Columbia University Opportunity Pool Project: Elucidating the origin and development of bladder resident macrophages

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Introduction

Macrophages are part of the phagocyte mononuclear system constantly replaced by the circulating blood monocytes. In the steady-state, the myeloid cell compartment is highly heterogenous, and contains cells of different origins and functions. These cells include macrophages and dendritic cells, each playing important roles in tissue maintenance, including development, homeostasis, immunity and repair following tissue injury.

There have been pivotal studies that have proposed and demonstrated that tissue macrophages originate in part from mesodermal erythro-myeloid progenitors from the yolk sac and it has been suggested that the embryonic derived tissue macrophages represent a founding cell type within organ anlagen. These resident macrophages are conserved independently from conventional hematopoiesis by self-renewal and/or longevity.

Macrophages reside in the bladder and are recruited during urinary tract infections. The bladders' primary function is to store urine before voiding and to protect the host from toxins that accumulate in the urine, inhibit the invasion of micro-organisms and removal of cellular debris. The multi-functions of the bladder will require this organ to be immunologically unique. Presently, there is limited data on the origins of bladder macrophages, nor a complete understanding of bladder macrophages in the steady state. Understanding tissue specific immunity in the steady state is essential to fully understand immune responses to immunological insults such as infection, in the case of the bladder urinary tract infections.

Aims

Deep phenotyping of the bladder macrophage population.
Utilizing high dimensional flow cytometry, we will identify resident and recruited macrophage and monocyte populations in the steady state during the development of C57BL/6J mice.

Hypothesis: Immunological markers will identify distinct macrophage populations in the bladder and demonstrate how these populations vary by age and sex.

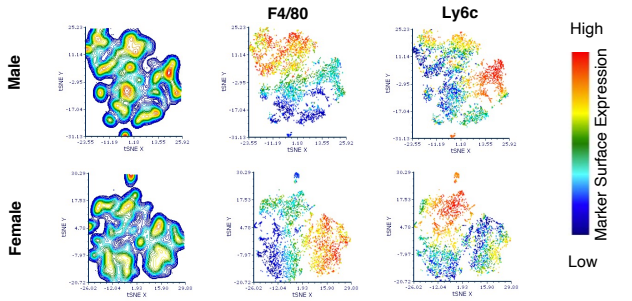
Methods

- **Bladders isolated from C57BL/6J mice aged 7-, 21-, and 84 days were disassociated into a single cell suspension.**
- **A multiparameter flow cytometry approach was used to deep phenotype the bladder macrophage populations.**
- **Flow cytometry markers:**

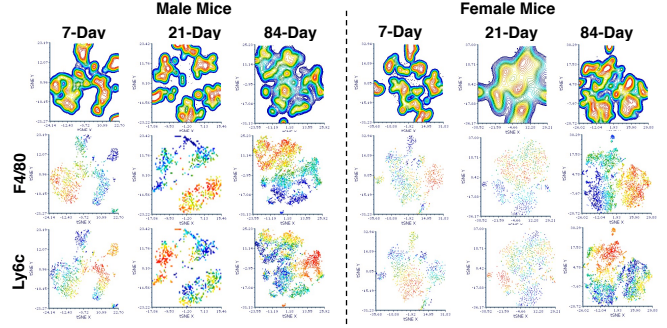
  Arg1, CD103, CD115, CD11b, CD16.2, CD163, CD192, CD206, CD3, CD43, CD45, CD64, CD68, CD80, CD86, CD90, Cx3cr1, F4/80, Fizz1, I-A/I-E (MHC class II), IL-10, IL-12, IL126, Ki-67, Ly6C, Ly6G, Siglec F, Tim4, TLR2, TLR4, Live/Dead

Results

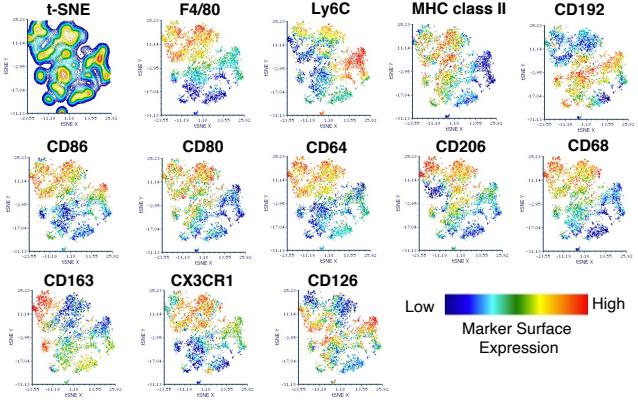
Multiple resident macrophage populations reside in the bladder in male and female adult mice



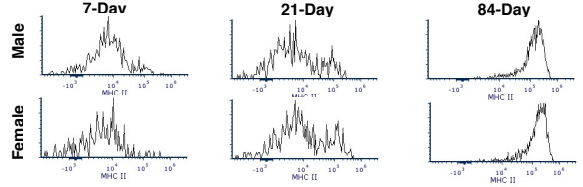
Multiple macrophage populations reside in the bladder in male and female young and old mice



Macrophage populations that reside in the bladder display different cell surface markers in adult mice



Induction of MHC class II on F4/80^{high} bladder macrophages



Conclusions

The data indicate a dynamic change in the bladder macrophage population, leading to an activated resident macrophage phenotype.
The composition of macrophage populations also differs by sex and age. These data suggest each population plays a role in bladder homeostasis.
Future studies will be directed towards elucidating the functions of each of the identified macrophage populations.

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