Inflammation and Cancer

Inflammation and Cancer: An Ancient Link with Modern Evidence

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Inflammation and Cancer

INFLAMMATION AND CANCER

Inflammation & cancer = 37,201 articles published in the last 10 years

In 1863 Rudolf Virchow noted leucocytes in neoplastic tissues and made a connection between inflammation and cancer.

Recent Evidence have supported Virchow’s hypothesis, and the links between inflammation and cancer may have potential implications in prevention and treatment.

Balkwill & Montovani; Lancet, 357; 2001
Aspirin Use

Aspirin Use Reduced Risk of Cancer Deaths

Rothwell PM et al., Lancet, 377, 2011
Chronic Inflammation and Infection

CHRONIC INFLAMMATION AND INFECTION CAN INCREASE CANCER RISK

<table>
<thead>
<tr>
<th>Inherited</th>
<th>Acquired</th>
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<tbody>
<tr>
<td><strong>Disease</strong></td>
<td><strong>Tumor Site</strong></td>
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<tr>
<td>Hemochromatosis</td>
<td>Liver</td>
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<tr>
<td>Hereditary Pancreatitis</td>
<td>Pancreas</td>
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<tr>
<td>Ulcerative Colitis</td>
<td>Colon</td>
</tr>
<tr>
<td>Crohn’s Disease</td>
<td>Colon</td>
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<tr>
<td><strong>Parasitic</strong></td>
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<tr>
<td>S. hematobium</td>
<td>Urinary Bladder</td>
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<tr>
<td>S. japonicum</td>
<td>Colon</td>
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<tr>
<td>Liver Fluke</td>
<td>Liver</td>
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“18% of human cancers, i.e., 1.6 million per year, are related to infection.”

Stress and Inflammation

Stress

Adaptive Immune Response

Innate Immune Response

Inflammation

Epithelium

Adaptive Immune Response

Stroma

Cancer

DNA Damage

Protein Modification

Proliferation

miRNA Expression

DNA and Histone Methylation

Angiogenesis

Landscape Effects

Metastasis

- Cytokines
- Chemokines
- Free Radicals
- Prostaglandins
- Growth Factors
- MMPs

- Cytotoxic T cell Activity
- Tumor Cell Lysis

- Humoral Immune Response
- T-Reg Cells
Free Radicals and Inflammation

FREE RADIALS AND INFLAMMATION

ROS
\[ \dot{\text{O}}\text{H} \quad \text{O}_2^- \] (Hydroxyl radical)

RNS
\[ \text{NO} \cdot \quad \text{ONOO}\cdot \quad \text{N}_2\text{O}_3 \] (Nitric Oxide, Peroxynitrite, Nitrous Anhydride)

Protein Damage
(DNA Repair Enzymes, Caspases)

DNA Damage and Mutation
Nitrosamines/Deamination
8-oxo-dG
8-nitroguanine
Etheno Adducts
M1G Adduct

Base Excision Repair

Lipid Peroxidation
Arachidonic Acid Cascade
Eicosanoids
Cell Proliferation
Tumorigenesis

Inflammation Contributes to Various Stages of Tumorigenesis

Inflammation Types

Types of Inflammation in Tumorigenesis and Cancer

Grivennikov, Greten and Karin, Cell, 140, 2010
Obesity, Metabolic Syndrome and Cancer

Macrophage and Adipocyte Functions

CONVERGENCE OF MACROPHAGE AND ADIPOCYTE FUNCTIONS IN OBESITY AND METABOLIC SYNDROME

M Lehrke and Lazar MA, Nature Medicine, 10: 126, 2004
Chronic Inflammation

CHRONIC INFLAMMATION AFFECTS MULTIPLE PATHWAYS

Chronic Inflammation

Cytokines → Free radicals

DNA Damage

ATM

NFκB Pathway → Cell Survival

ATM

p53 Pathway → Cell cycle arrest

ATM

DNA Repair → Apoptosis

p14ARF

RB Pathway

Nitric Oxide Synthase

NITRIC OXIDE SYNTHASE

Inactive cNOS

Inactive cNOS

Active cNOS (NOS1/NOS3)

Ca^{++}+2

CaM

CaM

CaM

CaM

L-ARGININE

Citrulline

NO^{••}

iNOS (NOS2)

Always active
NOS2 Induction

INDUCTION OF NOS2

INFLAMMATION

NFκB
HYPOXIA
OXIDATIVE STRESS

CYTOKINES
β-CATENIN/TCF4
ENDOTOXIN

NOS2

NO°
p53 Transrepresses NOS2

p53 TRANSREPRESSES NOS2: EXAMPLE OF p53 NEGATIVE FEEDBACK LOOP

- NOS2 - inducible nitric oxide synthase
  - Macrophages
  - Epithelia

Forrester et al., PNAS 93: 2442, 1996
Ambs et al., PNAS 95: 8823, 1998
Ulcerative Colitis Colon

ULCERATIVE COLITIS COLON

Normal  UC Lesions  UC Cancer

Ulceration  Cancer
Colorectal Cancer and Ulcerative Colitis

CUMULATIVE RISK OF DEVELOPING COLORECTAL CANCER FOR A PATIENT WITH ULCERATIVE COLITIS
NOS2 Expression

NOS2 EXPRESSION CORRELATES WITH p53 SER15 PHOSPHORYLATION AND EXPRESSION OF p21 and MDM2 IN ULCERATIVE COLITIS

Hofseth et al., PNAS, 2003
Nitric Oxide

NITRIC OXIDE CAN ACTIVATE THE PROTECTIVE p53 STRESS RESPONSE PATHWAY

UC → Cytokines → NOS2 → NO• → DNA damage

ATM ↓ ATR
p53 activation

Anticarcinogenic Effects
- Growth arrest
- DNA repair
- Apoptosis
- Anti-oxidative and -nitrosative stress*

* S. P. Hussain et. al., Cancer Res., 64, 2004
Hypothesis

HYPOTHESIS: NITRIC OXIDE AND OXYRADICALS ASSOCIATE WITH CANCER-RELATED MUTATIONS IN CHRONIC INFLAMMATORY DISEASES PRIOR TO CANCER

UC \text{ Cytokines} \rightarrow \text{NOS2} \rightarrow \text{NO}^* \rightarrow \text{DNA damage} \rightarrow \text{p53 activation} \rightarrow \text{Anticarcinogenic Effects}

- Growth arrest
- DNA repair
- Apoptosis
- Anti-oxidative and -nitrosative stress

p53 Mutation

INCREASED TISSUE p53 MUTATION LOAD IN CHRONIC INFLAMMATORY AND OXYOVERLOAD CONDITIONS

- Ulcerative Colitis
- Hemochromatosis
- Wilson Disease
- Lungs from Tobacco Smokers
- Chronic Pancreatitis?

Hussain et al., Cancer Res. 60: 333, 2000
Hussain et al., Cancer Res. 61: 6350, 2001
Hagiwara et al., Cancer Res., 66:8309, 2006
Stress Response Pathway

NITRIC OXIDE CAN ACTIVATE THE PROTECTIVE p53 STRESS RESPONSE PATHWAY AND INDUCE ONCOGENIC p53 MUTATIONS

Chronic Inflammation → Cytokines → NOS2 → NO^• → DNA damage → p53 mutations → p53 activation

Anti-carcinogenic Effects
- Growth arrest
- DNA repair
- Apoptosis
- Anti-oxidative and -nitrosative stress

Pro-carcinogenic Effects
- Genomic instability
- Decrease cell cycle checkpoints, apoptosis and DNA repair
- Pro-oxidant and -nitrosative stress
Inflammation and Pancreatic Cancer

Chronic Pancreatitis

Desmoplastic Stroma

NF-κB Signaling

Inflammatory Cytokines

COX2, NOS2, LOX

Pancreatic Cancer
Chronic Pancreatitis

Increased Risk of Pancreatic Cancer in Patients with Chronic Pancreatitis

Lowenfels, A.B., et. al., NEJM, 328, 1993
Components of Pancreatic Cancer

COMPONENTS OF PANCREATIC CANCER

Inflammatory Changes

INFLAMMATORY CHANGES DURING DEVELOPMENT AND PROGRESSION OF PANCREATIC CANCER

Oncogenic KRAS

Inflammation enhances and maintains a pathologic level of oncogenic KRAS in pancreatic cancer

Daniluk et. al., JCI, 2012
NOS2 expression

Increased NOS2 expression is associated with poor survival of resected pancreatic cancer patients (Stage I/II).

Human Pancreatic Ductal Adenocarcinoma

K/M Survival Curve

NOS2 (IHC)

Tumor

Nontumor

Tumor
(N=30)

Nontumor
(N=27)

P=0.03
Pancreatic cancer mouse model

Genetically Engineered Mouse Model of Pancreatic Cancer

*LSL-Kras-G12D X p53 LSL R172H X Pdx-Cre 1 (KPC)

Pancreatic Ductal Adenocarcinoma (PDAC)
(Median Survival = 4-5 months)

*Hingorani, S.R. et al., Cancer Cell, 2005
NOS2 knockout mouse

HYPOTHESIS: NO⁺ Enhances Pancreatic Cancer Progression

- NOS2-deficiency Enhances Survival in KPC Mice

NOS2 deletion in genetically engineered mouse model of pancreatic cancer
Nitric oxide and pancreatic cancer

NO$^\bullet$ and Pancreatic Cancer

FOXO: Fork-head box transcription factor (subclass O)
Nostrin

NOSTRIN is a Negative Regulator of Disease Aggressiveness

Wang, J. et. al., Clin Cancer Res., 2016
MIF
Macrophage Migration Inhibitory Factor (MIF)

- Increased Expression in Tumors
- NF-κB
- ERK1/2
- Akt
- COX2
- p53
- Rb/E2F
- NOS2

- Proinflammatory Cytokine
- Expressed in Epithelial and inflammatory Cells

Immune/Inflammatory Response
MIF, Inflammation, and Cancer

Cell Press

Macrophage Migration Inhibitory Factor: A Probable Link between Inflammation and Cancer

Richard Bucala and Seamus C. Donnelly

The pleiotropic effects of macrophage migration inhibitory factor (MIF) place it in a central position in the immunopathogenesis of many diseases. Here we discuss the current understanding of MIF's role and highlight it as a potential link between inflammatory activation and malignant progression.

J. Exp. Med., 190, 1999

At the Crossroads of Inflammation and Tumorigenesis

By Carlos Cordon-Cardo and Carol Prives
MIF expression

Increased expression of MIF in tumors from pancreatic ductal adenocarcinoma cases
Higher MIF Expression and PDAC

A Higher MIF Expression is Associated with Poorer Survival in Resected PDAC Cases

Funamizu et al., Int. J. Cancer, 2012
MIF expression

A higher expression of MIF is associated with poor survival in human PDAC

Validation in Independent Cohorts
MIF Overexpression

MIF-Overexpression Enhanced Tumor Growth and Metastasis in Orthotopic Mouse Model

47th Day Post-Implantation of 1mm³ Sub-Cutaneous Tumor into Pancreas

Tumor Size (mm³)
P = 0.0011

Tumor Weight (g)
P = 0.0015

Visible Metastasis (Number)
P = 0.0064
MIF and EMT-related genes

MIF Induces a Marked Change in Global Gene Expression Profile including EMT-related Genes in Orthotopic Tumors

- MIF over-expressing tumors are poorly differentiated.
- MIF induces a change in global gene expression profile.
- MIF over-expressing tumors showed expression of EMT-related genes.
Pancreatic cancer exosomes initiate pre-metastatic niche formation in the liver


Pancreatic ductal adenocarcinomas (PDACs) are highly metastatic with poor prognosis, mainly due to delayed detection. We hypothesized that intercellular communication is critical for metastatic progression. Here, we show that PDAC-derived exosomes induce liver pre-metastatic niche formation in naive mice and consequently increase liver metastatic burden. Uptake of PDAC-derived exosomes by Kupffer cells caused transforming growth factor β secretion and upregulation of fibronectin production by hepatic stellate cells. This fibrotic microenvironment enhanced recruitment of bone marrow-derived macrophages. We found that macrophage migration inhibitory factor (MIF) was highly expressed in PDAC-derived exosomes, and its blockade prevented liver pre-metastatic niche formation and metastasis. Compared with patients whose pancreatic tumours did not progress, MIF was markedly higher in exosomes from stage I PDAC patients who later developed liver metastasis. These findings suggest that exosomal MIF primes the liver for metastasis and may be a prognostic marker for the development of PDAC liver metastasis.
MIF and pancreatic cancer

MIF-induced disease aggressiveness in pancreatic cancer
miR-301b and pancreatic cancer

MIF/miR-301b/NR3C2 Axis in Pancreatic Cancer
MIF deficiency

MIF-deficiency enhanced survival and reduced metastasis in KPC mice

KPC Mouse Model

- **Survival**
  - Percent Survival vs Days
  - MKPC (N=48) vs KPC (N=31)
  - p < 0.0001

- **Metastasis**
  - % of Mice with Metastasis
  - Comparison between KPC (N=31) and MKPC (N=48)
  - MKPC vs KPC
  - p = 0.03
  - MKPC vs KPC with Liver Metastasis
  - p < 0.01

MKPC = MIF-deficient KPC mice
miRNA and Cancer

miRNA and Cancer

Ryan, Robles and Harris, Nat. Rev. Cancer, 2010

Volinia et al., PNAS, 2006
Inflammation, miRNA, and Cancer

INFLAMMATION, microRNA AND CANCER

Infection/Acid-Reflux/Injury

IMMUNE RESPONSE

SELECTIVE GENE-EXPRESSION

INFLAMMATION

miRNA

miR-146

TLR

NF-κB

IRAK1

TRAF6

IL-6

IFNβ, IFNγ, TNFα

STAT3

Let-7a

HMGA-1

miR-155

Tp53INP1

Oncogenic

Mir-21

PTEN Tumor Suppressor

Chronic Inflammatory Diseases

Alterations in miRNA Expression in Cancer-Prone Chronic Inflammatory Diseases

- **Ulcerative Colitis** → miR-21, miR-192, miR-375, miR-126, miR-195, miR-23a
- **Chronic Pancreatitits** → miR-100, miR-10b, miR-125, miR-199, miR-99
Balance of Inflammatory Signals

Overall Balance of Inflammatory Signals Determine the Outcome

Schetter et. al., Carcinogenesis, 31, 2009
Many chronic inflammatory diseases increase cancer risk.

Inflammatory and immune mediators contribute to different stages of tumor development and progression.

NO can activate p53 stress response pathway and may also induce oncogenic p53 mutation in ulcerative colitis.

Increased Expression of inflammatory mediators (e.g., MIF and NO) enhances disease aggressiveness in pancreatic cancer.

Targeting MIF and NO-induced signaling pathways increased survival in mouse model with lethal pancreatic cancer.

The highly complex interactions of immune and inflammatory signaling determines the overall effect of inflammation on tumorigenesis.