression. We observe that Ti-Treg express high CD39 levels. CD39 is an ectonucleotidase that cooperates with CD73 to degrade the danger signal ATP into immunosuppressive Adenosine (Ado). The potency of Ado mediated suppression is illustrated in Adenosine-Deaminase (ADA)-deficient patients unable to degrade Ado and developing severe immunodeficiency.
In this study, we demonstrate that, in contrast to murine Treg, human CD39+Treg do not express CD73 enabling them to degrade ATP into AMP only. Within T cells, CD73 expression was mainly associated with naïve CD8+ T cells and a subset of memory conventional CD4+ T cells (Tconv) that exhibit Th1/Th17 characteristics (CXCR3+CCR6+CCR4negIFN□highIL17+). CD39+ Treg isolated from healthy donor blood, in presence of exogenous ATP, strongly inhibit purified CD73+ but not CD73neg Tconv, proliferation and cytokine production (IFN□, TNF□). The use of enzymatic inhibitors demonstrates the involvement of CD39 and CD73 through Treg/Tconv cooperation for Ado mediated immunosuppression. Of importance when integrated in [Treg -CD4+CD73+] coculture, CD4+CD73neg T cells expressing similar levels of Ado receptors (A2A, A2B) are also inhibited.
Interestingly, we observe that TCR-mediated proliferation of CD73+Tconv leads to CD73 downregulation. Moreover within the breast tumor environment, CD39highTreg and CD73+Tconv counts are positively correlated suggesting Ado-mediated inhibition of CD73+Tconv preventing CD73 downregulation and prolonging suppression.
In conclusion, our findings support the existence of an Ado mediated immunosuppression loop in the tumor through cooperation between CD39highTreg and CD73-expressing Th1/Th17 subsets in the breast tumor environment.
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