An apolipoprotein A-I mimetic targets scavenger receptor A on tumor-associated macrophages

A prospective anticancer treatment?

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Keywords: scavenger receptor, tumor-associated macrophage, ligand, apo A-I mimetic peptide, co-culture

Abbreviations: SR, scavenger receptor; TAM, tumor-associated macrophage

Tumor-associated macrophages (TAMs) constitute attractive therapeutic targets because they are a constant part of the tumor microenvironment in many distinct types of malignancy.1 We and others have previously shown that tumor cells can promote the switch of macrophages toward an anti-inflammatory M2 phenotype that is characterized by the expression of scavenger receptor A (SRA).2,3 We established a co-culture model to probe the crosstalk between macrophages and cancer cells for pro-oncogenic cues from either interaction partner. Interestingly, the expression of SRA on TAMs appeared to be necessary for tumor cell invasion in co-culture experiments as well as in vivo, since Msr1−/− mice (lacking SRA upon gene knockout) failed to develop metastases.4

Two hypotheses could be put forward to explain the involvement of SRA in tumorigenesis (Fig. 1): TAMs might use SRA to downregulate inhibitory signals in the tumor microenvironment or to trigger activatory pathways, resulting in release of tumor-promoting mediators. In either case, the identification of an SRA ligand in co-culture experiments was expected to provide mechanistic clues. We therefore screened culture supernatants for SRA ligands and identified both proteins and lipids that are selectively produced by co-cultures but not when TAMs and cancer cells are grown separately.

Several protein ligands that we identified are components of the extracellular matrix, a promising finding since scavenger receptors (SRs) are known to interact with modified collagens and extracellular proteoglycans. The repeated incubation of co-culture supernatants with wild-type (WT), but not Msr1−/−, macrophage monolayers depleted the ligand, supporting the hypothesis of SRA-mediated scavenging. Another SR has previously been implicated in clearing the way for migrating tumor cells: stabilin 2 (STAB2, also known as FELL). STAB2 efficiently removes hyaluronic acid from the circulation, and the loss of STAB2 has been shown to suppress metastatic spread in a mouse tumor model.5 If a similar extracellular matrix clearance mechanism proved to underlie the pro-metastatic functions of SRA, it will be worthwhile to investigate inhibitors that would simultaneously target multiple SRs.

The de-lipidation of co-culture supernatants also reduced SRA ligand activity. SRA is able to interact with a range of modified lipids and lipoproteins, and pro-inflammatory lipids not only stimulate tumor growth but also constitute predictive biomarkers, at least in some settings. In the future, it will hence be interesting to characterize the SRA-binding sites of tumor cells and cancer cell co-cultures. This will provide further insights into how bioactive lipids drive the TAM-tumor crosstalk.

We used known ligands to interfere with SRA function, and were able to block tumor cell invasiveness in vitro. This prompted us to test the therapeutic benefits of a small peptidic inhibitor of SRA, 4F, in vivo. This d-amino acid peptide, which is known to block SRA-mediated macrophage adhesion, prevented the progression and metastatic spread of two murine cancer cell lines, ovarian ID8 and pancreatic Panc02 cells, similarly to the complete absence of SRA. Since no additive effects were observed when 4F was administered to Msr1−/− mice, we believe that 4F and SRA operate in the same signaling pathway.

Initially designed to improve cholesterol homeostasis and to prevent atherogenesis through its antioxidant and anti-inflammatory properties, the apolipoprotein A-I (APOA1) mimetic peptide

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Submitted: 03/20/13; Accepted: 03/26/13
Citation: Neyen C, Mukhopadhyay S, Gordon S, Hagemann T. An apolipoprotein A-I mimetic targets scavenger receptor A on tumor-associated macrophages: A prospective anticancer treatment? OncoImmunology 2013; 2:e24461; http://dx.doi.org/10.4161/onci.24461
4F is emerging as a multi-faceted antican-
cer therapeutic. A study by Su et al. first
described the direct antineoplastic effects of
4F against mouse and human ovar-
ian cancer cell lines in vitro. The same
authors succeeded in reducing the growth
of ID8 ovarian cancer cells in mice, hence
improving the survival of tumor-bearing
animals.6 The proposed mechanism of
action hinges on the ability of 4F to reduce
the circulating levels of lysosphos-
phatidic acid (LPA), similar to overexpressed
human APOA1. Pro-inflammatory and
pro-angiogenic lysophospholipids such
as LPA have been repeatedly associated
with tumor progression and poor prog-
nosis, and are normally cleared from the
serum by APOA1, which is downregu-
lated in ovarian, gastric and pancreatic
cancer patients. In a subsequent study the
same group demonstrated that the LPA-
lowering, antitumor properties of 4F are
shared by other apolipoprotein mimetics
and can be observed in murine models
of both induced and spontaneous colon
cancer.7

A follow-up paper further unrav-
ellled the mechanism of action of 4F and
proposed that 4F-dependent oxidative
changes in cancer cells may be responsible
for its therapeutic effects.8 4F-treated ID8
cells upregulated the manganese-cont-
taining superoxide dismutase (MnSOD),
resulting in lower levels of oxidative stress
and oxidative damage to macromolecules
in the tumor microenvironment. In vivo,
ID8 cells depleted of MnSOD became
unresponsive to 4F, proving that one 4F
exerts antineoplastic effects, at least in
part, by modulating the oxidative status of
cancer cells.

With regards to our study, it will be
interesting to investigate whether APOA1,
a known SRA ligand, is indeed cleared from
the tumor microenvironmetn by SRA, and
whether this drives the local accumulation
of LPA. As noted above, the SRA ligand
activity generated by TAM/cancer cell co-
cultures decreased upon repeated passag-
ing on macrophage monolayers, which is
suggestive of scavenging activity, and we
also detected lipid ligands in co-culture
supernatants. Whether these two observa-
tions are mechanistically linked remains
to be determined.

Oxidative stress leads to the modifica-
tion of multiple macromolecules, includ-
ing proteins (carbonylation) and lipids
(peroxidation), thereby generating poten-
tial ligands for SRs. The ability of 4F to
exert antioxidant effects in cancer cells
may reduce the availability of both SRA-
specific and less specific SR ligands in the
tumor microenvironment, another plau-
sible connection to explore.

From a therapeutic standpoint, the
administration of 4F seems flexible: Su
et al. reported that 4F was active regard-
less of oral or subcutaneous delivery, and
although injection resulted in higher
plasma levels than ingestion, both routes
were efficient in lowering circulating
LPA in mice. Bioavailability and safety
tests in humans showed that oral, sub-
cutaneous and intravenous 4F is safe and
well-tolerated.9,10 Nevertheless, so far the
anti-inflammatory effects on circulating
serum lipids observed in 4F-treated mice
could not be replicated in humans.9 An
increasing number of studies reports anti-
neoplastic effects for 4F, raising the urgent
need to fully understand how 4F affects
tumor progression in order to best deploy
its therapeutic benefits.

Disclosure of Potential Conflicts of Interest
No potential conflicts of interest were
disclosed.

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Figure 1. Therapeutic effects of the scavenger receptor A inhibitor 4F: (A) The co-culture of
tumor-associated macrophages (TAMs) and cancer cells results in the production of unidentified
scavenger receptor A (SRA) ligands, which promote the TAM/cancer cell crosstalk and metastasis. (B) SRA
on TAMs may scavenge extracellular matrix (ECM) components, thereby releasing cancer
cells from their primary site and enabling metastasis. 4F might interfere with this interaction
and hence, similar to the loss of SRA, prevent tumor cell egress. (C) Lysosphosphatidic acid (LPA)
stimulates the proliferation and migration of cancer cells, and is scavenged from the circulation
by apolipoprotein A1 (APOA1) and/or APOA1 mimetic peptides (such as 4F). By interacting with
APOA1, SRA might promote the local release of LPA.

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