

Macrophages, apoptotic cells and cholesterol—strategies for survival: an interview with Dr. Ira Tabas

Helene F. Rosenberg¹

Laboratory of Allergic Diseases, National Institute of Allergy and Infectious Disease, National Institutes of Health, Bethesda, Maryland, USA

The manuscript “Macrophages become resistant to cholesterol-induced death after phagocytosis of apoptotic cells” was selected as a Pivotal Advance because it addresses novel mechanisms of macrophage function, specifically related to their role in atherogenesis. Of particular interest, this work elucidates several mechanisms via which macrophages survive despite having ingested massive amounts of cholesterol.

Dr. Tabas, to begin, what exactly led you to study the effects of cholesterol on macrophages? How important is cholesterol and its effects on macrophages to the process of atherogenesis *in vivo*?

IT: At the time when I was being introduced into the atherosclerosis field, which was in the early 1980s, the major cell type in atherosclerotic lesions had been definitively identified through the use of newly-developed monoclonal antibodies as macrophages. For me, this provided an opportunity to become a macrophage biologist with a direct application to the disease I wanted to study. The defining characteristic of macrophages in atherosclerotic lesions, as opposed to macrophages in other tissues or pathologies, is the fact that they are capable of accumulation of tremendous amounts of cholesterol which is acquired from the uptake of lipoproteins from the arterial wall.

How important is cholesterol accumulation *in vivo* to the process of atherogenesis? That is an incredibly fundamental but, surprisingly, not very well-resolved issue. Certainly, cholesterol and its oxidized metabolites have profound effects in cultured macrophages. Moreover, genetic manipulations in mice have shown that promotion of cholesterol efflux from macrophages decreases atherosclerosis, and vice versa. In terms of our specific model of cholesterol-induced macrophage death, which depends upon cholesterol trafficking to the endoplasmic reticulum and triggering of the Unfolded Protein Response (UPR), we published a paper in 2003 [1] in which we showed that blocking cholesterol trafficking to the endoplasmic reticulum (ER) *in vivo* also blocked macrophage death and plaque necrosis in advanced atherosclerotic lesions in a widely used model of mouse atherosclerosis, the ApoE knockout mouse. These findings are consistent with an important role for ER cholesterol in macrophage death in advanced atheromata, at least in this specific mouse model.



Dr. Ira Tabas received his M.D. and Ph.D. degrees from the joint program at Washington University in St. Louis, and did residency and fellowship training at Columbia University in New York. Dr. Tabas is currently Professor of Medicine and Anatomy and Cell Biology at Columbia University and Attending Physician of Medicine at Columbia-Presbyterian Medical Center. Dr. Dongying Cui, the first author on the manuscript, is a post-doctoral fellow in Dr. Tabas' laboratory. She graduated from Pennsylvania State University College of Medicine with a Ph.D. in Physiology.

In this manuscript, you and your colleagues demonstrate that cultured mouse peritoneal macrophages can initiate several survival responses when confronted with cholesterol-laden apoptotic cells (ACs). However, it seems as though only a fraction of cultured macrophages actually take up these ACs. Do you have any understanding of the features of macrophages that permit phagocytosis of cholesterol-loaded ACs to take place?

IT: It is intriguing that only ~30% of the macrophages in culture take up cholesterol-loaded ACs. We have investigated this observation, asking whether there is a defined macrophage subset that is capable of uptake, or whether the system is more dynamic in nature. To do this, we evaluated responses of

¹ Correspondence: hrosenberg@niaid.nih.gov
doi: 10.1189/jlb.1307192

cultured macrophages to both primary and secondary challenges with differentially-labeled apoptotic cells. Interestingly, secondary challenge resulted in uptake by some cultured macrophages that had been resistant to primary challenge, and, conversely resistance by some macrophages that had taken up apoptotic cells at primary challenge. Given these mixed results, we cannot conclude definitively that there are intrinsic differences between macrophages in culture. However, there may be intrinsic differences that are then complemented by cell-to-cell interactions or secretory phenomena resulting from the primary challenge.

Have any of the macrophage survival responses reported here been explored in vivo?

IT: A comprehensive picture of how macrophages handle different types of phagocytosed material is part of a new and emerging field of study, and we certainly do not understand how all of this works in vivo. Despite the fact that macrophages in atherosclerotic lesions ingest massive amounts of cholesterol, we do in fact find macrophages that are very much alive in atheromata. In human atheromata, cholesterol-filled macrophages are found in regions surrounding dead cells in advanced lesions. Could these macrophages have been "given a second life" by ingestion of apoptotic cells, as suggested by the findings we present here? We see no reason to rule out this mechanism, but further studies will be needed to prove that it is operative in vivo.

In a recent review article [2], you discussed the possibility of searching for therapeutics based on macrophage apoptosis. Of the various strategies considered, what do you see as the most likely to be effective?

IT: Although we discussed a number of different therapeutic strategies in this review, the approaches that make the most sense overall are those that enhance efferocytosis, or clearance of apoptotic cells. These strategies would be quite beneficial in advanced lesions, particularly if factors that inhibit phagocytosis of apoptotic cells could be identified and eliminated. Indeed, the long range clinical implications of the findings presented in this manuscript relate to the fact that inducing engulfment of cholesterol-loaded apoptotic macrophages in lesions would not simply perpetuate a cycle of further macrophage death in the phagocyte itself.

On a more personal note, can you tell us what motivated you to become a physician/scientist?

IT: I was originally interested in more basic science, particularly cell biology, but as time went on, I increasingly became intrigued with the possibility of applying insights from basic research to human disease. Regarding my interest specifically

in atherosclerosis—my father died at a very young age of a heart attack and that inspired me to devote myself to research in this area.

In your career as a physician and a scientist, you have clearly determined how to strike an appropriate balance between clinical responsibilities and scientific pursuits. Do you have any advice to young physician/scientists as to how to deal with these issues most effectively?

IT: As one of the associate directors of the MD/PhD program at Columbia, I am frequently called upon to discuss just these issues. My advice is quite simple. It is important to make the right choices and to keep research as the most important focus. As an example from my own life—despite the fact that I study atherosclerosis, I decided to turn down a cardiology fellowship, because the time spent on clinical emergencies and procedures would have been too de-focusing for me. Instead, I opted for a fellowship in endocrinology, which always fascinated me and, in my outlook, was more compatible with a career in basic and translational research. I enjoy serving as a clinician, attending and participating in teaching rounds in internal medicine and endocrinology, but in a structured, time-controlled fashion. To make this work, one simply has to understand that the time and infrastructure for research needs to be an absolute priority.

What are some of the things you enjoy doing other than clinical work and research?

IT: I enjoy keeping physically fit, I'm an avid skier and gardener, and enjoy spending time with my family. I also do quite a bit of outside reading, with a particular interest in history.

Is there anything else you would like to add to this interview?

IT: I would like to acknowledge the contributions of Dr. Dongying Cui, the first author on this publication. Dr. Cui is a wonderful postdoctoral fellow who deserves tremendous credit for this work. Her perseverance on this project is what ultimately led to our success, given the complexities and challenges presented by the experiments featured in this manuscript.

REFERENCES

1. Feng, B., Zhang, D., Kuriakose, G., Devlin, C. M., Kockx, M., Tabas, I. (2003) Niemann-Pick C heterozygosity confer resistance to lesion necrosis and macrophage apoptosis in murine atherosclerosis. *Proc. Natl. Acad. Sci. USA* **100**, 10423–10428.
2. Tabas, I. (2005) Consequences and therapeutic implications of macrophage apoptosis in atherosclerosis. The importance of lesion stage and phagocytic efficiency. *Arterioscler. Thromb. Vasc. Biol.* **25**, 2255–2264.