Lung Cancer Research

Oncology for Scientists II (RPN 532)

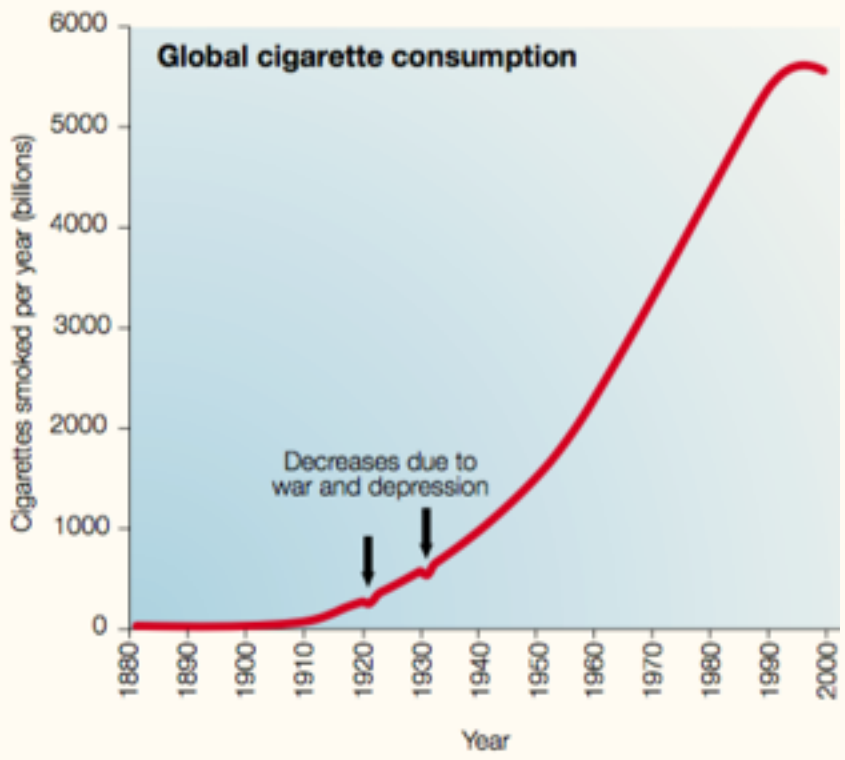
Santosh Patnaik, MD, PhD
Assistant Professor
Department of Thoracic Surgery
Roswell Park Cancer Institute

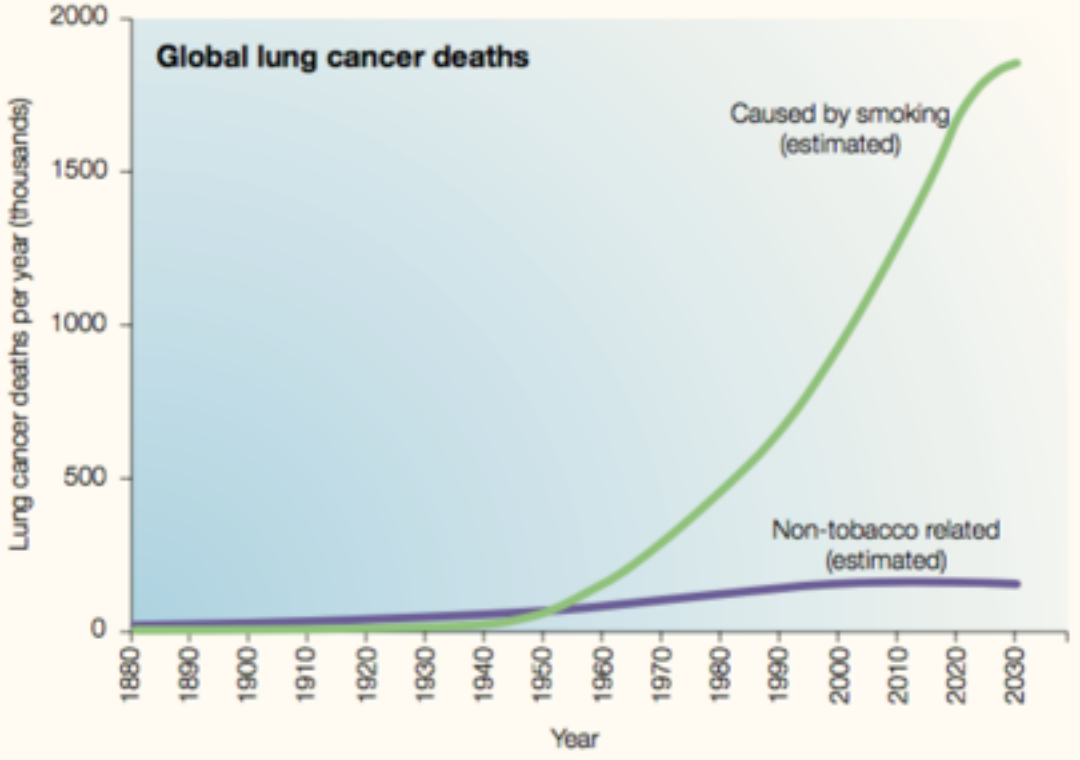
Almost a man-made disease

First case of lung cancer		1751
World-wide lung cancer cases	*up to* 1898	140
World-wide lung cancer cases	up to 1912	374
World-wide lung cancer cases	*in* 2012	1800000

NATURE REVIEWS | CANCER
VOLUME 1 | OCTOBER 2001 | 83

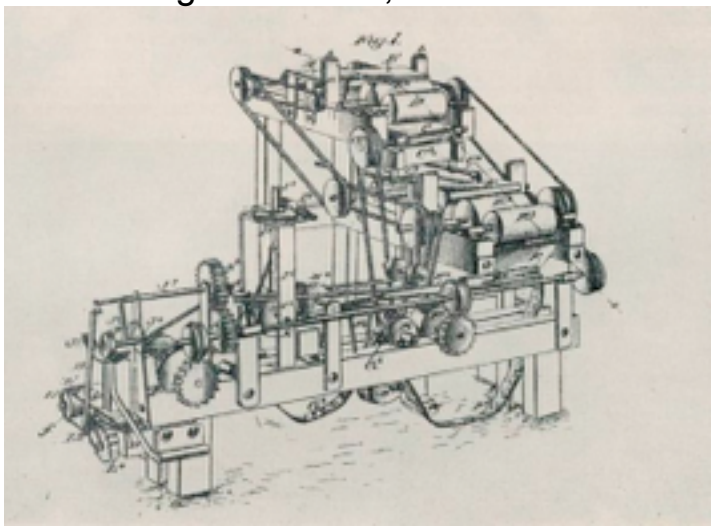
Mayan god smoking (ca. 1c BC)

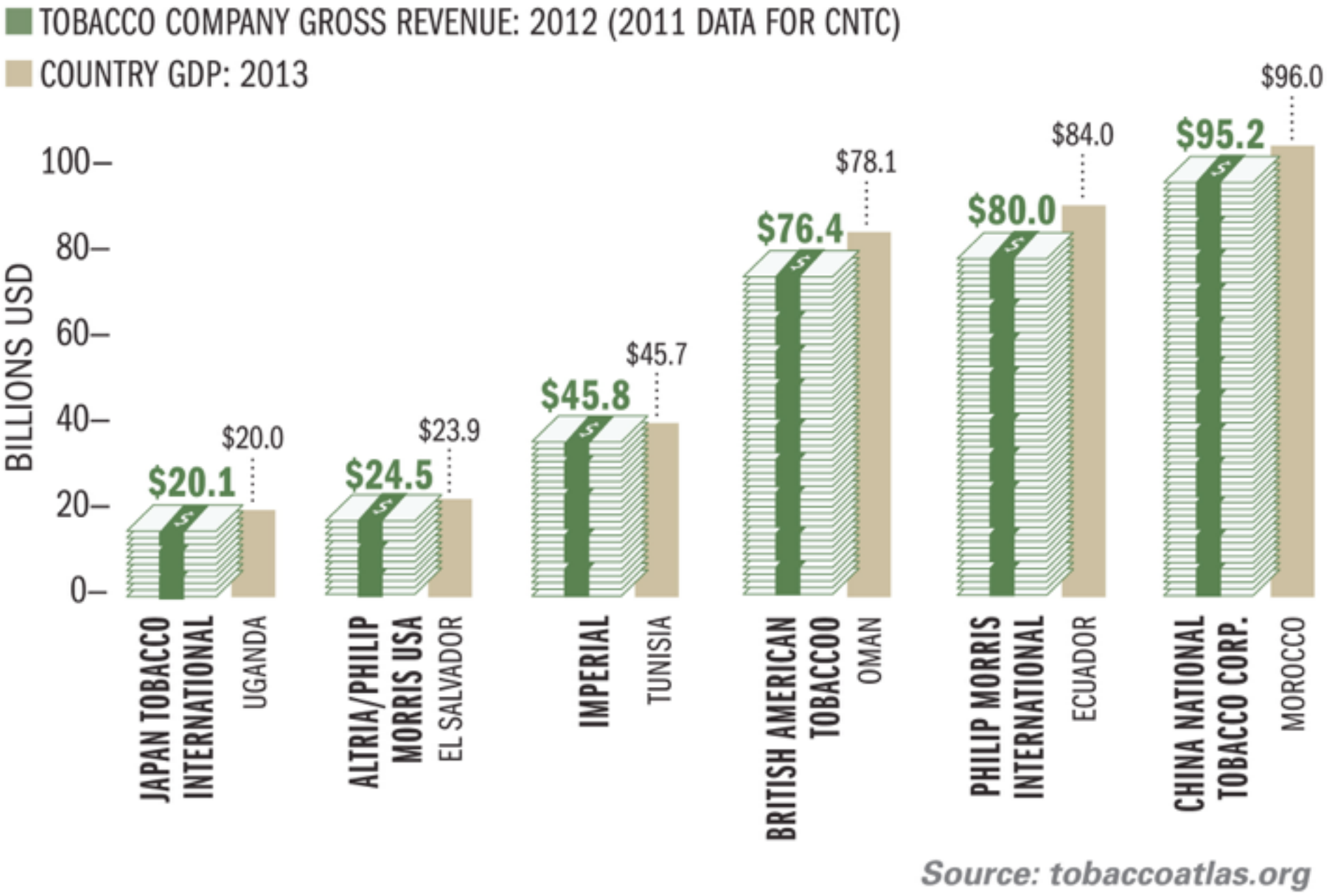




1612 China
1723 Berlin
–1920s 14 US states

Bonsack cigarette roller, 1880 invention

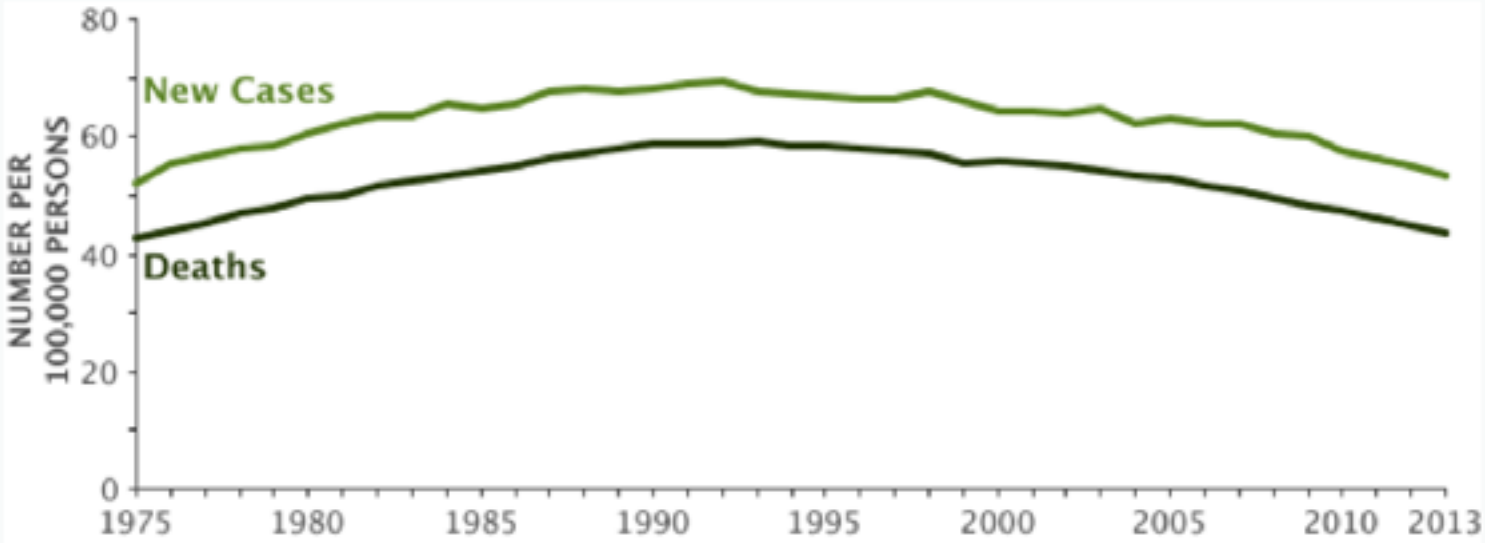




Source: tobaccoatlas.org

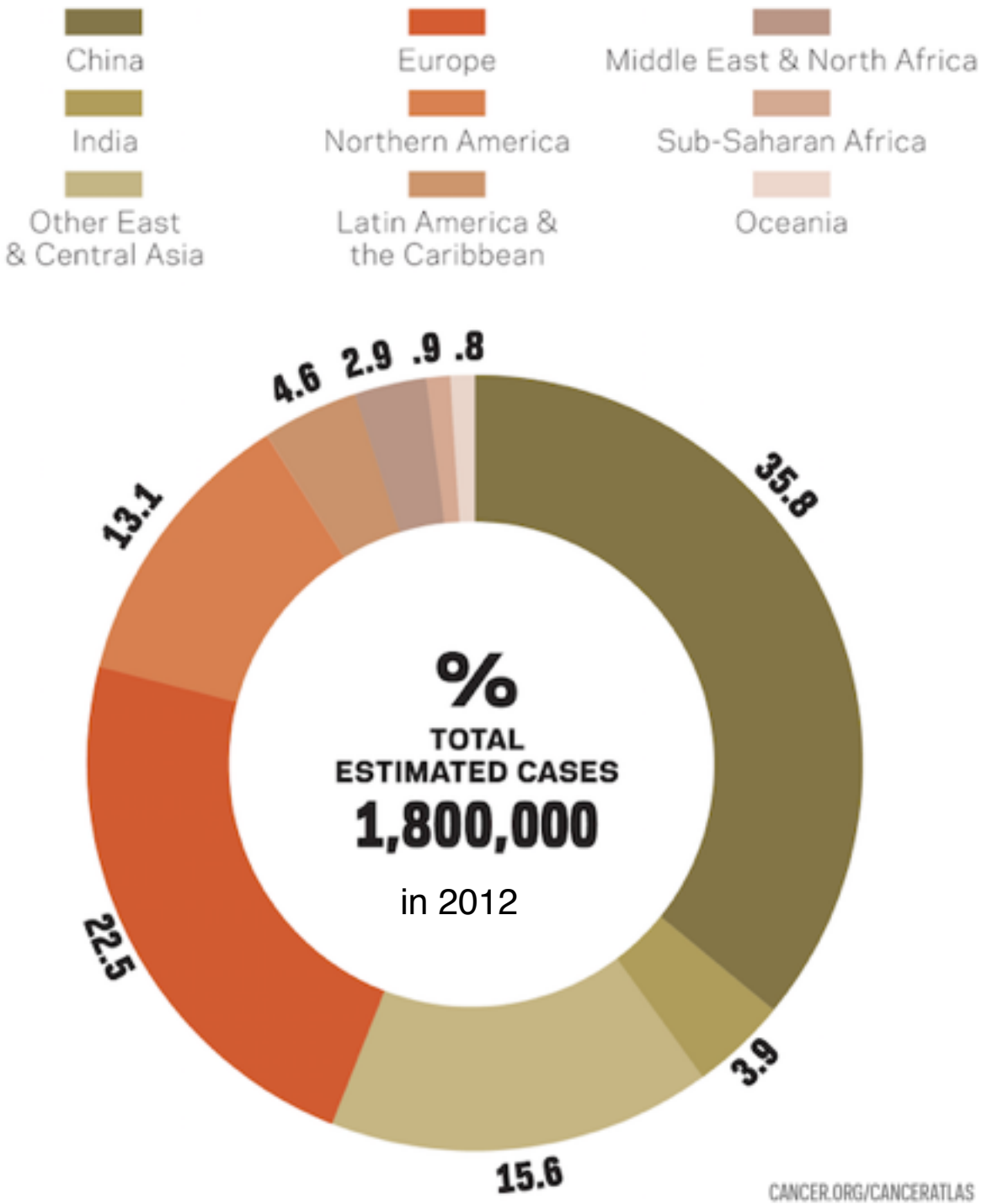
New Cases, Deaths and 5-Year Relative Survival

View Data Table



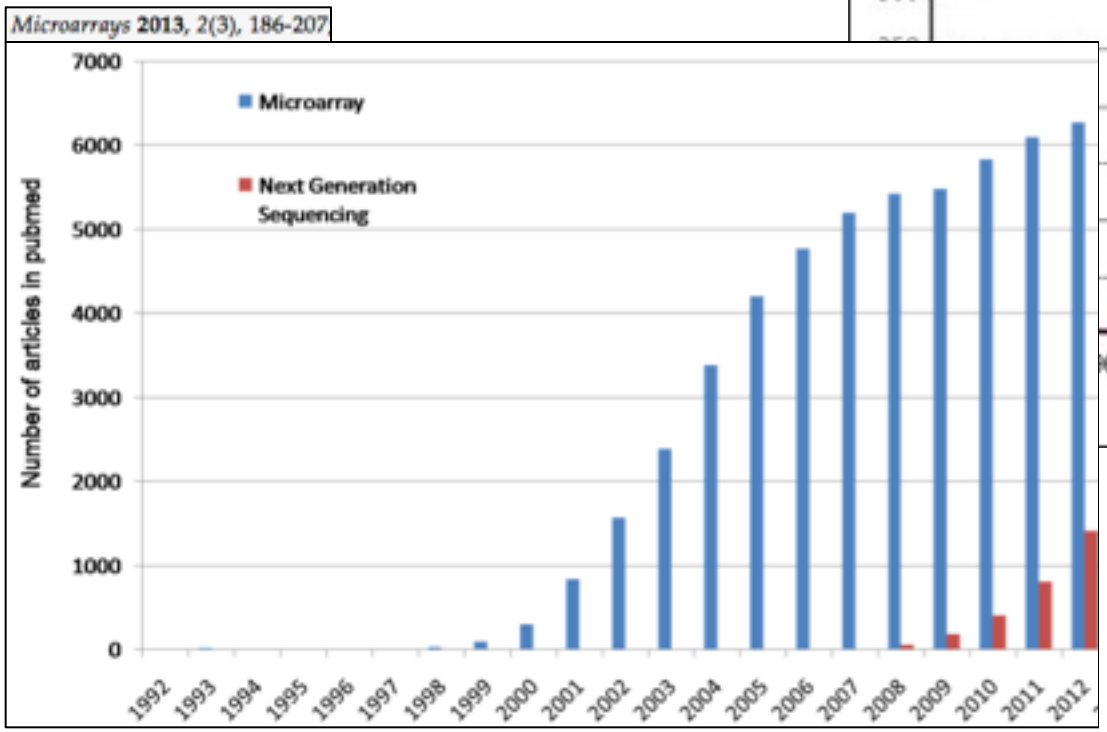
Year	1975	1980	1985	1990	1995	2000	2004	2008
5-Year Relative Survival	11.4%	12.5%	13.1%	13.3%	14.5%	15.7%	16.8%	18.7%

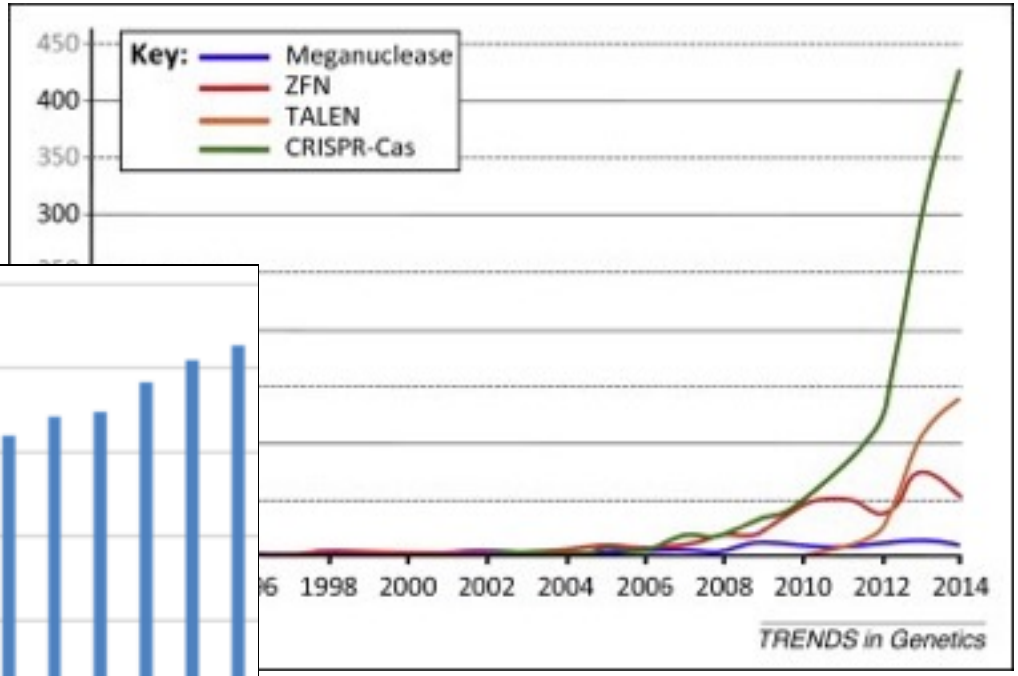
SEER 9 Incidence & U.S. Mortality 1975-2013, All Races, Both Sexes. Rates are Age-Adjusted.


China
Europe
Middle East & North Africa
India
Northern America
Sub-Saharan Africa
Other East & Central Asia
Latin America & the Caribbean
Oceania
35.8
3.9
15.6
22.5
13.1
4.6
2.9
.9
.8
%
TOTAL ESTIMATED CASES
1,800,000
in 2012
CANCER.ORG/CANCERATLAS

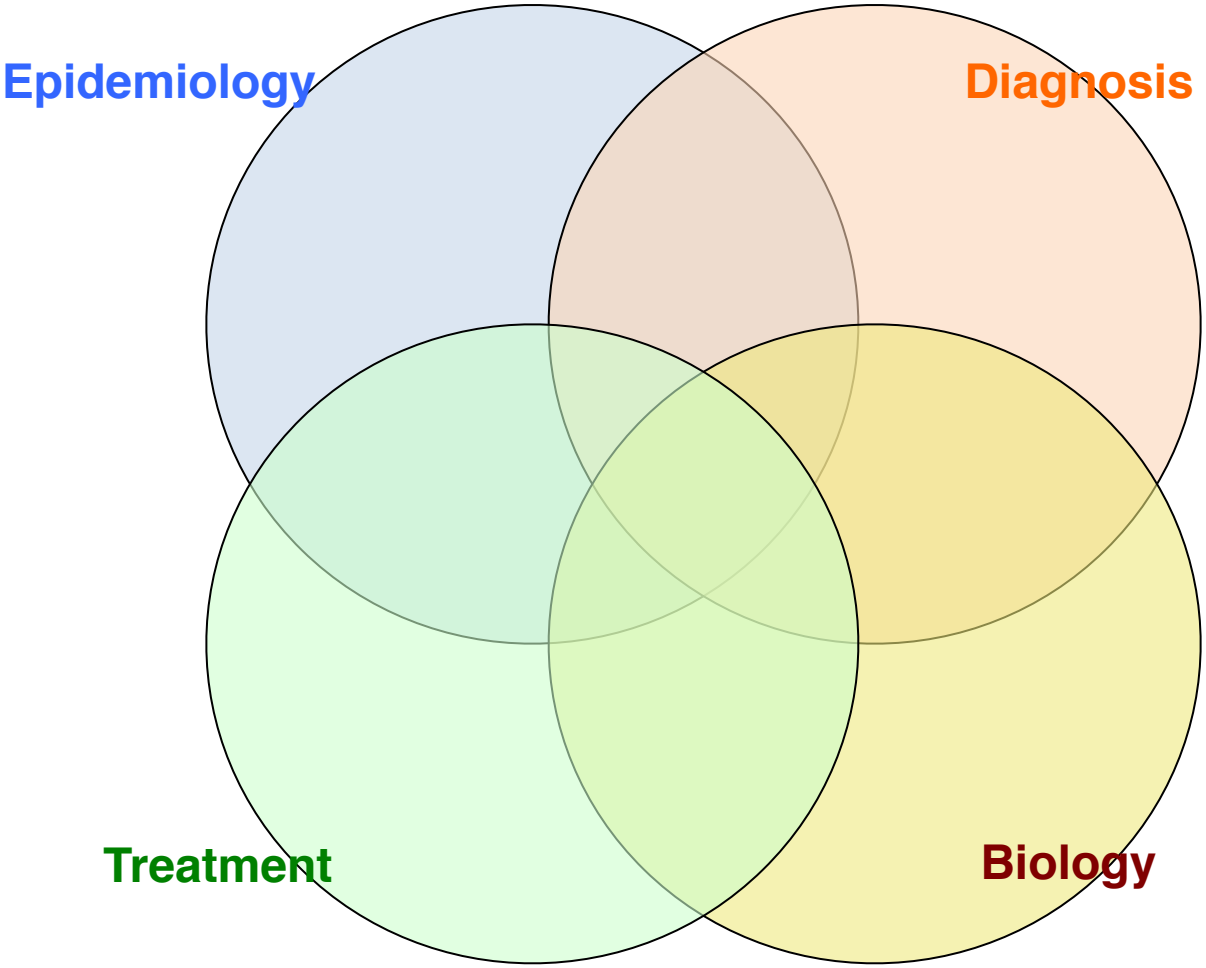
Almost a man-made disease

Ephemeral value of research



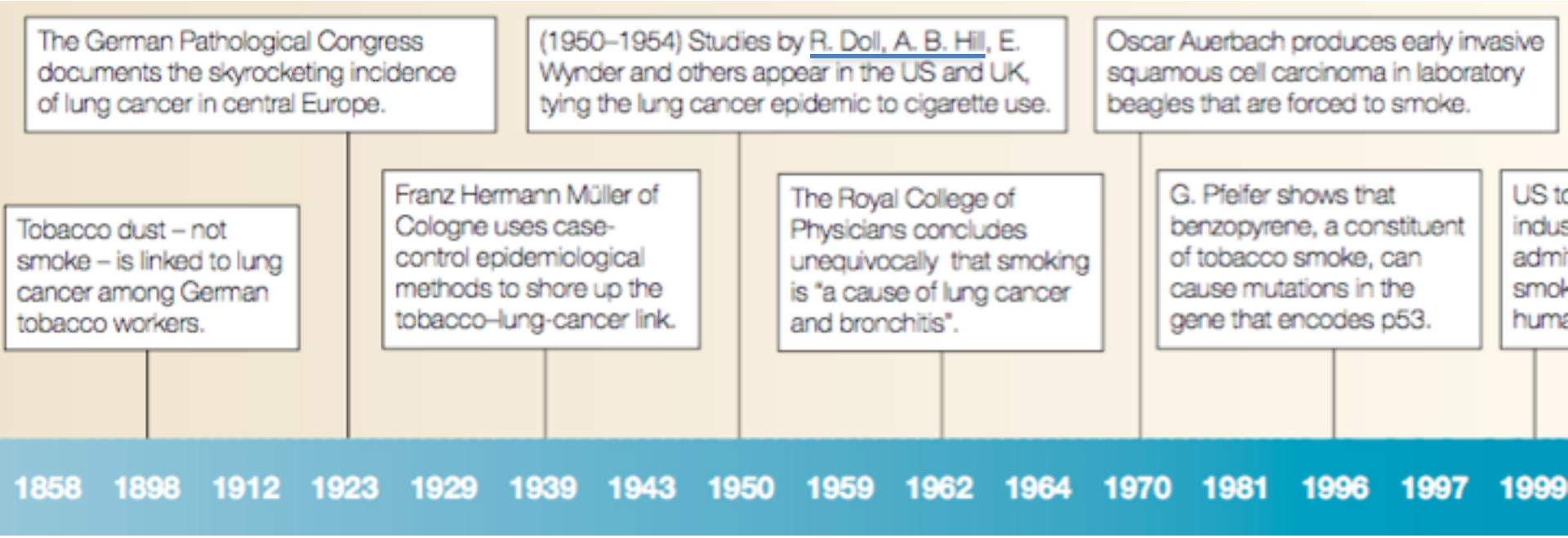


Facets of research in lung cancer



Epidemiology: Smoking & cancer

British Doctors' Study 1951–2001
80% UK doctors smoked in 50s



Epidemiology: Asbestos and mesothelioma

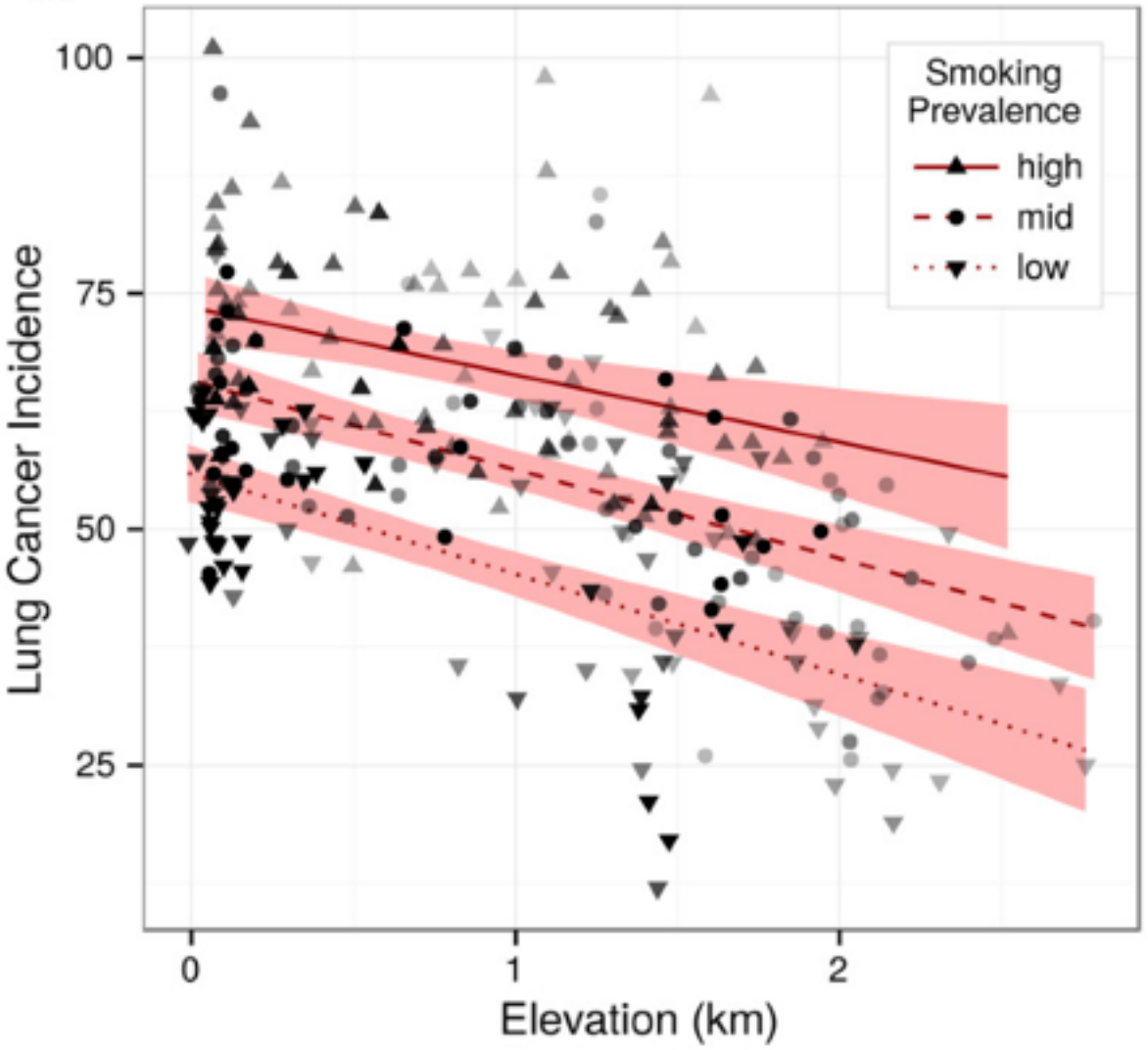
80% of mesothelioma (pleural cancer) by asbestos

2013: 50,000 mesothelioma cases world-wide

2002: US asbestos production ends

Epidemiology: Altitude & lung cancer

PeerJ 3:e705; DOI 10.7717/peerj.705

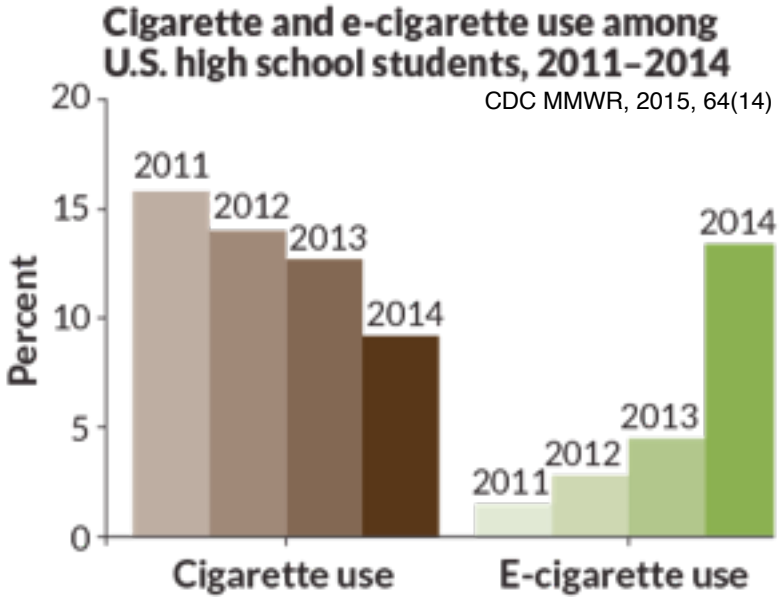


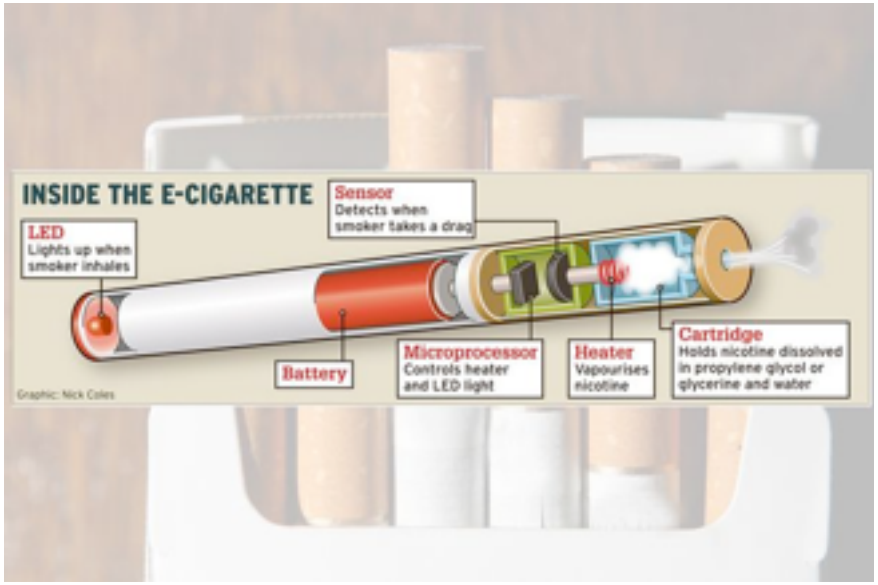
Not because of UV, radon, etc.

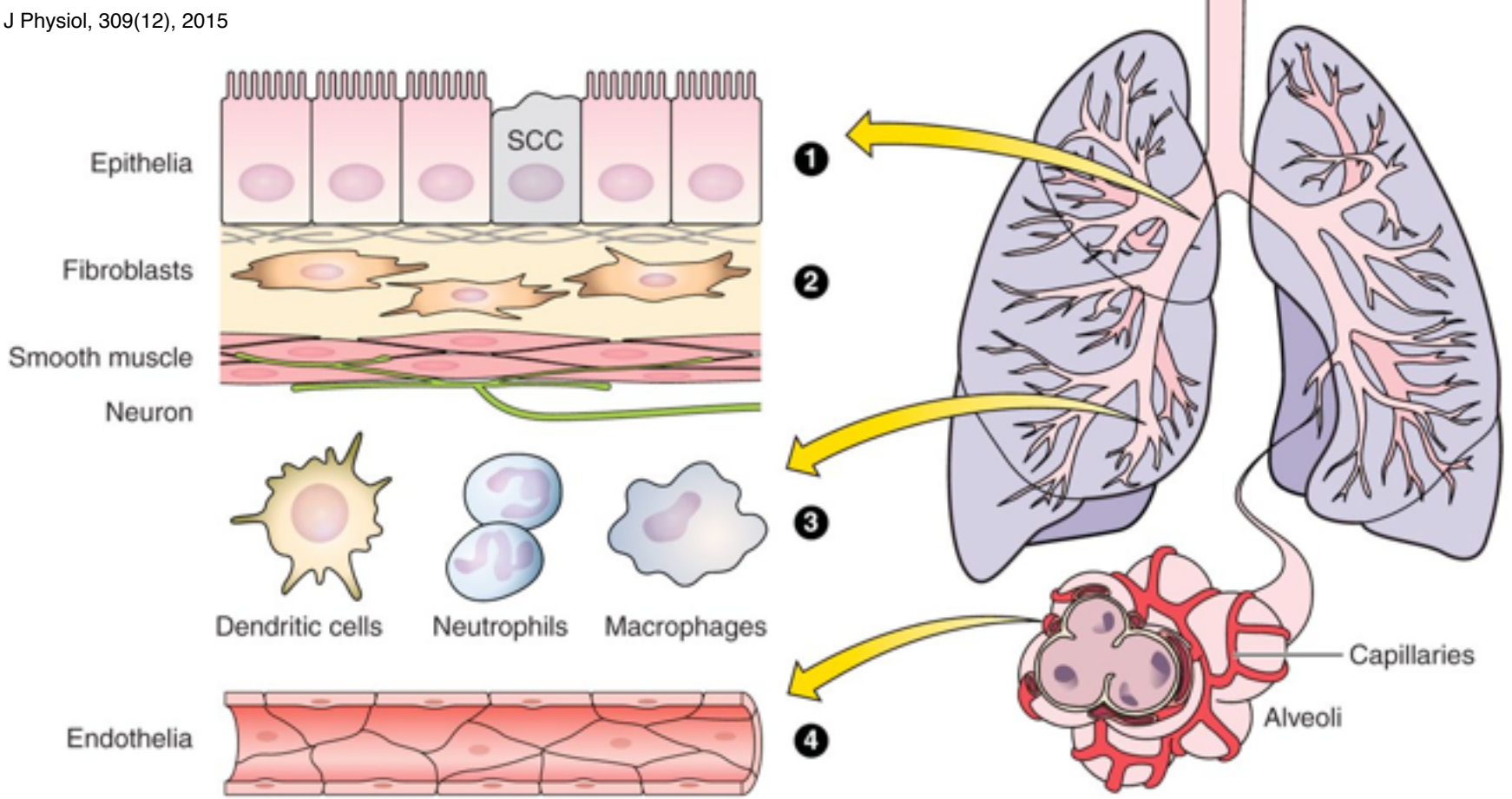
Not seen for breast cancer

Oxygen-driven tumorigenesis?

Epidemiology: E-cigarettes / vaping

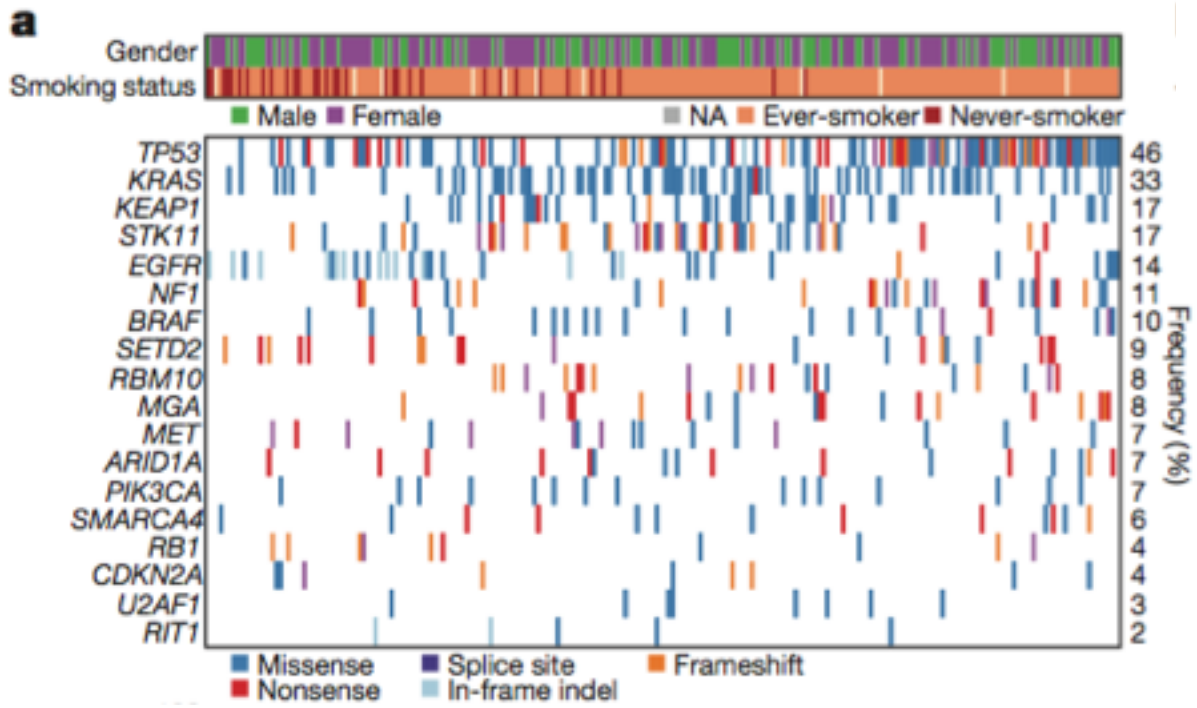






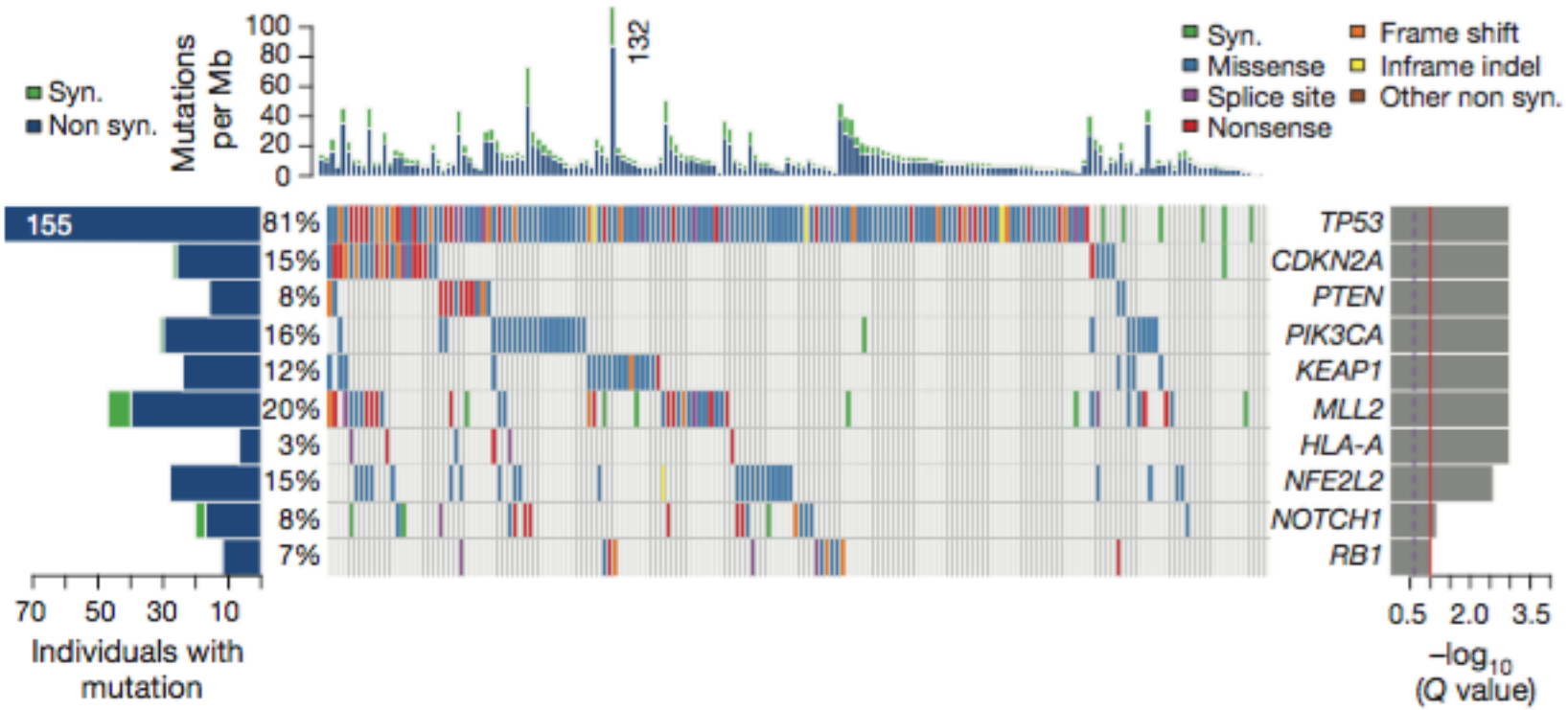
Current data for the effects of E-cigarettes/E-liquids on the lung	
Tissue/cell type	**Effects**
(1) Epithelia	↑Cytotoxicity[31], ↓Cell viability[31], ↑Inflammation[94,159], ↑Infection[159]
(2) Fibroblasts	↑Cytotoxicity[10,94], ↓Cell viability[10,94], Altered morphology[94]
(3) Inflammatory cells (BALF)	↑Macrophages[142], ↑Cytokine secretion[31], ↑Infection[159]
(4) Endothelia	↓Cell viability[133], ↓Electrical resistance[133]

Genetic alterations in lung cancer



Lung adenocarcinoma

Cancer Genome Atlas; Nature 511:543, 2014

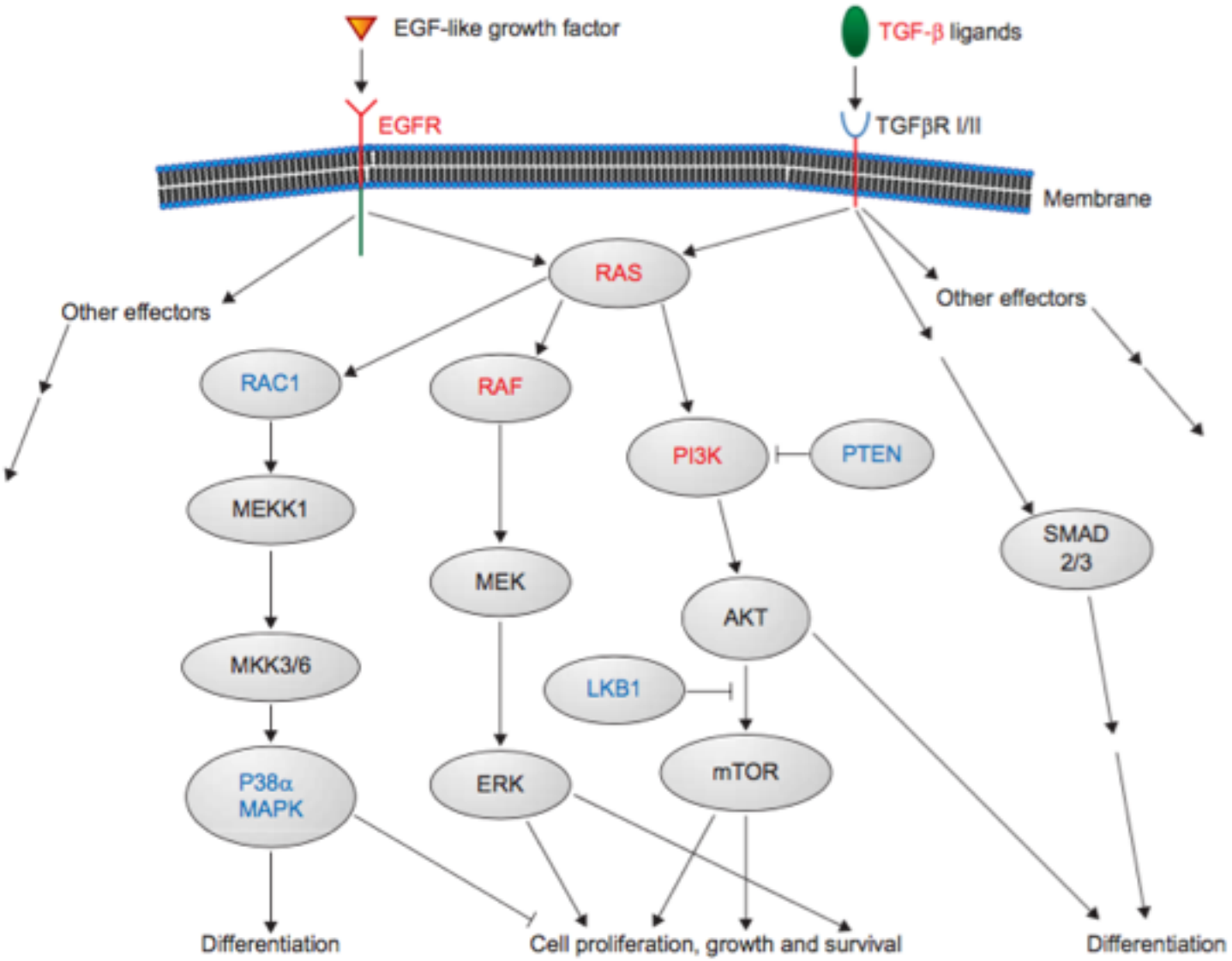


Lung squamous cell carcinoma

Cancer Genome Atlas; Nature 598:520, 2012

Animal models of lung cancer

Major lung cancer-associated signaling pathways



Eur Resp J, 35(2), 2009

Spontaneous/treatment-induced mouse models of lung cancer

Spontaneous

~3% of wild mice
100% of A/J strain mice (*KRAS* polymorphism)

Treatment-induced

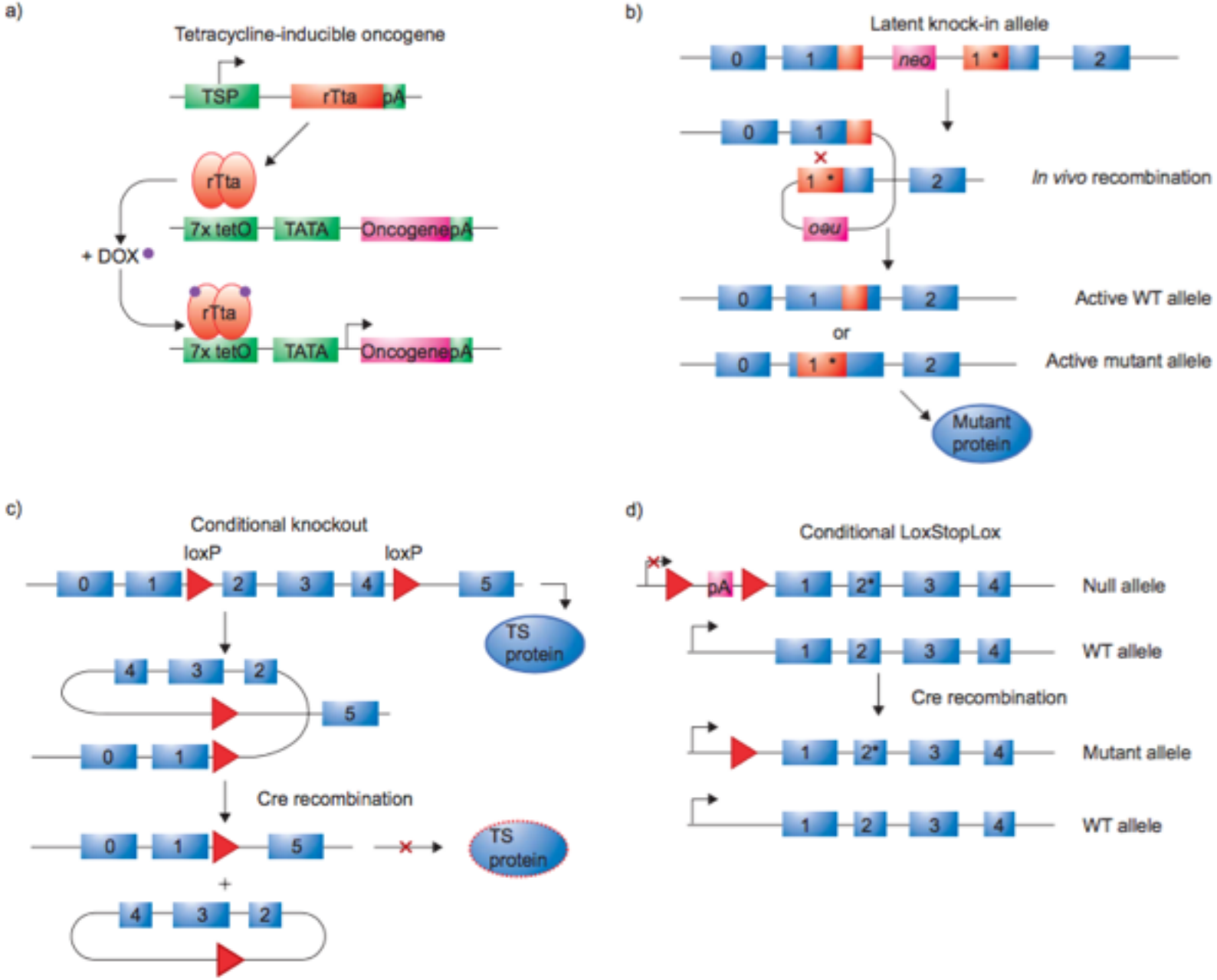
Cigarette smoke: very weak
Cigarette smoke constituents: stronger

Model	Strain	Carcinogen	Tumor
Mouse AD/ADC	A/J	B (a)P, i.p. 100 mg/kg	20 w: 8-10 tumors (AD), 100% incidence (20, 21)
		B (a)P, i.g. 100 mg/kg (3X)	
	A/J	NNK, i.p. 100 mg/kg	20 w: 6-8 tumors (AD), 100% incidence (20, 22, 23) 52 w: 15 tumors (95% AD, 5% ADC), 70-80% incidence (ADC)
	A/J	Urethane, i.p. 1 g/kg	16 w: 20-25 tumors (AD) (21, 24-26)
	A/J	Vinyl carbamate, i.p. 60 mg/kg	24w: 25 tumors (AD), 12% incidence (ADC) 52 w: 30% incidence ADC (27, 28)
	Swiss albino - newborn	Main-stream cigarette smoke, 120 days	26-33w: 6-14 tumors (AD), 80% incidence (AD), 5-20% incidence (ADC) (29)
	A/J	Main- and side-stream cigarette smoke, 5 mos smoke + 4 mos air	3 tumors (AD) vs 1 spontaneous tumor (AD) (30)
	B6C3F1	Mainstream cigarette smoke, lifetime	10X increase in hyperplasia, 4.6X AD and papilloma, 7.3X ADC, 5X metastatic pulmonary ADC (31)
Rat AD/ADC	F344	NNK, s.c. 1.5 mg/kg (3X, 20 w)	98w: 67% incidence (AD), (33% ADC) (32)
	F344	Mainstream cigarette smoke, up to 30 months	Incidence increased from 0% in control to 6% (light smoke) to 14% (heavy smoke) (33)
Mouse squamous	Swiss – 8 w	NTCU, 3 µmol, 2x week (22 w)	24w: 50% hyper/metaplasia, 10% CIS/SCC (34)

i.p. = intraperitoneal, i.g. = intragastric, i.t. = intratracheal, AD = adenoma, ADC = adenocarcinoma
Benzo(a)pyrene = B(a)P
4- (methylnitrosamino)-1- (3-pyridyl)-1-butanone = NNK
N-nitroso-tris-chloroethylurea = NTCU

Frontiers in Bioscience E5, 939-946, June 1, 2013

Systems for genetically engineering mouse models



Eur Resp J, 35(2), 2009

Model type	Phenotype
Transgenic	
CC10-Tag and Sp-C-Tag	Multifocal bronchioloalveolar hyperplasias develop into mixed solid and papillary adenocarcinomas
K5-E6/E7[#]	Adenocarcinomas
MMTV-TGF-β1 DN[¶]	Adenocarcinomas
MMTV-RARβ4[+]	Alveolar hyperplasia
CC10-Tag; CC10-hASH1	Adenocarcinomas with focal NE differentiation
CC10-hASH1	Bronchial hyperplasia
CC10-hK14	Squamous differentiation hyperplasia with occasional squamous metaplasia
Sp-C-lgEGF	Alveolar hyperplasia
Sp-C-cMyc; SpC-lgEGF	Bronchioloalvealor adenocarcinomas
Sp-C-cMyc	Mixed bronchioloalveolar adenomas and adenocarcinomas
Sp-C-cRaf-1	Adenomas
MMTV-RARβ2[§]	Adenomas and adenocarcinomas
CC10-cMyc	Bronchioloalveolar hyperplasia
CGRP-H-Ras	NE hyperplasia and non-NE adenocarcinomas
Conditional transgenic	
Using Cre/lox system[/] β-actin-lox GFP lox-K Ras^{V12}-IRES-hPLAP	Alveolar hyperplasia, adenomas and adenocarcinomas
Lox-stop-lox-K Ras^{G12D}[##]	Epithelial hyperplasia of bronchioles, adenomatous hyperplasia, adenomas, both solid and papillary adenocarcinomas
Doxycycline regulatable	
CC10-rtTA; $(tetO_7)$CMV-FGF7	Epithelial cell hyperplasia and adenomatous hyperplasia
CC10-rtTA; $(tetO_7)$CMV-K Ras^{G12D}	Bronchogenic adenocarcinomas. Phenotype is completely reversible upon Dox removal
CC10-rtTA; $(tetO_7)$CMV-K Ras^{G12D} in a $Trp53^{-/-}$ or $Ink4a^{-/-}$ background	Bronchogenic adenocarcinomas. Phenotype is completely reversible upon Dox removal
CCSP-rtTA; $TetO_7$ PIK3CA(H1047R)	Adenocarcinomas
CCSP-rtTA; $TetO_7$-BRAFV600E	Mixed adenocarcinoma and bronchioalveolar carcinoma
SP-C-rtTA; TetO7-Cre; $Pten^{F/F}$	Impaired alveolar epithelial differentiation and hyperplasia
CCSP-rtTA; Tet-O_7-$hEGFR^{L858R}$	Adenocarcinoma with bronchioalveolar carcinoma features
	Local invasive adenocarcinoma after longer incubation
CCSP-rtTA; Tet-O_7-$hEGFR^{DEL}$	Adenocarcinoma with bronchioalveolar carcinoma features
	Local invasive adenocarcinoma after longer incubation but with longer latency
Spontaneous activatable knock-in latent allele $K^{RasG12D\ LA}$	Epithelial hyperplasia of bronchioles, adenomatous hyperplasia, adenomas, both solid and papillary adenocarcinomas
K $Ras^{G12D\ LA}$ in $Trp53^{-/-}$ background	Epithelial hyperplasia of bronchioles, adenomatous hyperplasia, adenomas, both solid and papillary adenocarcinomas above but with shorter latency
K $Ras^{G12D\ LA}$; $TGF\text{-}\beta^{+/-}$	Increase of adenocarcinoma formation
Conditional knockout	
Using Cre/lox system	
Trp53	Adenocarcinomas
$Rb^{F/F}$; $Trp53^{F/F}$	NE hyperplasia, SCLC with metastases
Compound conditional knock out and transgenes	Phenotypes compared with LSLKrasG12D control
Spry-$2^{F/F}$; $LSLKras^{G12D}$	Increased number of adenocarcinoma
$Rac1^{F/F}$; $LSLKras^{G12D}$	Adenoma formation with long latency
$Pik3r2^{-/-}$; $Pik3r1^{F/F}$; LSL $Kras^{G12D}$	Strong decrease of adenoma and adenocarcinoma formation
$Dicer^{F/F}$; $LSLKras^{G12D}$	Increased number of adenocarcinoma
$Trp53^{F/F}$; $LSLKras^{G12D}$	Progressed adenocarcinoma with lymph node metastases
$Trp53^{LSL\ R172H/-}$; $LSLKras^{G12D}$	Idem
$Trp53^{LSL\ R270H/-}$; $LSLKras^{G12D}$	Idem but with higher penetrance
$Lkb1^{F/F}$; $LSLKras^{G12D}$	Shorter latency with mixed adenocarcinoma/squamous cell carcinoma with occasional large cell carcinoma; frequent metastases
CC10-Cre; $Pten^{F/F}$; $LSLKras^{G12D}$	Bronchioalveolar lesions followed by adenocarcinoma with local invasion and stromal interactions
$Rb^{F/F}$; $p130^{F/F}$; $LSLKras^{G12D}$	Shorter latency and higher number of adenocarcinoma
	No neuroendocrine tumours

Eur Resp J, 35(2), 2009

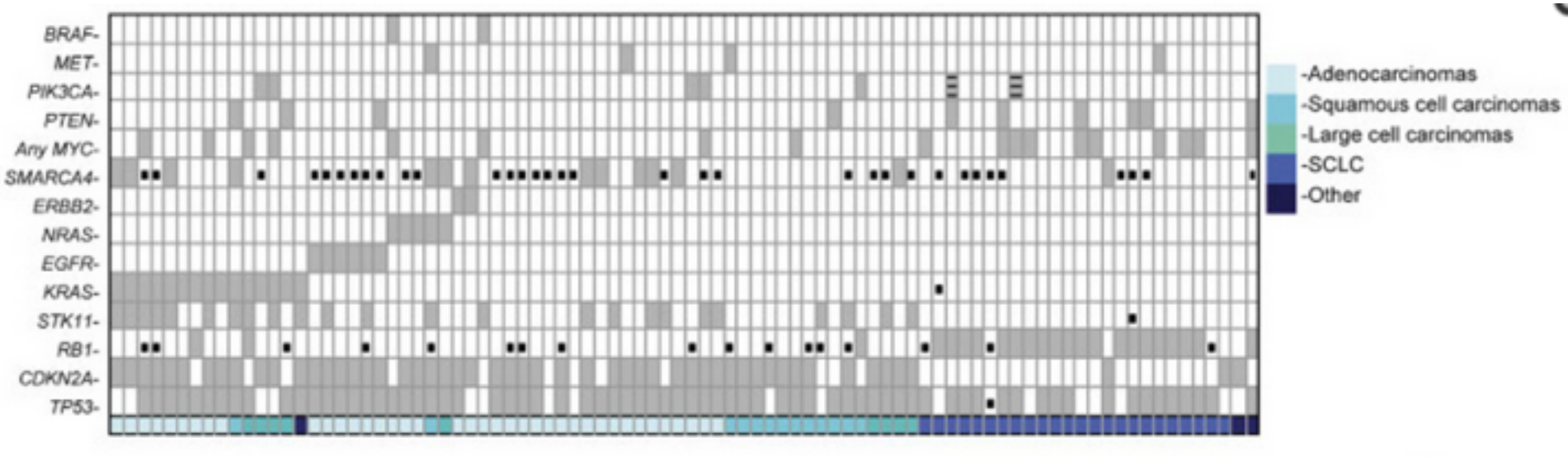
Lung cancer cell lines

Human lines >> mouse

NCI-H1703	Human	Lung	Adenocarcinoma, non-small cell
NCI-H2135	Human	Lung	Cancer, non-small cell lung
NCI-H2172	Human	Lung	Cancer, non-small cell lung
NCI-H2444	Human	Lung	Cancer, non-small cell lung
NCI-H835	Human	Lung	Carcinoid
UMC-11	Human	Lung	Carcinoid
NCI-H720	Human	Lung	Carcinoid, atypical
A549	Human	Lung	Carcinoma
A-427	Human	Lung	Carcinoma
NCI-H596	Human	Lung	Carcinoma, adenosquamous
SW 1573	Human	Lung	Carcinoma, alveolar cell
NCI-H1688	Human	Lung	Carcinoma, classic small cell lung cancer
NCI-H1417	Human	Lung	Carcinoma, classic small cell lung cancer
NCI-H1672	Human	Lung	Carcinoma, classic small cell lung cancer
NCI-H2227	Human	Lung	Carcinoma, small cell lung cancer
NCI-H1963	Human	Lung	Carcinoma, small cell lung cancer
SHP-77	Human	Lung	Carcinoma, small cell lung cancer, large cell, variant
NCI-H2170	Human	Lung	Carcinoma, squamous cell
NCI-H520	Human	Lung	Carcinoma, squamous cell
SW 900	Human	Lung	Carcinoma, squamous cell
NCI-H358	Human	Lung	Carcinoma, bronchioalveolar, non-small cell
NCI-H727	Human	Lung	Carcinoid
LA-4	Mouse	Lung	Adenoma
LL/2 (LLC1)	Mouse	Lung	Carcinoma, Lewis lung
KLN 205	Mouse	Lung	Carcinoma, squamous cell

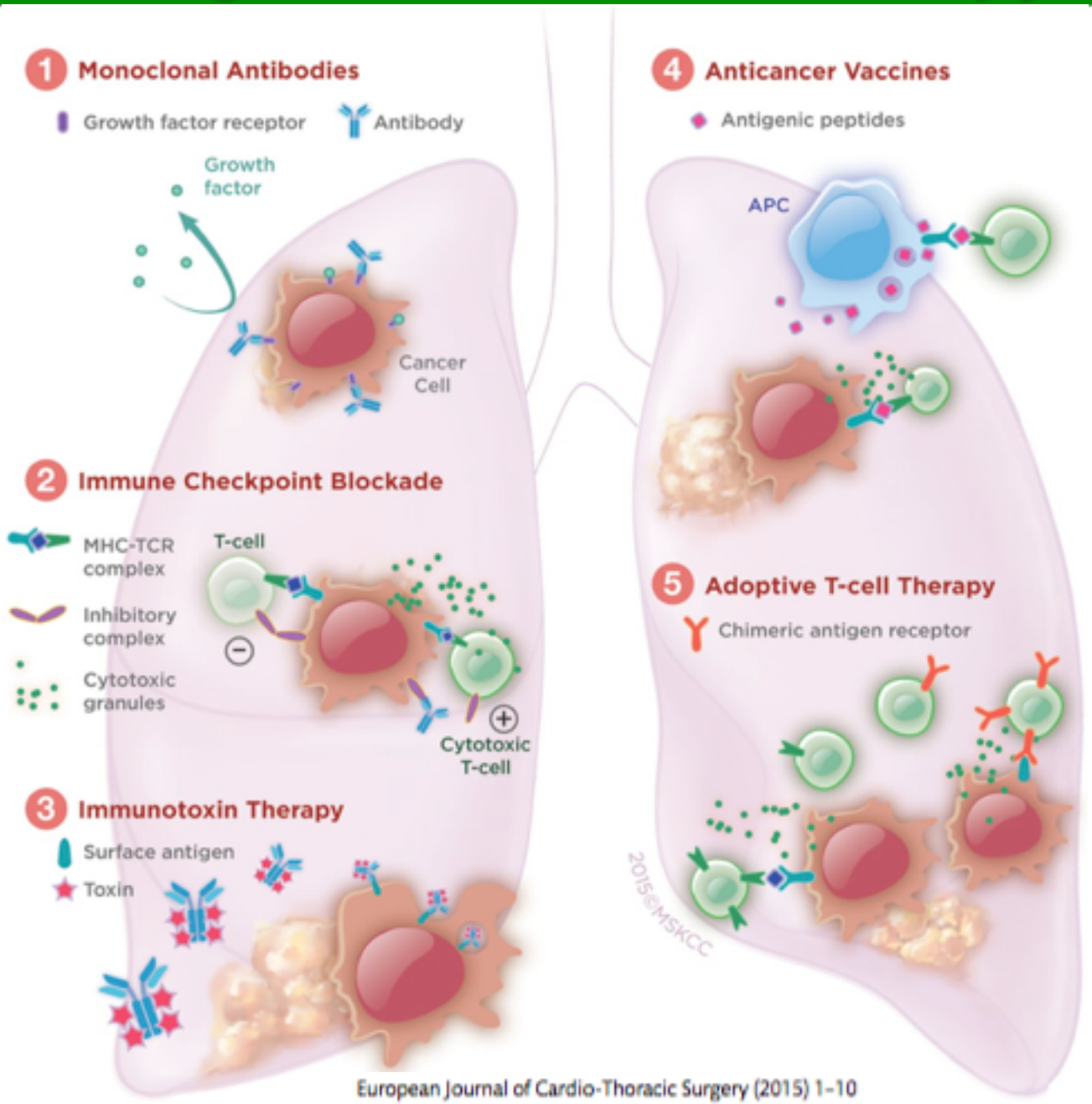
Examples; ATCC®

Genetic alterations in lung cancer cell lines



Human Mut, 30(8), 2009

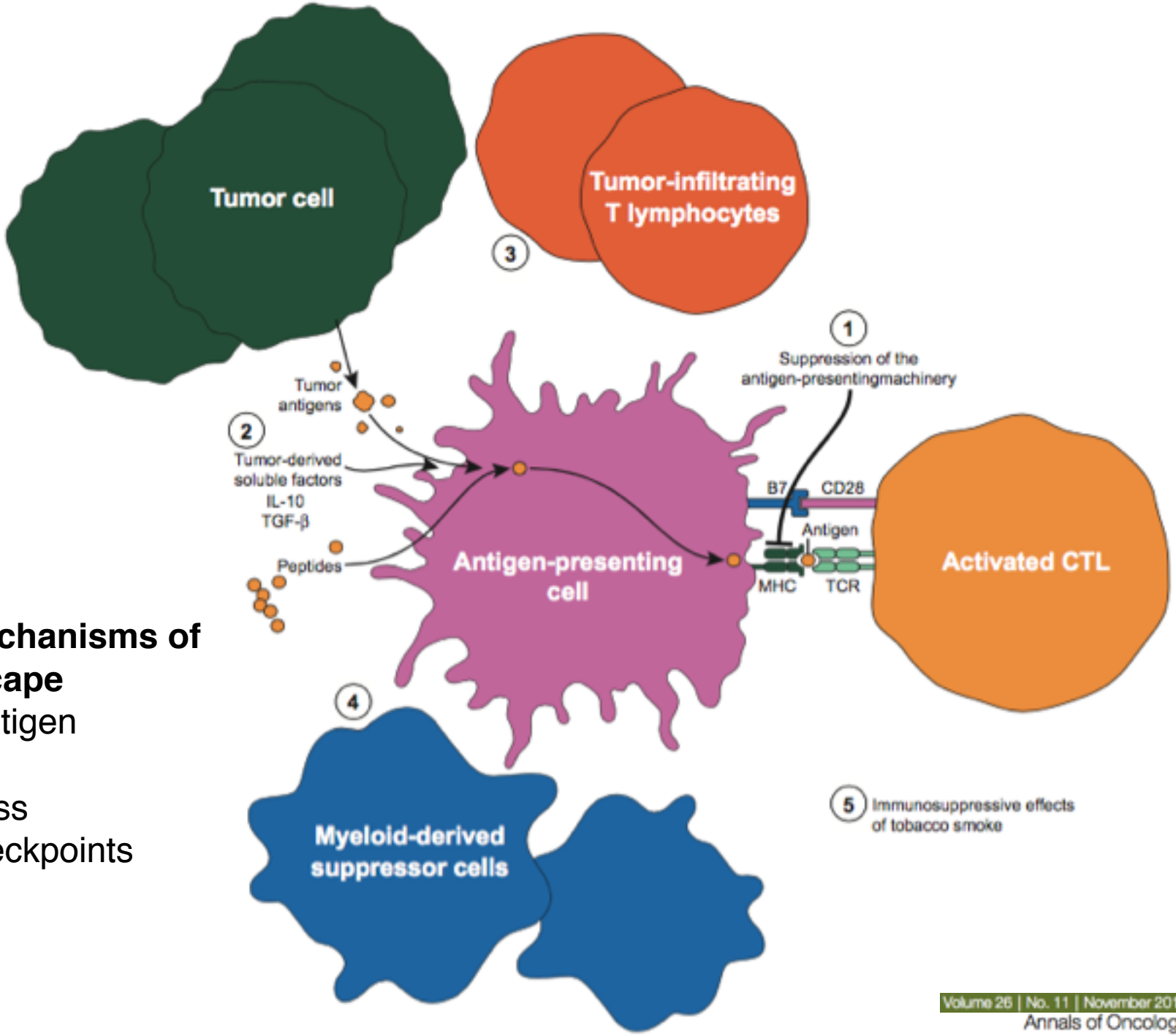
Lung cancer immunotherapy



European Journal of Cardio-Thoracic Surgery (2015) 1–10

Tumoral mechanisms of immune escape

- Reduced antigen presentation
- Antigenic loss
- Immune checkpoints



Volume 26 | No. 11 | November 2015
Annals of Oncology

Table 1: Monoclonal antibodies: ongoing clinical trials

	Phase	*n*	Comments	Ref.	Representative clinical trials
EGFR-targeted antibodies					
Cetuximab	Completed Phase II and Phase III	6	Modest benefit when used in combination with chemotherapy in first-line treatment of NSCLC; ongoing investigations into the efficacy of combination therapy with other chemo-immunotherapeutic or radiotherapy regimens	[1, 2]	NCT00408499, NCT00533949, NCT00673738, NCT01090011, NCT01451632, NCT00397384
Necitumumab	Ongoing Phase II and III	7	Results from the SQUIRE trial presented at American Society of Clinical Oncology 2014 Annual Meeting showed improvements in OS in patients treated with necitumumab plus chemotherapy; ongoing investigations into efficacy in combination therapy	[3, 4]	NCT01763788, NCT02411591, NCT00981058, NCT00982111, NCT02392507, NCT01769391, NCT01788566
c-MET blocking antibodies					
Onartuzumab	Ongoing Phase II and III	5	A Phase III trial in combination with erlotinib for MET-positive NSCLC stopped due to futility	[6]	NCT01887886, NCT01519804, NCT02031744, NCT01456325, NCT01496742
Ficlatuzumab	Ongoing Phase II	1	Ongoing trial in combination with chemical inhibition of EGFR in EGFR mutant NSCLC	[9]	NCT02318368
VEGF-targeted antibodies					
Bevacizumab	Completed Phase II, III; ongoing Phase II, III, IV	48	Evaluation in combination chemotherapy, vaccine and radiation regimens	[10–12]	NCT02054052, NCT00324805
Ramucirumab	Completed Phase II; ongoing Phase III	4	Preliminary results from ongoing Phase III trial have demonstrated increases in OS and PFS	[14]	NCT01168973, NCT01703091, NCT01160744, NCT02411448

c-MET: MET receptor; EGFR: epidermal growth factor receptor; NSCLC: non-small-cell lung cancer; VEGF: vascular endothelial growth factor; OS: overall survival; PFS: progression-free survival.

Table 1. Phase II and phase III studies of selected antigen-specific immunotherapeutic approaches in nonsmall-cell lung cancer

Investigational agent	Phase of study	*N*	Patients	Primary end point	Primary end point outcome		Significance of differences between treatment group and control group
					Treatment group	Control group	
Tecemotide (MUC-1 epitope)	Randomized phase II (Butts and Maksymiuk et al. [12])	171	IIIB or IV NSCLC SD or OR after first-line chemotherapy or chemoradiation	OS	17.2 m	13 m	NS
	Randomized, double-blind placebo-controlled phase III (Butts and Socinski et al. [14])	1513	IIIA (T3, N2 only), IIIB and IV SD or OR after first-line chemotherapy or chemoradiation	OS	25.6 m	22.3 m	NS
Belagenpumatucel-L (4 irradiated cell-lines with TGF-b2 antisense)	Randomized, dose-variable phase II (Nemunaitis et al. [7])	75	II, IIIA, IIIB and IV; low tumor burden Completed conventional therapy	OS	Dose-related improvements in survival in three treatment arms[a]	NA	No control arm
	Randomized, double-blind placebo-controlled phase III (Giaccone et al. [8])	532	IIIA (T3, N2 only), IIIB and IV SD or OR after primary platinum-based chemoradiotherapy	OS	20.3	17.8	NS
Melanoma-associated antigen-A3 vaccine (MAGE-A3 protein)	Randomized phase II (Vansteenkiste [15])	182	Completely resected IB/II MAGE-A3-expressing tumor	DFI	HR 0.74 (95% CI 0.44–1.20) $P = 0.107$[b]	NA	NS
	Randomized, double-blind placebo-controlled phase III (release 2014)	2312	Completely resected IB, II, or IIIA MAGE-A3-expressing tumor	DFS	Not available	Not available	NS

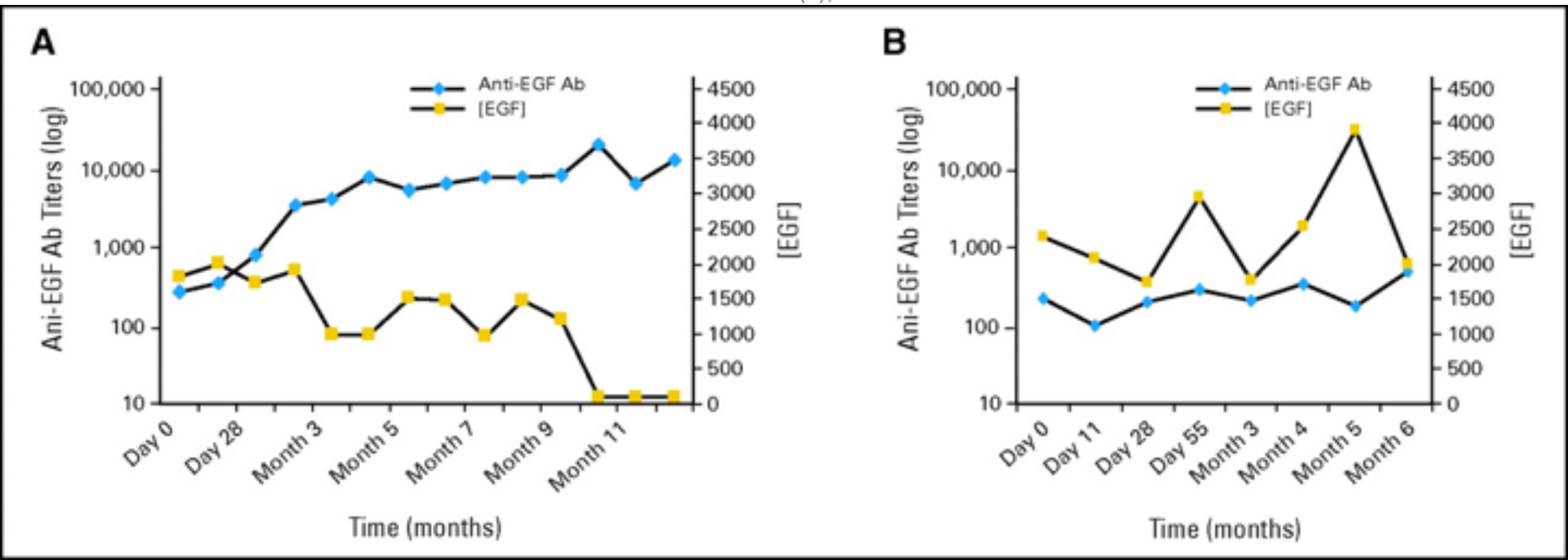
CIMAvax-EGF

- First therapeutic cancer vaccine for non-small cell lung cancer
- Immunization with EGF–*Neisseria* P24 protein (+adjuvant)
- Center of Molecular Immunology, Cuba (25 y to develop)
- Preventive?
- Available in Cuba
- Testing to begin in US, Japan, etc.

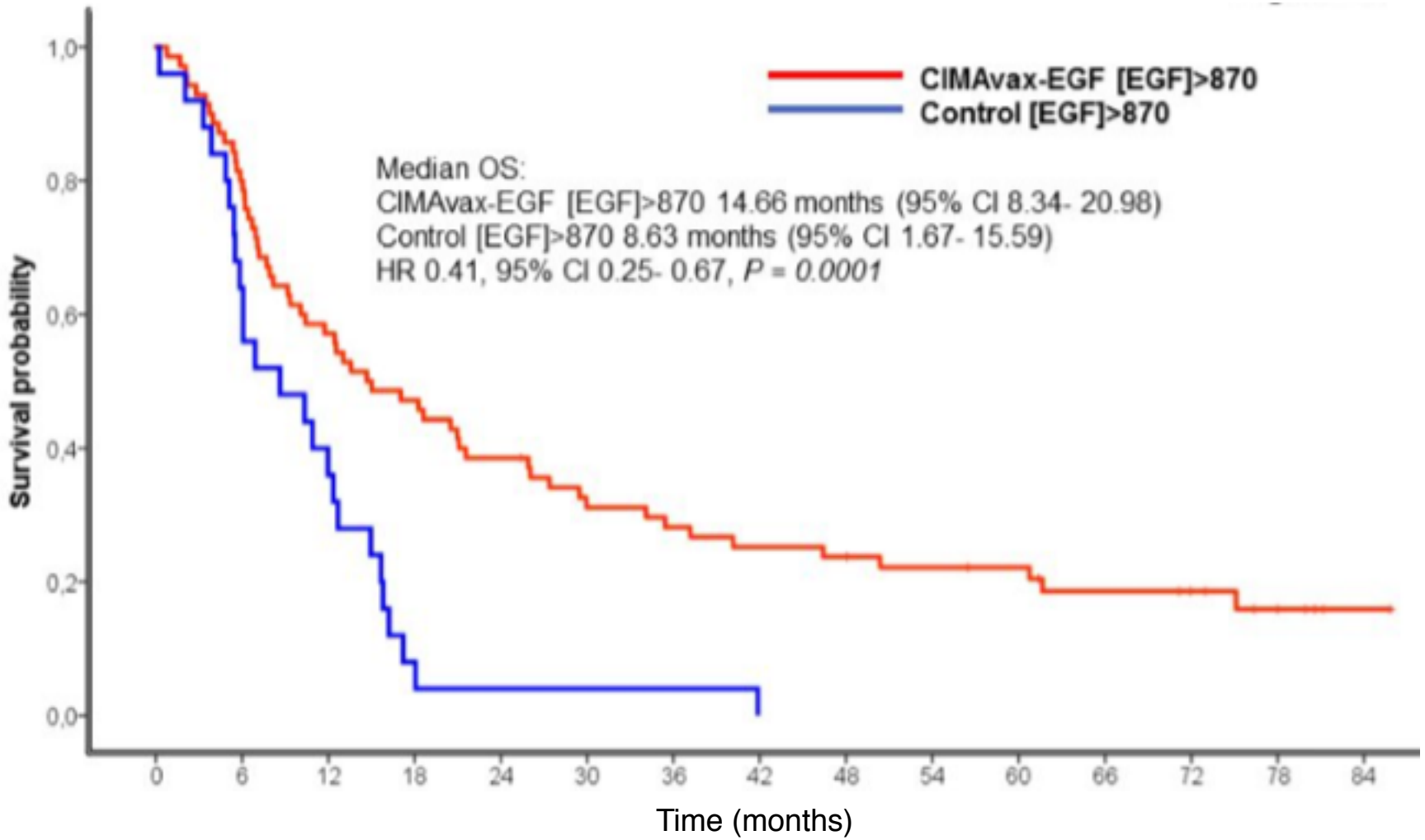
Vaccinated *Control*

stage IIIB/IV NSCLC

JCO 26(9), 2008



Phase III, randomized trial
Stage IIIB/IV NSCLC

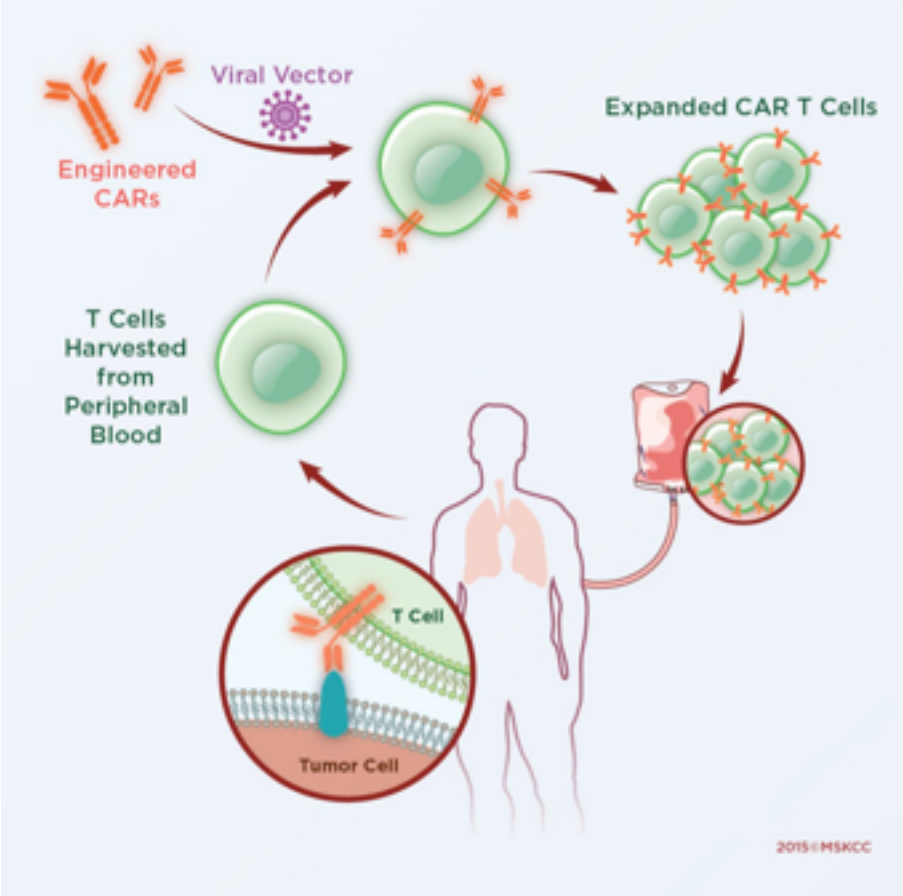


Clin Cancer Res, 2016 Feb 29

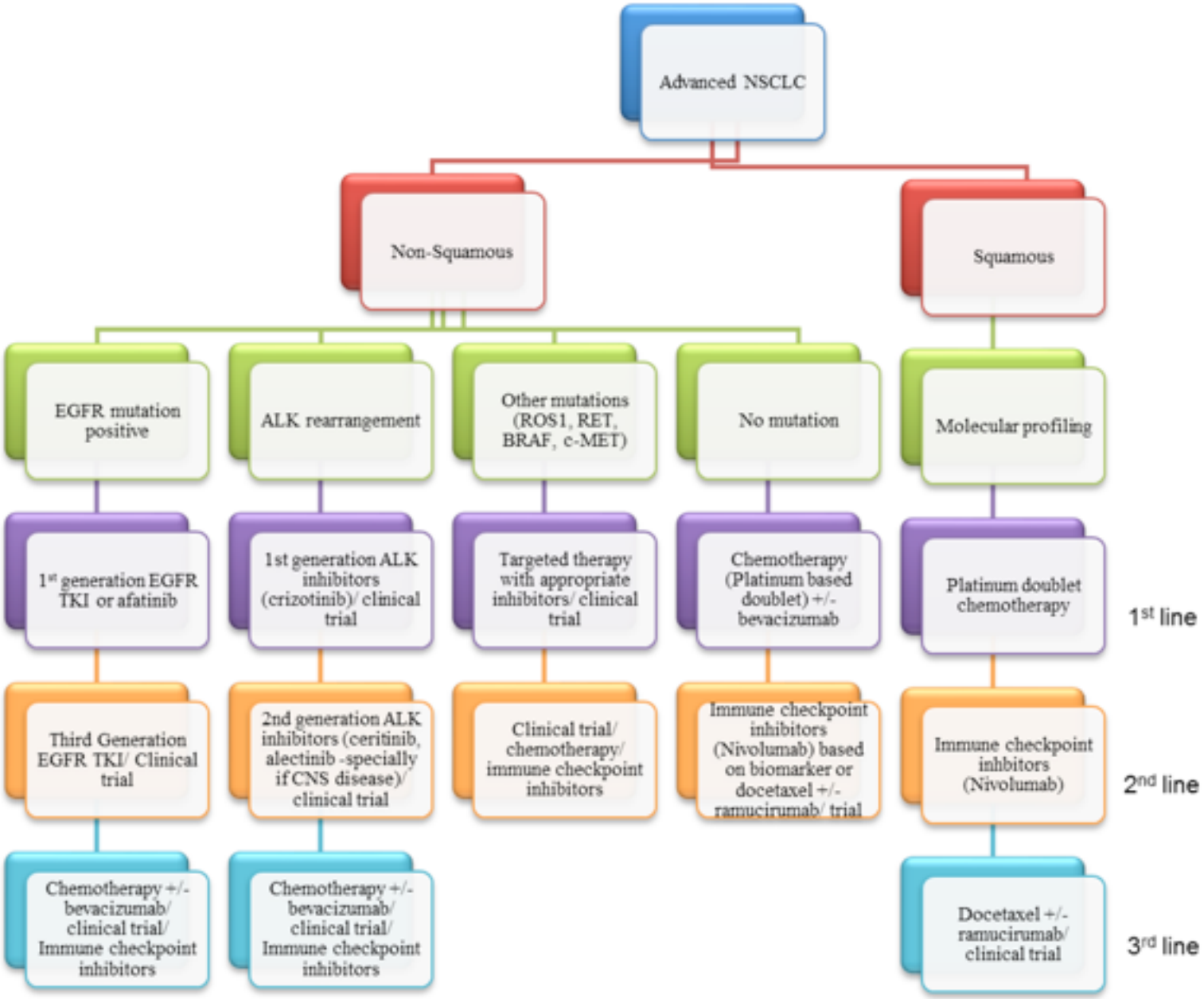
Adoptive cell therapy
Isolate immune cells > engineer in vitro > inject

1. Introduce chimeric antigen T cell receptor (CAR) against cancer antigens

2. Expand killer or infiltrating cell populations

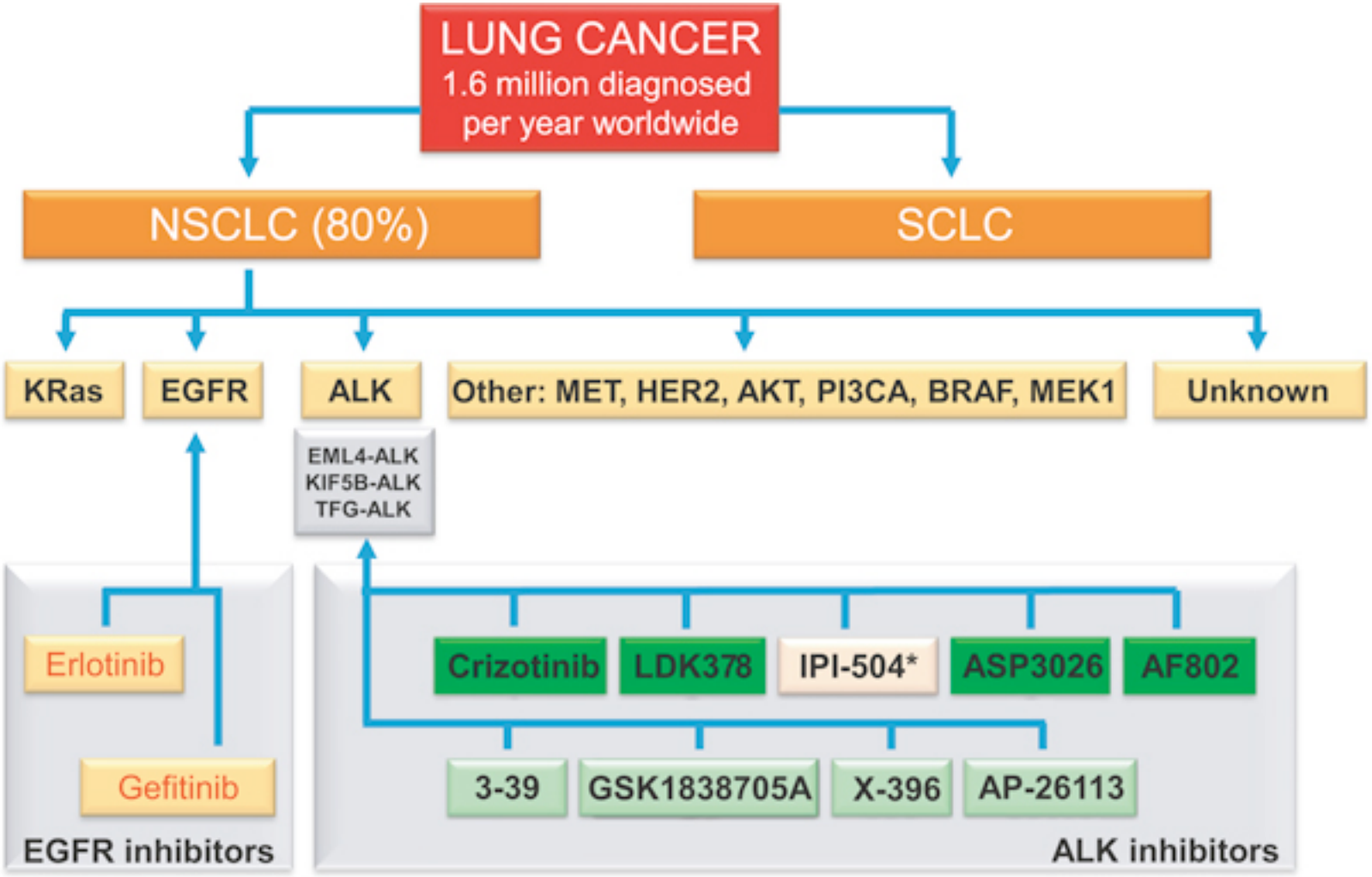


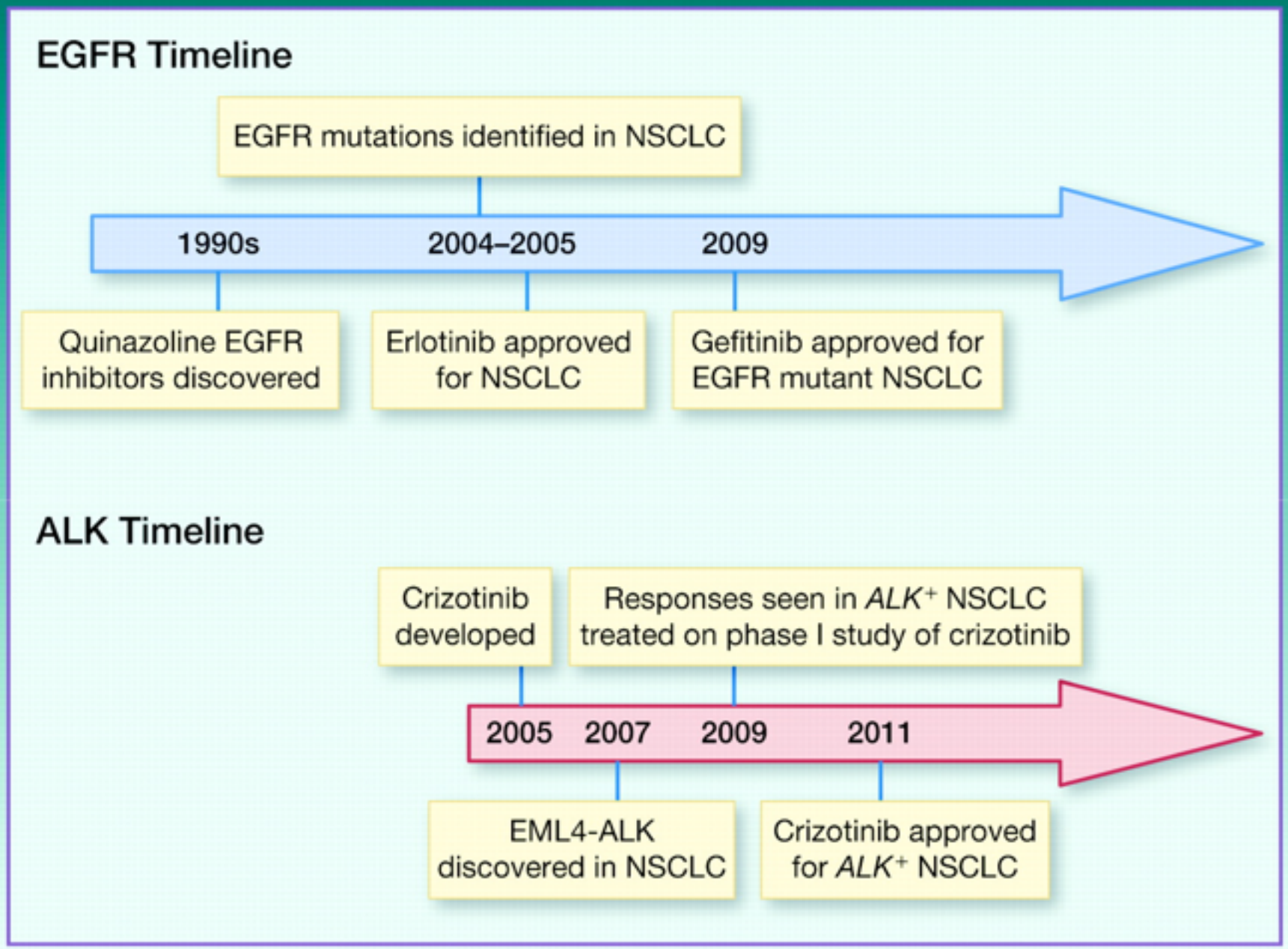
Lung cancer therapy



Cancers 2015, 7(3)

Targeted therapies



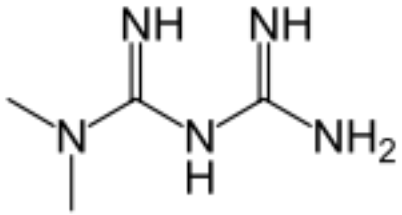


Preventive medicines

Vitamins, anti-oxidant, anti-inflammatory, anti-lipid...

Metformin

Biguanide

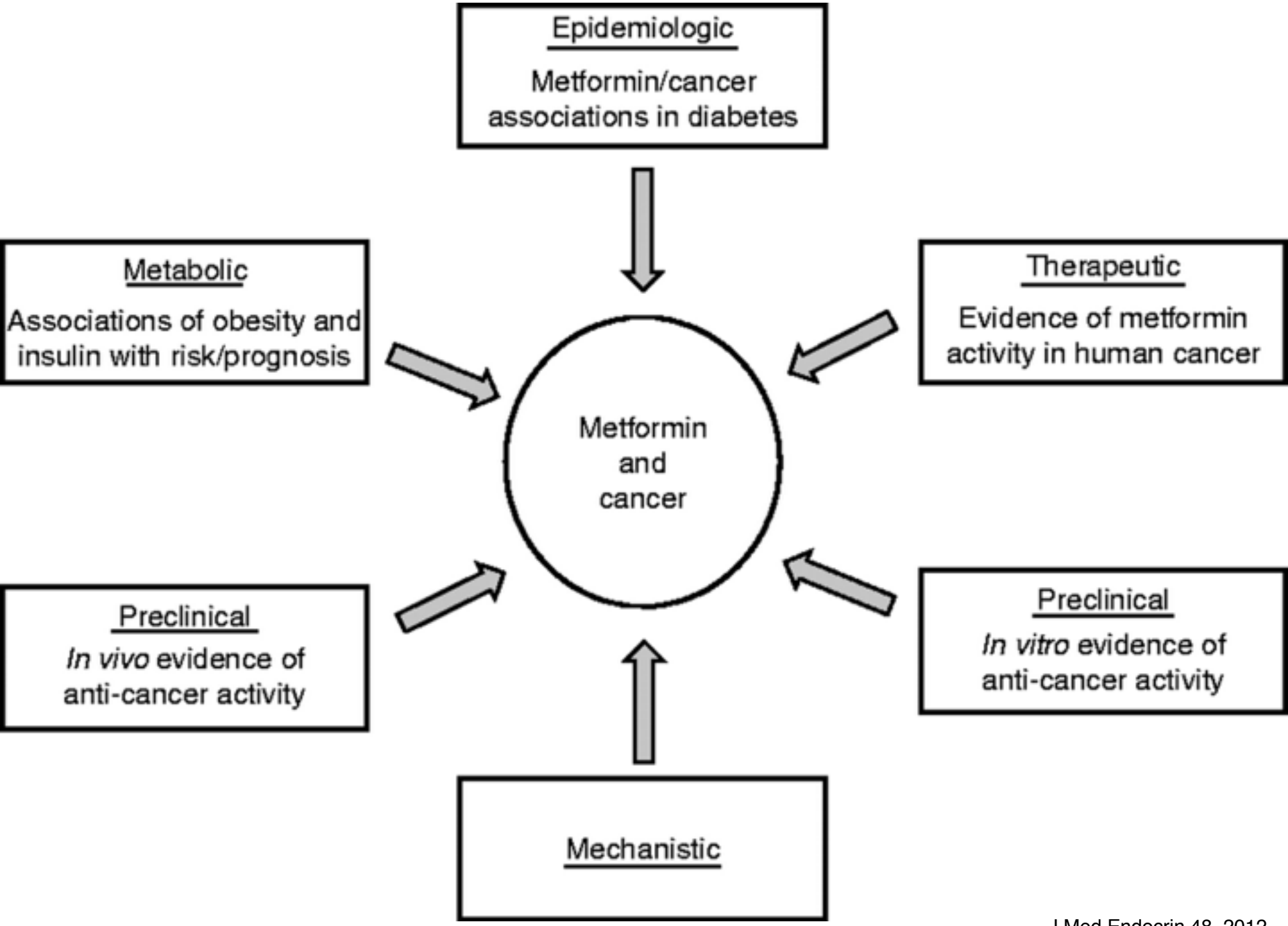


French lilac used against diabetes for centuries

Synthesized in 1920s
Hypoglycemic effect noticed in 1920s
Anti-diabetic trial 1957
US use 1995

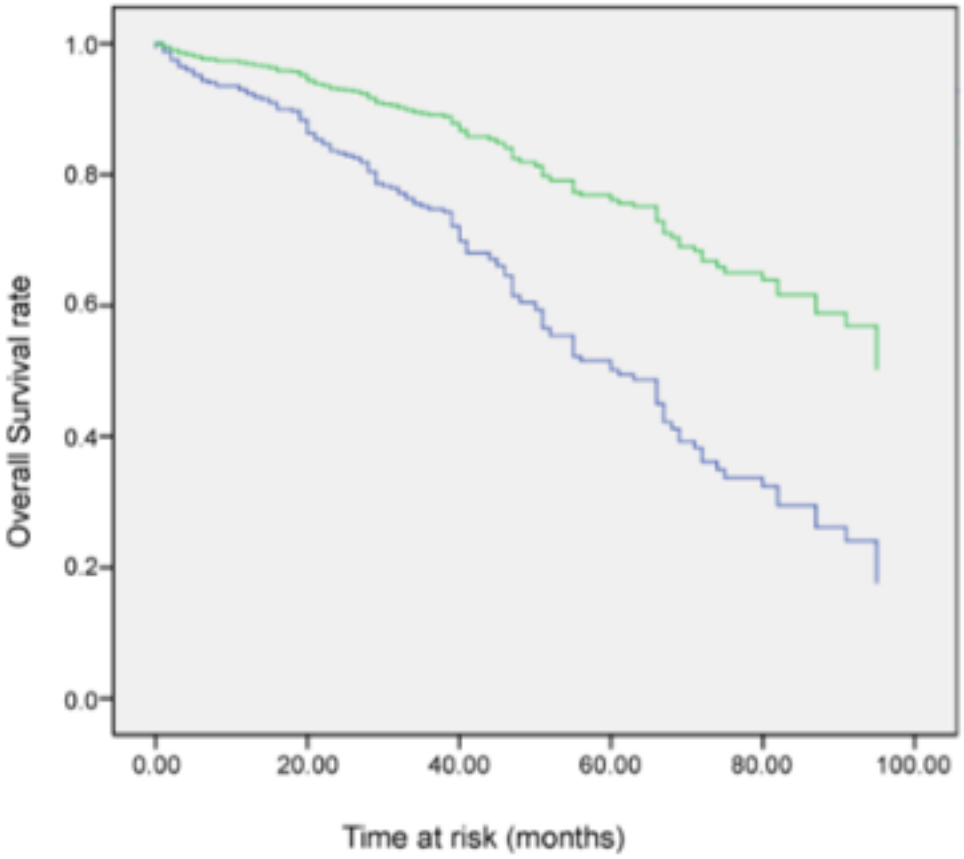
Now most widely used anti-diabetic

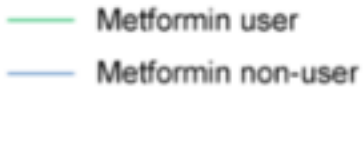
Metformin may reduce
occurrence of cancer
cancer mortality, recurrence,
metastases...
variety of cancers



J Med Endocrin 48, 2012

Metformin and survival after early-stage NSCLC





Ovarian cancer. Cancer. 2013 Feb 1;119(3):555-62

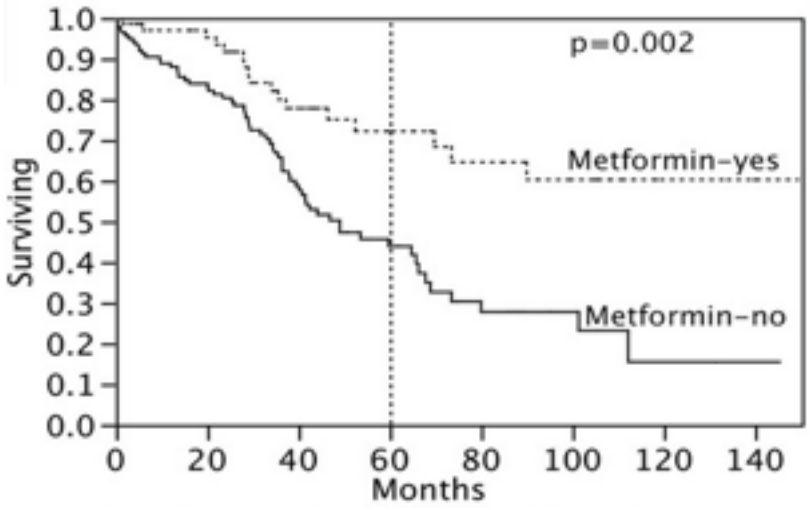
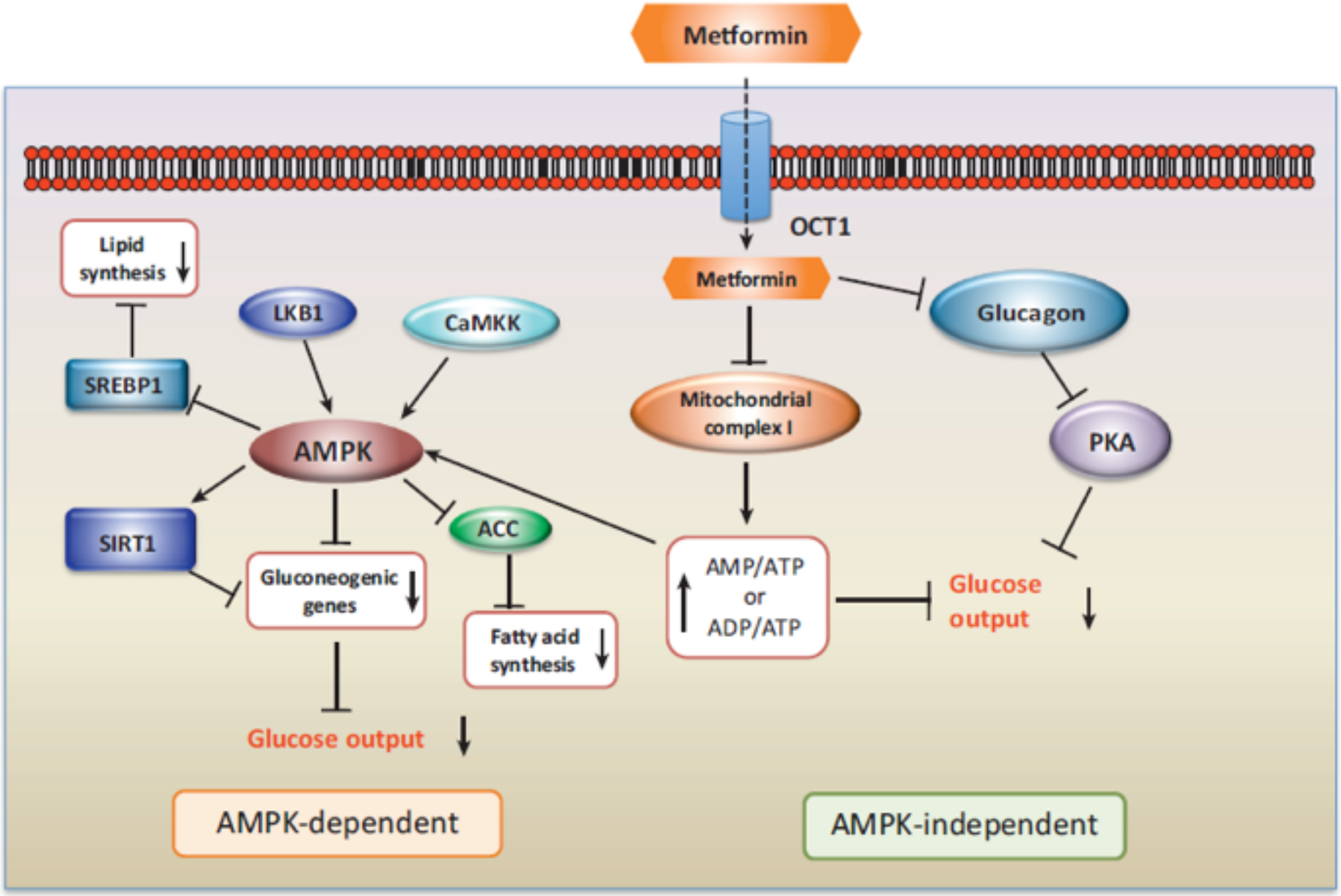


Figure 2.
Kaplan-Meier plot showing significantly better OS in patients on metformin after controlling for the effect of age, gender, race, stage, smoking and histology.

J Cancer Sci Ther. Author manuscript; available in PMC 2015 October 07.



TRENDS in Endocrinology & Metabolism

Questions?