The following relationships exist related to this presentation:

No relationships to disclose.
LOSS OF HLA-DR EXPRESSION ON CD14+ CELLS; A COMMON MARKER OF IMMUNOSUPPRESSION IN CANCER PATIENTS

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Mayo Clinic
2010 iSBTC Annual Meeting
Assessing patient immunity by peripheral blood immunophenotyping

- Flow Cytometry
- Whole blood
- Broad unbiased approach
  - > 40 phenotypes
- Patients: prior to therapy or
  - > 8 weeks off treatment

**Diseases**
- Glioma Stage IV (GBM) n=50
- Prostate Cancer n=40
- Non-Hodgkin’s Lymphoma (NHL) n=40
- Clear Cell Renal Cell Carcinoma (RCC) n=23
- Chronic Lymphocytic Leukemia (CLL) n=29
- Patients at risk for sepsis n=29
- Diabetes and ALS n=11

**TOTAL=193 patients**

<table>
<thead>
<tr>
<th>Lymphocytes</th>
<th>T cells</th>
</tr>
</thead>
<tbody>
<tr>
<td>T helper/T killer subsets</td>
<td></td>
</tr>
<tr>
<td>T regulatory cells</td>
<td></td>
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<tr>
<td>Costimulatory/inhibitor receptors</td>
<td></td>
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<tr>
<td>Activated T cells</td>
<td></td>
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<tr>
<td>Central and effector memory</td>
<td></td>
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<tr>
<td>Naïve T cells</td>
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<tr>
<td>B cells</td>
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<tr>
<td>Naïve and Mature B cells</td>
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<tr>
<td>Transitional B cells</td>
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<tr>
<td>Plasma cells</td>
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<td>Isotyped switched</td>
<td></td>
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<tr>
<td>NK cells</td>
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<tr>
<td>NKT cells</td>
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**Myeloid cells**

<table>
<thead>
<tr>
<th>Monocytes</th>
<th></th>
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<tbody>
<tr>
<td>M1 and M2 (CD14+ vs CD16+)</td>
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<tr>
<td>HLA-DR</td>
<td></td>
</tr>
<tr>
<td>Co-stimulatory receptors</td>
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<tr>
<td>Signaling receptors</td>
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</table>

<table>
<thead>
<tr>
<th>Granulocytes</th>
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</thead>
<tbody>
<tr>
<td>CD15+CD66b+</td>
<td></td>
</tr>
<tr>
<td>Myeloid Derived Suppressor cells</td>
<td></td>
</tr>
<tr>
<td>Classical Lin-HLA-DR-CD33+</td>
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<tr>
<td>“Granulocytic” CD15+CD14-</td>
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</tr>
</tbody>
</table>
Generating “Immune Profiles” using bioinformatics

Phenotypes converted to cells/uL

Data analyzed using Partek

Each sphere representing individual immune profiles

Clustering identifies relationships

Provides ‘unbiased’ method to look for phenotypes of interest
CD14<sup>+</sup>HLA-DR<sup>lo/neg</sup> monocytes profoundly change the immune profile of patient PBMCs.

With enough disease profiles, average profile of patients can be determined.
CD14^+HLA-DR^{lo/neg} monocytes are elevated in cancer patients.
**CD14\(^+\)HLA-DR\(^{lo/neg}\) monocytes mechanisms of immunosuppression**

Inhibition of T cell proliferation

Unable to fully differentiate into mature DCs


Lin Y, Gustafson MP, et al, Submitted manuscript.
CD14^+ HLA-DR^+ Monocytes

CD14^+ HLA-DR^+ → Mature DC → Responder T cells

CD14^+ HLA-DR^{lo/neg} Monocytes

CD14^+ HLA-DR^{lo/neg} → Mature DC → Responder T cells
CD14<sup>+</sup>HLA-DR<sup>lo/neg</sup> in other clinical settings

- Found in melanoma, ovarian cancer, and hepatocellular carcinoma.
  - Valenti R, Cancer Res. 2006;

- Associated with sepsis, acute pancreatitis, liver failure, burns, and trauma.

- Correlated with survival in sepsis.
  - Cheadle WG, Am. J. Surg. 1993; Wakefield CH, et al,
  - Br. J. Surg. 1993; van den Berk JM, et al,
Loss of HLA-DR on CD14+ cells is a prognostic factor in cancer patients

GBM

NHL

N=40

N=25

Percent survival

Overall survival (percent)

Days

Time (days)

p=0.0177

p = 0.13

Normal HLA-DR + 1 other phenotype

Abnormal HLA-DR + 1 other phenotype
Summary

- Immunophenotyping by flow cytometry and multiparameter analysis will continue to be extremely important in characterized baseline immunity in patients.
- Bioinformatics approach is likely to yield new relationships between immune cells.
- CD14$^+$HLA-DR$^{lo/neg}$ monocytes are elevated in every cancer type that we’ve analyzed.
- CD14$^+$HLA-DR$^{lo/neg}$ monocytes inhibit T cell proliferation and cannot fully mature into potent DCs.
- The combination of CD14$^+$HLA-DR$^{lo/neg}$ monocytes and other phenotypes are prognostic; independent of therapy.
Implications

- The presence of CD14\textsuperscript{+}HLA-DR\textsuperscript{lo/neg} monocytes may identify potential responders/non-responders on patients receiving vaccines or other immunotherapeutic approaches.

- Mechanisms of immunosuppression/immunoparalysis are very similar in cancer patients and in infection/sepsis patients.
Acknowledgments

Human Cellular Therapy Lab

- Dr. Allan Dietz
  - Scientific Director
- Dr. Dennis Gastineau
  - Medical Director
- Peggy Bulur
- Mary Maas

Other collaborators:
- Dr. Yi Lin
- Dr. Roshini Abraham
- Dr. Stanimir Vuk-Pavlovic
- Dr Eugene Kwon
- Dr. Clive Zent
- Dr. Jann Sarkaria
- Dr. Brian O’Neill
- Dr. Ian Parney
- Dr. Kent New
- Dr. Thomas Witzig