

# Marijuana Smoke

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# Identity of Marijuana Smoke



- Smoke created when flowers, leaves, stems, seeds, and/or resins of marijuana plants (genus *Cannabis*) are burned
- Mixture of thousands of compounds
  - Gas phase, particulate phase, and semi-volatile
  - Organic and inorganic (including metals)
  - ~350 constituents analytically identified (Table 1)
  - 60+ cannabinoids (e.g.,  $\Delta^9$ -THC)
  - 33 are Proposition 65 carcinogens (Table 6)



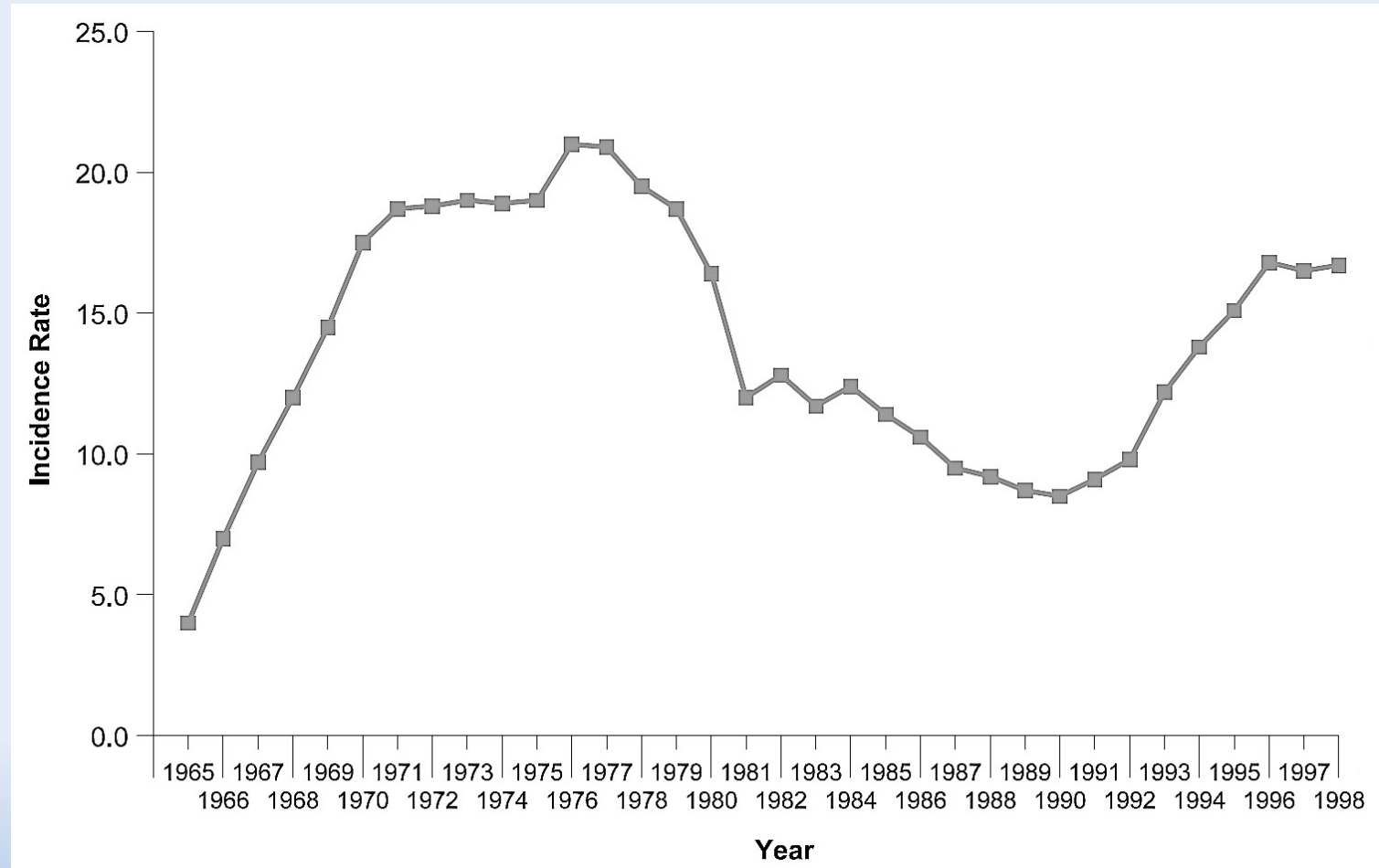
# Occurrence and Use



- Smoked in many parts of the world for thousands of years.
- In the U.S. and other western cultures, marijuana smoking became popular in the late 1960s and 1970s.
- Legal under California state law since 1996 for physician-recommended purposes (Proposition 215, Compassionate Use Act).
- Mixed with tobacco in some parts of the world.



# Marijuana first-time use rates per 1,000 per year, U.S., 1965-1998



Gfroerer *et al.*, 2002. Excludes children age <12.



# Controlled Cancer Studies in Humans (n=26)

- Direct marijuana smoking: n=21
- Parental marijuana smoking (mothers and/or fathers): n=6



# Issues of Validity in Studies in Humans



- Information bias from:
  - Under-reporting of marijuana smoking
    - Due to illegality, social stigma, employment restrictions, and lack of privacy during interviews
    - Cancer cases may under-report less than healthy controls
  - Categorizing people with little exposure as exposed
  - Proxy interviews of fathers
- Confounding bias from adding tobacco to marijuana
- Selection bias from nonparticipation
- Effect modification from cancer latent period (e.g., 20+ yrs)



# Cancer Categories Reported for Direct Marijuana Smoking

(Numbers of studies statistically significant/total)

- Acute myeloid leukemia (0/1)
- Anus (0/1)
- **Bladder (1/2)**
- **Brain (1/1)**
- Breast (0/1)
- Cervix (0/1)
- Colorectal (0/1)
- Esophagus (0/1)
- **Head and neck (2/4)**
- Larynx (0/1)
- **Lung (3/6)**
- Non-Hodgkin's lymphoma, (0/2)
- Melanoma (0/1)
- Oral cavity (0/4)
- Penis (0/1)
- Pharynx (0/1)
- Prostate (0/1)
- **Testis (1/1)**
- "Tobacco-related" (0/1)



# Studies of Lung Cancer in Humans

First Author and Year	Geographic Location	Example Results	
		MS Exposure	Rate Ratio Estimate
Hsairi 1993	Tunis	<i>Habitual smoker</i>	<b>8.2 (1.3-15.5)*</b>
Sidney 1997	San Francisco and Oakland	<i>7+ joints</i>	<b>0.9 (0.5-1.7) men</b> <b>1.1 (0.5-2.6) women</b>
Sasco 2002	Casablanca	<i>Ever</i>	<b>2.0 (0.6-6.3)</b>
Hashibe 2006	Los Angeles County	<i>60+ joint-years</i>	<b>0.6 (0.3-1.2)</b>
		<i>1+ joint-years</i>	<b>1.1 (0.5-2.6) non-tobacco</b>
Voirin 2006	Tunis	<i>Ever</i>	<b>4.1 (1.9-9.0)</b>
		<i>5+ years</i>	<b>3.4 (1.1-10.1)</b>
Aldington 2008a	New Zealand	<i>Ever</i>	<b>1.2 (0.5-2.6)</b>
		<i>&gt;10.5 joint-years</i>	<b>5.7 (1.5-21.6)</b>

\*95% confidence interval. **Red** = statistically significant with 2-sided probability < 0.05.



# Studies of Head and Neck Cancer in Humans

First Author and Year	Location	Example Results	
		MS Exposure	Rate Ratio Estimate
Zhang 1999	New York City	<i>Ever</i>	<b>2.6 (1.1-6.6)*</b>
Aldington 2008b	New Zealand	<i>Ever</i>	<b>1.0 (0.5-2.6)</b>
Gillison 2008	Los Angeles County	<i>15+ joint-years</i>	<b>2.0 (0.5-7.8)</b> HPV-16 neg <b>6.4 (1.6-26.0)</b> HPV-16 pos
Berthiller 2009@	Seattle, Tampa, Los Angeles, Houston, and Latin America (seven cities)	<i>Ever</i>	<b>0.9 (0.7-1.2)</b> <b>0.9 (0.6-1.4)</b> non-tobacco <b>1.1 (0.5-2.4)</b> non-tob/alc
		<i>&gt;5 joint-years</i>	<b>0.9 (0.5-1.4)</b>

\*95% confidence interval. **Red** = statistically significant with 2-sided probability < 0.05.

@The Berthiller, 2009, article was published after the draft document was completed.



# Studies of Bladder, Brain, and Testicular Cancer

Cancer, First Author, and Year	Geographic Location	Example Results	
		MS Exposure	Rate Ratio Estimate
<b>Bladder</b> Bedwani 1997	Alexandria, Egypt	<i>Ever 1+ times per day and 1+ years</i>	<b>0.4</b> (0.1-2.5)*
<b>Bladder</b> Chacko 2006	Palo Alto and Augusta	<i>Joint-years in regression</i>	<b>p=0.01</b>
<b>Brain</b> Efird 2004	San Francisco and Oakland	<i>Ever</i>	<b>1.9</b> (0.9-4.0)
		<i>Ever 1+ times/month</i>	<b>2.8</b> (1.3-6.2)
<b>Testicular</b> Daling 2009	Seattle/Puget Sound region (3 counties)	<i>Former (ever)</i>	<b>1.2</b> (0.8-1.8) seminoma <b>1.2</b> (0.9-1.7) non-seminoma and mixed
		<i>Current</i>	<b>1.3</b> (0.8-2.1) seminoma <b>2.3</b> (1.3-4.0) non-seminoma and mixed

\*95% confidence interval. **Red** = statistically significant with 2-sided probability < 0.05.



# Childhood Cancer Categories Reported for Parental Marijuana Smoking

(Numbers of studies statistically significant/total)

- Brain (astrocytoma) (0/1)
- **Leukemia, all types**
  - All childhood leukemia (**1**/1)
  - Infant leukemia (**1**/1)
- **Leukemia, acute lymphoblastic** (**1**/1)
- **Leukemia, acute myeloid** (**1**/2)
- **Neuroblastoma** (**1**/1)
- **Rhabdomyosarcoma** (**1**/1)



# Childhood Cancer Studies with Results for Maternal Marijuana Smoking

Cancer, First Author, and Year	Geographic Location	Example Results	
		MS Exposure	Rate Ratio Estimate
Leukemia, acute myeloid Robison et al, 1989	U.S.	<i>5+ times in year before or during pregnancy</i>	<b>“Tenfold”</b> , p=0.005
Neuroblastoma Bluhm 2006	North America	<i>Ever in 10 months before birth</i>	1.4 (0.8-2.5)
		<i>Ever 1<sup>st</sup> trimester</i>	<b>4.8</b> (1.6-16.5)
Rhabdomyosarcoma Grufferman 1993	U.S.	<i>Ever in year before birth</i>	<b>3.0</b> (1.4-6.5)

\*95% confidence interval. **Red** = statistically significant with 2-sided probability < 0.05.



# Childhood Cancer Studies with Results for Paternal Marijuana Smoking

Cancer, First Author, and Year	Geographic Location	Example Results	
		MS Exposure	Rate Ratio Estimate
<b>Leukemia, all types</b> Wen 2000	U.S. and Canada	<i>Ever in year before birth</i>	<b>1.5</b> , p<0.01 <b>2.0</b> , p<0.05 infant
<b>Leukemia, acute lymphoblastic</b> Wen 2000	U.S. and Canada	<i>Ever in year before birth</i>	<b>1.5</b> , p<0.05
<b>Leukemia, acute myelogenous</b> Trivers 2006	U.S. and Canada	Ever	<b>1.4</b> (1.02-1.8)
<b>Neuroblastoma</b> Bluhm 2006	North America	<i>Ever around time of pregnancy</i>	<b>2.0</b> (1.2-3.2)
<b>Rhabdomyosarcoma</b> Grufferman 1993	U.S.	<i>Ever in year before birth</i>	<b>2.0</b> (1.3-3.3)

\*95% confidence interval. **Red** = statistically significant with 2-sided probability < 0.05.



# Carcinogenicity Studies in Animals (n=4)

- Female rats: marijuana smoke inhalation
- Newborn rats: smoke condensate s.c. injection
- Female mice: smoke condensate dermal
  - Cancer bioassay
  - Tumor promotion



# Marijuana Smoke Inhalation Study in Female Wistar Rats (Murthy *et al.*, 1985)

Study Design	Treatment group	Tumor Type	Tumor bearing animals
20/group, 15 min/day, 6 days/week, 36 months	Marijuana smoke	<i>Ovary:</i> Benign serous cytoma Follicular cysts <i>Uterus:</i> Adenofibroma Telangiectatic cysts and polyps Adenosarcoma	50 %
	Control		0 %



# Marijuana Smoke Condensate Injection Study in Newborn CD Rats (Repetto *et al.*, 1979)

Study design	Treatment group	Tumor Type	Tumor incidence
S.C. injection of condensate or vehicle control (ethanol/olive oil) on days 1, 4, 7, 11, 14 and 18 of life	194 mg/kg condensate  Vehicle control	Mixed mesenchymatous tumors (malignant)	Not reported

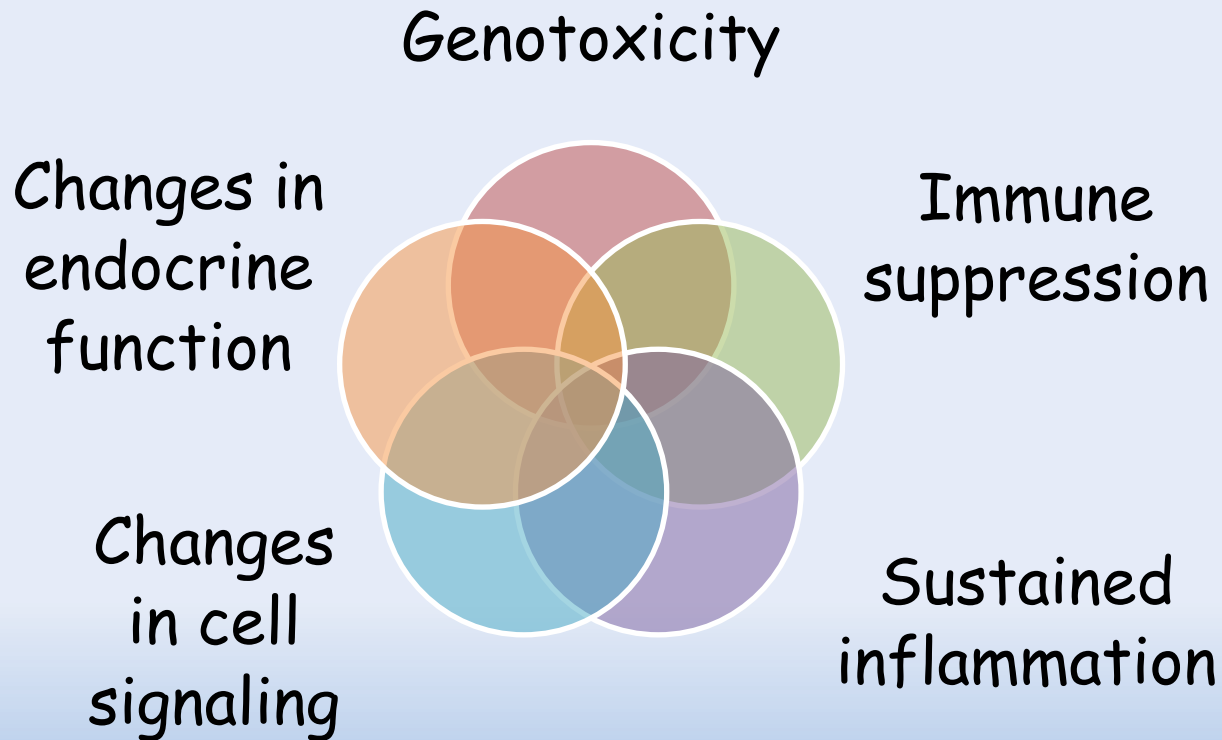


# Dermal Marijuana Smoke Condensate Studies in Female Swiss Mice (Hoffman *et al.*, 1975)

Study Type & Design	Treatment group	Skin Tumor Type	Tumor incidence
Bioassay:  100/group, 3X/week, 74 weeks ≈75 mg “tar”	Marijuana	Squamous cell papilloma	6/99
	Tobacco	Squamous cell papilloma & carcinoma	14/97
	Historical acetone-treated controls	“Skin tumors”	Rarely observed (e.g., ≤ 1%)
Tumor promotion:  60/group, Initiation: 75 µg DMBA Promotion: 3x/week, 56 weeks	Marijuana	Squamous cell papilloma & carcinoma, fibrosarcoma (3)	26/60
	Tobacco	Squamous cell papilloma & carcinoma	34/60
	Initiator only	Squamous cell papilloma & carcinoma	5/60



# Possible Mechanisms of Action



# Genotoxicity

- Increased lymphocyte *hprt* mutation frequency in mothers who smoke marijuana and their newborns
- Increased DNA/chromosome damage (PBL, bone marrow, alveolar macrophages) in some studies
- Marijuana smoke condensate increased mutations in *Salmonella*
- A number of individual chemical constituents of marijuana smoke are genotoxic



# Suppressive Effects on the Immune System

## Marijuana smoke

- Alveolar macrophages from marijuana smokers show reduced tumoricidal and bactericidal activity
  - Similar effects in rats
- Increased progression from HIV infection to AIDS in marijuana smokers



# Immune System Effects (continued)

## $\Delta^9$ -THC

- Reduces thymus & spleen weight and cellularity via CB2-R
- Disrupts
  - Host resistance to microbial infection
  - Macrophage function
  - NK and T cell cytolytic activity
  - Macrophage and T cell cytokine production
- Increases viral hemagglutinin titer, & decreases macrophage, CD4<sup>+</sup>, and CD8<sup>+</sup> T cell counts
- Varied effects on anti-tumor immune response
  - Dependent upon mouse model, cancer cell line



# Histopathological Changes in Lungs

- Marijuana smokers: Inflammation, proliferation and preneoplastic changes
- Rat: Dose-related inflammatory and proliferative lesions
- Dog: Bronchiolitis, metaplasia
- Monkey: Inflammatory fibrosis, metaplasia, hyperplasia



# Preneoplastic Changes in Skin

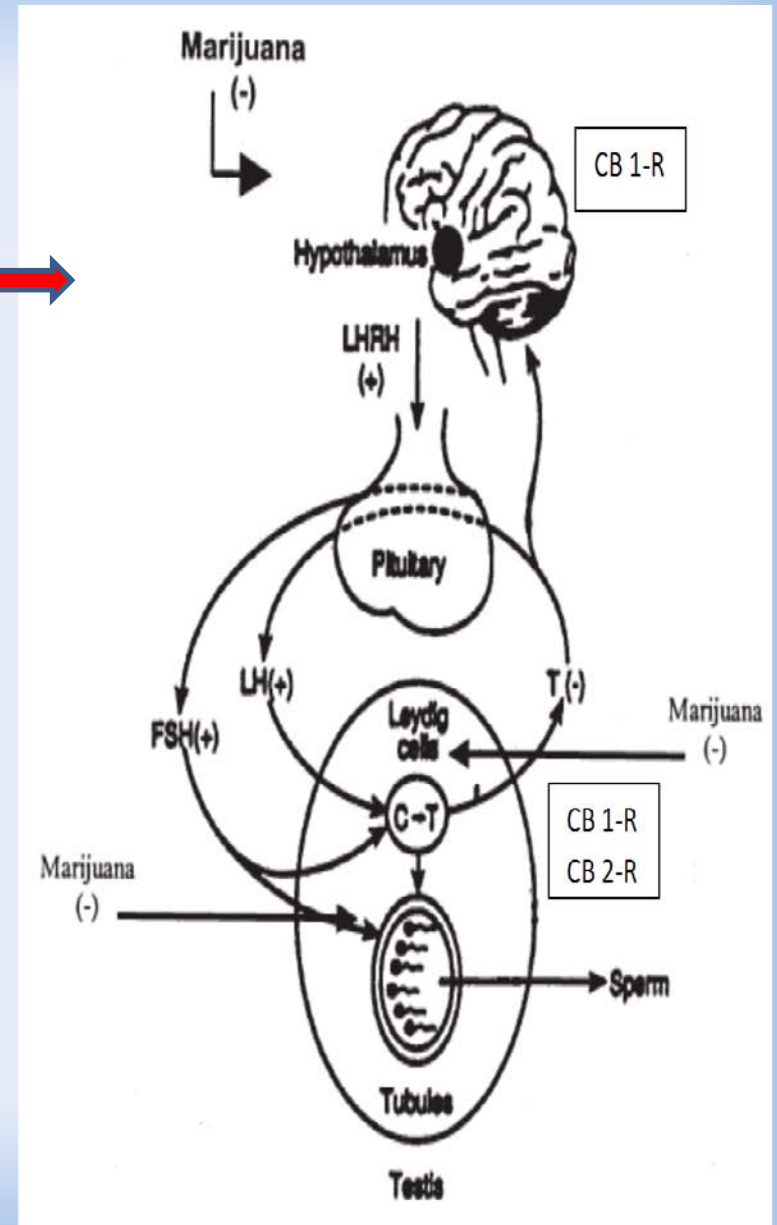
- Mice: Sebaceous gland metaplasia
  - tobacco smoke condensate causes the same lesions



# Effects on Endocrine Systems

Hypothalamic-pituitary-gonadal (HPG) axis

Other effects



# Effects on Endocrine Systems (Continued)

## Hypothalamic-pituitary-gonadal (HPG) axis

### Direct estrogen receptor (ER)-mediated effects

- Uterotropic effects & ER-responsive gene expression

### Indirect ER-mediated effects

- Interaction with the AhR
- AhR cross-talk with the ER: plausible
- Aromatase inhibition

### Cannabinoid receptor (CB-R)-mediated effects

- Hormone secretion: FSH, LH, prolactin, GH, TSH, corticotropin, E2, T, Progesterone
- Cross talk with EGF receptor signaling pathways: cell proliferation

### Androgen receptor (AR)-mediated effects

- AR Antagonism
- Effects on androgen metabolism



# Comparison of Marijuana and Tobacco Smoke

- Similar, except MS contains cannabinoids, TS contains nicotine
- Similar particle size distributions for both
- ~4x greater MS tar deposited in the lungs than TS tar, from similar amount of plant material
- 33 carcinogens present in both
- Similar effects in mouse skin, *Salmonella*, human and dog lungs



# Summary of Evidence: Epidemiological Studies

- Some epidemiological studies suggest increased cancer risk from direct and parental marijuana smoking.
- Limitations of the epidemiological studies include:
  - Small numbers of studies for most cancer types.
  - Potential biases from
    - Mixing of tobacco and marijuana
    - Differential under-reporting of use between cases and controls
    - Low participation
    - Proxy interviews for fathers



# Summary of Evidence: Carcinogenicity Studies in Animals

- MS or its condensate induce skin papillomas in mice, and malignant uterine & mesenchymatous tumors and benign ovarian tumors in rats.
- MS condensate exhibits tumor promoting activity in mouse skin similar to that of TS condensate.



# Summary of Evidence:

## Other Relevant Data

- Studies in smokers suggest that MS induces mutations and chromosomal abnormalities; MS condensate induces mutations in Salmonella, similar to TS condensate.
- MS suppresses multiple parameters of immune function.
- MS affects multiple hormonal and other cell signaling pathways leading cells to potential tumor transformation.
- Marijuana smokers' lungs exhibit lesions similar to those of tobacco smokers, including inflammation, proliferation, and preneoplastic changes.
- MS induces preneoplastic lesions in mouse skin, similar to TS.
- MS contains 33 of the same carcinogenic constituents as TS.

