



PROCEEDINGS OF THE XXVII NATIONAL CONFERENCE OF CYTOMETRY

Centro Congressi Fiera

Ferrara

14–17 Ottobre 2009

EDITED BY

R. DE VITA and G. MAZZINI

Organizing Committee

F. Lanza (Ferrara)
R. De Vita (Roma)
G. Mazzini (Pavia)
L. Del Vecchio (Napoli)
E. Erba (Milano)
G. Pirozzi (Napoli)

Scientific Coordinators

P. Bianchini (Genova)
R. Casotti (Napoli)
G. Gaipa (Monza)
S. Lucretti (Roma)
S. Pepe (Napoli)
M. Spanò (Roma)
G. Starace (Roma)
L. Zamai (Urbino)

Scientific Board

B. Bacarani (Bologna)
G.P. Bagnara (Bologna)
G. Basso (Padova)
P. Bonara (Milano)
B. Brando (Milano)
A. Calugi (Roma)
S. Capitani (Ferrara)
G. Carandina (Ferrara)
A. Cuneo (Ferrara)
M. Danova (Pavia)
A. Diaspro (Genova)
M. Dominici (Modena)
S. Garattini (Milano)
M. Giroto (Ivrea)
L. Gugliotta (R. Emilia)
A. Kunkl (Genova)

G. Lanza (Ferrara)
C. Ortolani (Venezia)
S. Papa (Urbino)
G. Pizzolo (Verona)
M. Rocchi (Bari)
R. Rossi (Ferrara)
A. Russo (Palermo)
L. Teodori (Roma)
D. Tirindelli (Roma)
C. Usai (Genova)
M. Valentini (Pesaro)
M. Vitale (Parma)
S. Volpe (Avellino)

SOCIETÀ ITALIANA DI CITOMETRIA
c/o Unità Tossicologia e Scienze Biomediche
ENEA Centro Ricerche Casaccia s.p. 016
Via Anguillarese, 301 - 00123 Roma
tel.: 06 30484671 fax: 06 30484891
e-mail: devita@enea.it
<http://biotec.casaccia.enea.it/GIC/>

SUPPORTED BY ENEA - ENTE PER LE NUOVE TECNOLOGIE, L'ENERGIA E L'AMBIENTE

XXVII National Conference of the Italian Society of Cytometry GIC

October 14—17, 2009

Ferrara - Italy

Following the first experience in 2005, also this year an issue of Cytometry is partly dedicated to the programme and abstracts of the National Conference of the Italian Society of Cytometry, GIC. The XXVII edition of the Conference has been organized in October 2009 in Ferrara City, Italy. From 1995 on, UNESCO has included the historical centre of Ferrara in the list of World Cultural Heritage as a wonderful example of a town planned in the Renaissance and still keeping its historical centre intact. Its beauty has been linked to one of the most important courts in the political scenario of the 15th-16th century: the Estense court, which was one of the major actors in that precious season we call the Renaissance period.

As far as the GIC meeting is concerned, we want to stress the fact that all abstracts were carefully reviewed by the Scientific program Committee and published here in full and categorized by scientific track (1. cell cycle and apoptosis; 2. environmental sciences and toxicology, 3. hematology, 4. immunology, 5. methodology and technology, 6. oncology).

Following a continuous growth in these years, to date there are over 850 members actively involved in educational programs, promotion of quality controls programs, drafting/validation of guidelines and accreditation, providing information for people involved that actively work in the field of basic and applied cytometry.

This year, a great number of abstracts (>100) have been selected by the Scientific Committee among those submitted by basic and clinical researchers operating in the various Italian Institutions.

Each session involved invited lectures and was focused on the emerging role of cytometry techniques in Hematology, Stem Cell Biology, Immunology, Oncology and Environmental Sciences and Toxicology.

In addition, different topics of general interest in biological and medical sciences, new data on the study of a rare disease such as PNH, accreditation, standardization of ZAP70 measurement across Italy, and on the Methodological and Technological advances were reviewed by experts from Italy. Two of these lectures were dedicated to the loss of two “top” scientists, Prof Bruno Rotoli (Naples) and Prof Antonio Tabilio (Perugia). Both of them tirelessly helped young researchers and research students, and they were active in disseminating research findings to and communicating with the public. We do all miss them!

The Conference had been also characterized by a round table dealing with the possible interactions between parental scientific Societies having different levels of interest in cytometric techniques and applications. Since many years ago the GIC Society did promote such kind of scientific interactions.

A substantial contribution was obtained from the principal industries in the field that have been located in a large exhibition area inside the conference center.

This national event is growing each year and, once again, represents Italian cytometry's scientific contribution to the international community.

Guest Editors:
R. De Vita - G. Mazzini

Francesco Lanza
GIC President

not only produce Th1/Th17 cytokines, but express a number of homing and chemokine receptors, thus being equipped for recirculation through lymph nodes and peripheral tissues.

PRODUCTION OF SOLUBLE HLA-G MOLECULES BY MESENCHYMAL STROMAL CELLS AFTER IN VITRO IL-10 ACTIVATION: A MARKER FOR "A PRIORI" EVALUATION OF THEIR IMMUNOREGULATORY ACTIVITY

Rizzo R.,² Campioni D.,¹ Stignani M.,² Lanzoni G.,³ Melchiorri L.,² Bonsi L.,³ Alviano F.,³ Costa R.,³ Ricci F.,⁴ Tazzari PL.,⁴ Cuneo A.,¹ Bagnara GP.,³ Baricordi OR.,² and Lanza F.¹

¹Department of Biomedical Sciences and Advanced Therapies, Hematology Section, Azienda Ospedaliera-Universitaria Arcispedale S. Anna, Ferrara, Italy

²Department of Experimental and Diagnostic Medicine, Laboratory of Immunogenetics, Section of Medical Genetics, University of Ferrara, Italy

³Department of Histology, Embryology and Applied Biology, University of Bologna, Stem Cell Research Centre, University of Bologna, Italy

⁴Sant'Orsola-Malpighi Hospital, Service of Blood Trasfuzion (Bologna), Italy
rbr@unife.it

Graft versus host disease (GvHD) is the main unfavorable evolution of allogeneic hematopoietic cell transplantations (HSCT). Even though GvHD is now controlled by pharmacologic treatment, recent studies have proposed a beneficial effect of mesenchymal stromal cell co-transplantation (MSCs). These cells are able to inhibit the innate and adaptive cell-mediated immune response with a variable efficacy between MSCs from different subjects. For this the availability of markers of MSC inhibitory activity would be of extreme interest in HSCT. Several soluble factors have been recognized as responsible of MSC immunoregulation. In our study we have evaluated if HLA-G molecules could be implicated in MSCs functions. HLA-G are non-classical HLA class I molecules implicated in the immune response, inhibiting T CD8+, T CD4+, natural killer, B and dendritic cell activation.

By flow cytometric analysis and immunosorbent assay, we have analyzed the production of membrane-bound and soluble HLA-G by MSCs after IL-10 treatment.

The bone marrow derived (BM) MSCs with or without rIL-10 treatment have been analyzed in particular for IL-10R1 expression by flow cytometry with anti-IL-10R1 MoAb. The rIL-10 treatment has increased IL-10R1 expression ranging from 12.0 and 58.3% with a mean value of 32.9%. To

confirm that rIL-10 treatment is involved in IL-10R1 up-modulation the MSC cultures have been pre-treated with an anti-IL-10R1 MoAb. By flow cytometric analysis we observed that this pre-treatment has significantly reduced the up-regulation of IL-10R1, membrane HLA-G1 expression and sHLA-G secretion (ranging from 0.0 and 3.1 ng/ml).

In conclusion, our data demonstrate the role of sHLA-G molecules in the immunoregulatory effect of MSCs. The *in vitro* treatment with IL-10 induces different levels of HLA-G secretion by MSCs which seems to be a marker of MSC functionality.

EARLY CD4+ LYMPHOCYTE RECOVERY CORRELATES TO CLINICAL OUTCOME AFTER ALLOGENEIC HEMATOPOIETIC STEM CELL TRANSPLANTATION

Spiniello E., Fedele R., Garreffa C., Dattola A., Princi D., Imbalzano L., Andidero P., Moscato T., Irrera G., Console G., Messina G., Martino M., Massara E., Cuzzola M., and Iacopino P.

Centro Trapianti Midollo Osseo Az. Osp. B.M.M., Reggio Calabria, Italy
elisa_spiniello@hotmail.it

Recent reports suggested that early CD4+ cell recovery after allogeneic stem cell transplant (SCT) has a strong impact on acute graft versus host disease (aGVHD), overall survival (OS), transplant-related mortality (TRM). We evaluated CD4+ cell count at 20 days after SCT (r. 12-34) on 99 patients (pts), with a median age of 46 years (r. 11-67), underwent to bone marrow (23 pts) and peripheral blood (76 pts) SCT. The median follow-up was 46 months (r. 12-86). Donors were 83 matched sibling and 16 alternative. Conditioning regimens were myeloablative (48 pts) or at reduced intensity (51 pts). The incidence of aGVHD (grade II-IV) was 44%. Univariate analysis showed that early CD4+ cell recovery is correlated with OS and TRM but not with aGVHD. Roc curve of CD4+ cell count indicated that the cut-off was 115/ μ l. At 2 years follow-up, pts achieving this cut-off showed significantly lower cumulative TRM respect on pts who did not. At 5 years, OS was better in pts with more than 115 CD4+/ μ l, respect on pts with less. We evaluated, with multivariate analysis, the predictive role of other factors associated to OS as donor type and sex, ABO identity, recipient sex and age, stem cell source, conditioning regimen, disease type and status and we found that the main predictive factor for clinical outcome after allogeneic SCT is represented by early T helper count. Patients with low early CD4+ count need to be followed more carefully to avoid transplant complications. The graft manipulation may represent an opportunity to obtain an improvement in early immune recovery and overall survival.

METHODOLOGY AND TECHNOLOGY

MICROSCOPIC EVALUATION OF PHAGOCYTIC ACTIVITY OF HUMAN MACROPHAGES AGAINST ASPERGILLUS CONIDIA AFTER IMMUNO-STIMULATING TREATMENT

Andreola F.,¹ Psaila R.,¹ Zonfrillo M.,¹ Mercuri L.,¹ Moroni N.,¹ Gaziano R.,² Sinibaldi-Vallebona P.,² Piermarchi P.,¹ and Serafino A.¹

¹Institute of Neurobiology and Molecular Medicine (INMM-ARTOV), CNR, Rome, Italy

²Department of Experimental Medicine and Biochemical Science, Univ. of Rome "Tor Vergata", Italy
federica.andreola@artov.inmm.cnr.it

Aspergillus species are recognized as major fungal pathogens in severely immunosuppressed or neutropenic patients, in which invasive pulmonary aspergillosis (IPA), characterized by hyphal invasion and destruction of pulmo-