



# Toxicity Assessment of F-T Jet Fuel (JP-8/SPK)

**LT Vish Mokashi, PhD, MSC, USN**

Naval Health Research Center/ EHEL

2729 R St., Bldg. 837, Area B

Wright Patterson Air Force Base, OH 45433

937-904-9473

[Vishwesh.mokashi@wpafb.af.mil](mailto:Vishwesh.mokashi@wpafb.af.mil)

CDR Gail Chapman, PhD, MSC, USN

Officer-in-Charge EHEL

David R. Mattie, PhD, DABT

AFRL/RHPB

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# Toxicity Assessment of F-T Jet Fuel (JP-8/SPK)

- Background on major jet fuels
- Background on JP-8/SPK
- Development of the JP-8/SPK toxicity program
- Dermal Irritation
- Genotoxicity
- Inhalation
- Conclusions
- Questions

# Background

➤ DoD consumes ~ 5 B gallons of aviation jet fuel per year (US consumer ~25B)

## ➤ Major Jet Fuels:

➤ Jet-A

➤ Commercial

➤ JP-4

➤ Issued 1951

➤ 50/50 kerosene-gasoline blend

➤ Contains aliphatic and aromatic hydrocarbons, corrosion inhibitor and anti-static and -icing compounds

➤ Phased out by USAF 1976-1996, replaced with JP-8

➤ JP-5

➤ US Navy Fuel

➤ Primarily kerosene

➤ Lower volatility

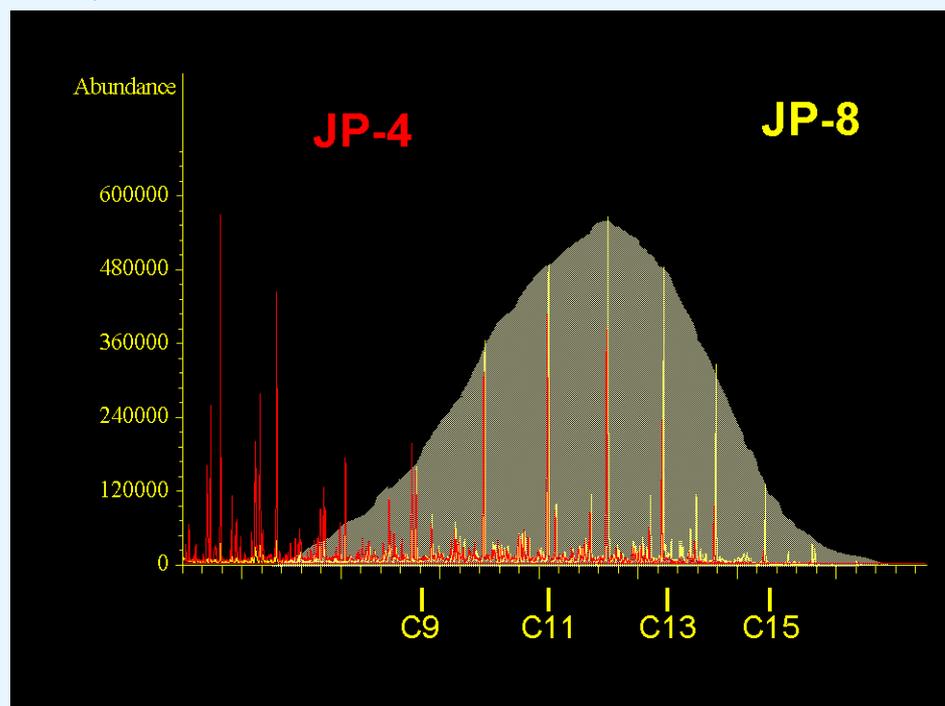


# Background

## ➤ Major Jet Fuels:

### ➤ JP-8 (MIL-DTL-83133)

- Kerosene-based replacement for JP-4
- Mixture of aliphatic, aromatic, and substituted naphthalene hydrocarbon compounds
- Reduced concentrations of benzene, n-hexane
- Contains icing inhibitor, corrosion inhibitors, lubricants, and anti-static agents
- +100 thermal stability additive can also contain metal deactivators, anti-oxidants



Erica Becvar  
AFCEE/TDV

John Hinz, Health Risk  
Assessment Support  
USAF School of Aerospace  
Medicine

# Background



- Jet fuel toxicity risks traditionally associated with aromatic hydrocarbons
  - Conventional petroleum-based military aviation fuels contain ~20% Aromatics and 1-3% naphthalene.
- JP-4
  - toxicity studies have reported skin irritation, defatting, neurotoxicity nephrotoxicity and, in male rats, renal carcinogenicity.
  - In humans, symptoms of neurasthenia, psychasthenia, polyneuropathy and sexual dysfunction has been reported in military and civilian workers.

# Background

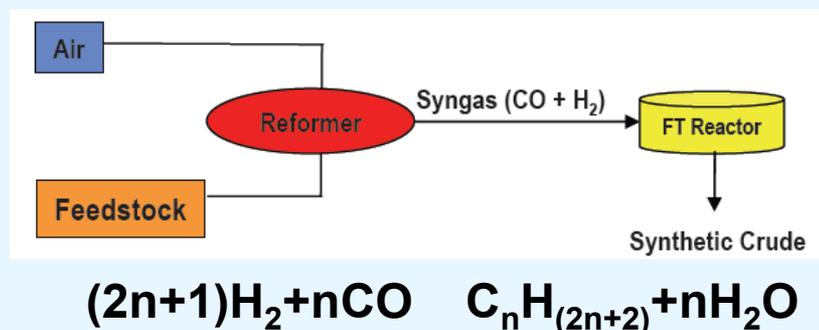
## ➤ JP-8 Toxicity

- Considered slightly toxic
- Oral Lethal Dose ( $LD_{50}$ ) > 5.0g/kg (highest tested)
- Dermal – non-irritating to slightly irritating
  - Repeated and continuous exposure does cause irritation in animals and humans
- Acute Vapor Inhalation Lethal Concentration ( $LC_{50}$ ) > 3.43 mg/L (highest tested)
- Acute Aerosol  $LC_{50}$  > 4.39 mg/L (highest tested)
- Subchronic Inhalation- decreased body weight and renal hydrocarbon nephrothopy in male rats
- Chronic Inhalation – male rat nephrothapy



# Background

- **JP-8/SPK (aka F-T, S-8...)**
  - USAF initiated the “alternative fuels” program in 1999
    - MIL-HDBK-510 Aerospace Fuels Certification
    - Goal of system certification and use of 50:50 synthetic jet fuel:JP-8 blend by 2016
  - To be used as a “drop in” fuel (JP-8:JP-8/SPK (50:50))
    - Synthetic jet fuel derived converting natural gas using coal-to-liquid (CTL) technology
    - Current synthetic jet fuels (S-8) alkane-rich, have reduced risk - essentially no aromatics or naphthalene



# Background

## ➤ JP-8/SPK (aka F-T, S-8...)

- B-52 Certification (Aug 07)
- C-17 Ground/flight demos and FSE complete
- F101 engine (B-1B) test at AEDC
- CFM56 (F108) engine (KC-135) operability testing
- B-1B Demo flight – late Spr 08
- Acceleration of USAF Comm-Derivative aircraft certification by FAA Type-Cert (Dec 08?)
- F-22 certification effort - complete Fall 08
- F119 engine testing at P&W (May 08)
- F-16, KC-135, B-2 cert planning in-process



# F-T Jet Fuel Toxicity Assessment



- Funded by Air Force
- Goal - Complete initial program by end FY08
- To ensure that toxicity program addresses all issues associated with testing a complex mixture, such as jet fuel, a review panel of toxicologists was established to discuss the toxicity testing program and provide advice on testing procedures.

# F-T Jet Fuel Toxicity Assessment

## ➤ Panel members:

- David Mattie, Ph.D., DABT, Chair, AFRL/RHPB
- John Hinz, AFIOH/RSRE
- Gunda Reddy, PhD, USACHPPM
- LT Dean Wagner, PhD, MSC, USN, NHRC/EHEL
- David Steup, PhD, Shell Oil and Chairman of the Toxicology Committee, API
- Errol Zeiger, Ph.D., J.D., Errol Zeiger Consulting, Chapel Hill, NC

# F-T Jet Fuel Toxicity Assessment

- **Develop appropriate study design for each toxicity test proposed:**
  - *In vitro* genotoxicity tests
  - Dermal irritation test
  - Acute inhalation study
  - *In vivo* genotoxicity test in tandem with inhalation rangefinder study
  - 90-day inhalation toxicity study

# F-T Jet Fuel Toxicity Assessment

- All studies are completed.
- Final analysis and/or report preparation stage.
- Occupational exposure level will be developed by the review panel for the F-T fuel during the last quarter of FY09.

# F-T Jet Fuel Toxicity Assessment

## ➤ Dermal Irritation Test

- Dermal Irritation Test
  - GLP Compliance
  - 100% concentrations of either SPK or JP-8
  - 50/50 mixture of SPK/JP-8
  - Occluded and semi-occluded exposures (each animal its own control)
  - Primary irritation index calculated to assign a descriptive rating to each test article and mixture

# Dermal Toxicity

- ◆ JP-8 Occluded
- ◆ Primary Irritation Index = 2.1      Descriptive Rating = Moderately Irritating
- ◆ JP-8 Semi-Occluded
- ◆ Primary Irritation Index = 1.8      Descriptive Rating = Slightly Irritating
- ◆ SPK Occluded
- ◆ Primary Irritation Index = 2.3      Descriptive Rating = Moderately Irritating
- ◆ SPK Semi-Occluded
- ◆ Primary Irritation Index = 0.8      Descriptive Rating = Slightly Irritating
- ◆ JP-8/SPK Occluded
- ◆ Primary Irritation Index = 1.9      Descriptive Rating = Slightly Irritating
- ◆ JP-8/SPK Semi-occluded
- ◆ Primary Irritation Index = 1.5      Descriptive Rating = Slightly Irritating

# F-T Jet Fuel Toxicity Assessment

## *In vitro* genotoxicity tests

Performed by Eurofins/ Product Safety Labs

GLP Compliance

# F-T Jet Fuel Toxicity Assessment

## ➤ **Chromosomal Aberration (CA) Test**

- Establish potential for fuel to induce chromosomal aberrations in human lymphocytes with and without activation.

## ➤ **Conclusion**

- Chromosomal aberrations were not induced by the fuel
- JP8/SPK is considered to be non-clastogenic

# F-T Jet Fuel Toxicity Assessment

- **Reverse Mutation Assay (Ames Test)**
- Establish potential for fuel to induce gene mutations
- Detect point mutations in *Salmonella typhimurium*
  - Histidine reversion system His<sup>-</sup> → His<sup>+</sup>

# F-T Jet Fuel Toxicity Assessment

## ➤ **Reverse Mutation Assay (Ames Test)**

### ➤ Preliminary Results

- No toxic effects in any of five tester strains
- No relevant increase in revertants
- F-T jet fuel did not cause base pair or frame shift genetic mutations in any of the tester strains

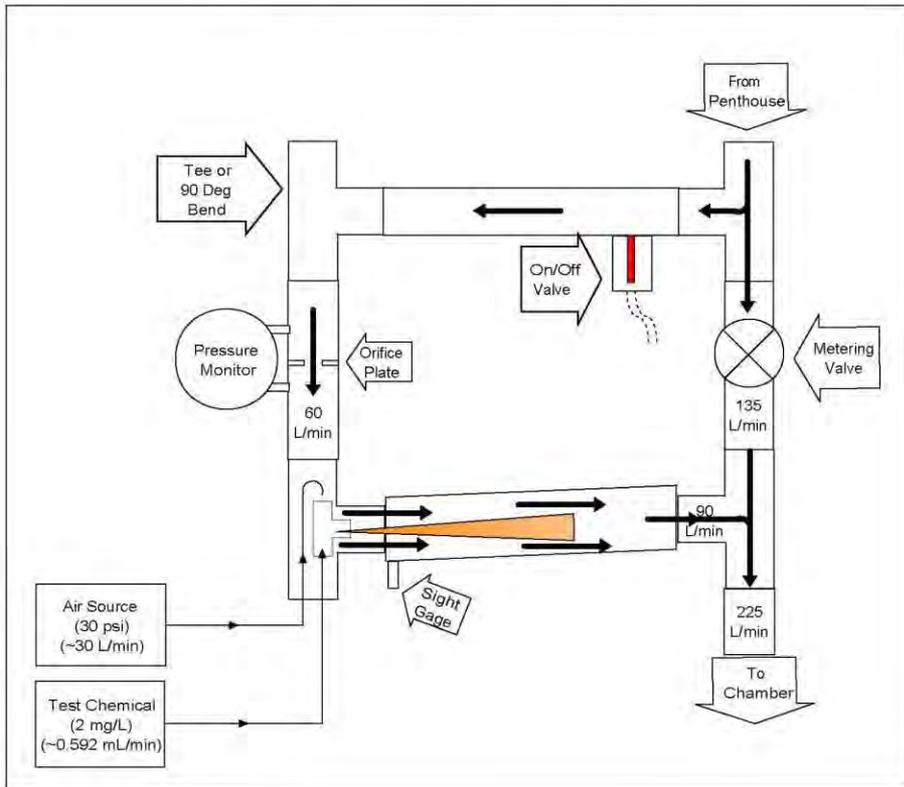
## ➤ **Conclusion**

- F-T jet fuel is considered to be non-mutagenic in this bacterial reverse mutation assay

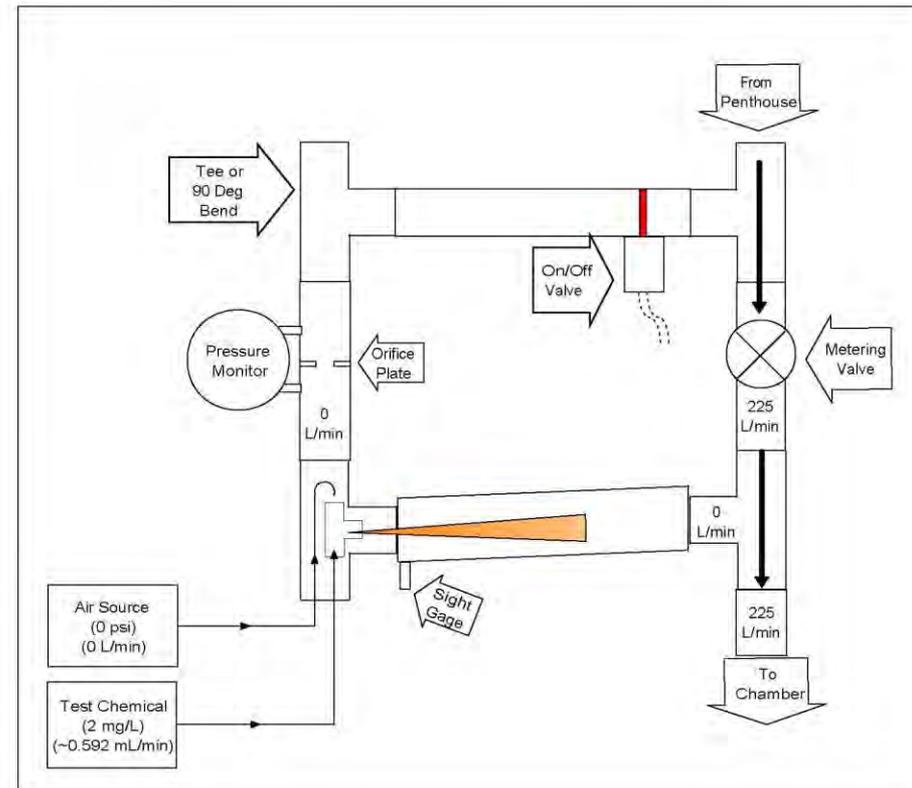
# F-T Jet Fuel Toxicity Assessment

## Inhalation studies

Protocol 07034: Generation of Jet Fuel: On/Off Valve in On Position



Protocol 07034: Generation of Jet Fuel: On/Off Valve in Off Position



# F-T Jet Fuel Toxicity Assessment

## ➤ Acute Inhalation Study

### ➤ Completed:

➤ 4 hour exposure

➤ 2000 mg/m<sup>3</sup> dose

➤ 5 males/5 females

➤ No clinical symptoms observed

# F-T Fuel Toxicity

## 2-Week Study

- **2-week - Histological findings in:**
  - **lung: Inflammatory foci evident in highest doses**
  - **Olfactory epithelia: degeneration in highest doses**
  - **Kidney: hyaline droplet accumulation in all males exposed**
  - **Liver: Panlobular hepatocyte changes in all males exposed and females exposed to the highest concentration**

# F-T Jet Fuel Toxicity Assessment

## ➤ 90-day Inhalation Study

### ➤ Study design based on:

➤ U.S. Environmental Protection Agency (U.S. EPA)  
Harmonized Test Guideline developed by the Office of  
Prevention, Pesticides and Toxic Substances (OPPTS)  
870.3465 90-Day Inhalation Toxicity and

➤ Organization for Economic Cooperation and Development  
(OECD) guideline 413, Subchronic Inhalation Toxicity:

➤ 90-day Study

➤ U.S. EPA Health Effects Test Guideline OPPTS 870.6200  
Neurotoxicity Screening Battery.

# F-T Jet Fuel Toxicity Assessment

## ➤ 90-day Inhalation Study

### ➤ Started April 28

### ➤ Route, Duration and Frequency of Administration

➤ Test substance will be administered as aerosol/vapor combination for 6 h/d, 5 d/w for 13 w

### ➤ Exposure Levels

➤ Animals will be exposed to three concentrations, high, intermediate and low, with a control group (0 mg/mm<sup>3</sup>).

➤ 2000 mg/m<sup>3</sup>, 700 mg/m<sup>3</sup> and 200 mg/m<sup>3</sup>

# F-T Jet Fuel Toxicity Assessment

## ➤ 90-day Inhalation Study

### ➤ Observations

- Animals will be observed before and after exposures for overt signs of toxicity
- Tissues and organs will be examined for gross pathology
- Blood will be collected for clinical pathology
- Tissues collected for histopathology including:
  - Nasal airways, trachea, larynx, lungs, liver, kidney, spleen, adrenals, heart and others

# F-T Jet Fuel Toxicity Assessment

## ➤ 90-day Inhalation Study

### ➤ Motor Activity and Functional Observational Battery

➤ To assess the neurobehavioral effects

### ➤ Sperm Morphology and Vaginal Cytology Examinations

➤ Sperm will be stained and examined for percentage of abnormal sperm

➤ Alterations in the estrous cycle will be assessed by examination cytology from vaginal smears

# F-T Jet Fuel Toxicity Assessment

## 90-day Inhalation Study

Group	Exposure Level mg/m <sup>3</sup>	Number of Animals	
		Males	Females
Control Replicate 1	0	5	5
Control Replicate 2		5	5
Low Replicate 1	200	5	5
Low Replicate 2		5	5
Intermediate Replicate 1	700	5	5
Intermediate Replicate 2		5	5
High Replicate 1	2000	5	5
High Replicate 2		5	5
Total		40	40

# 90-Day Inhalation Study

- Observations
  - Animals observed before and after exposures for overt signs of toxicity
  - Tissues and organs examined for gross pathology
  - Blood collected for clinical pathology
  - Full tissue list collected for histopathology
    - Analyzed male rat kidneys for alpha 2 microglobulin

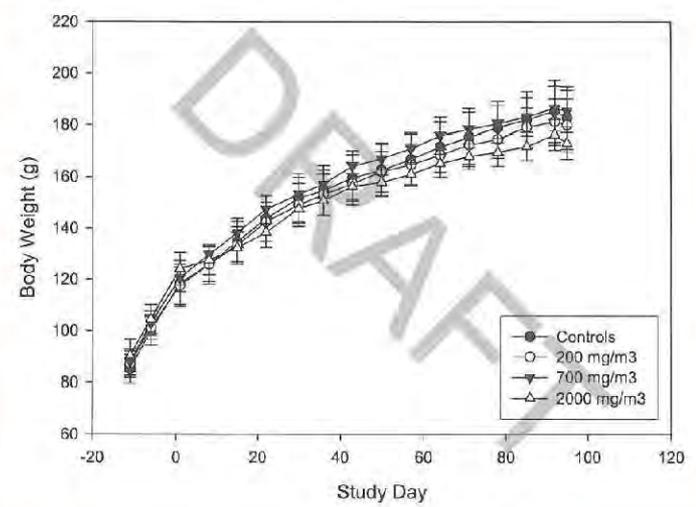
# 90-Day Inhalation Study

## Mean Exposure Levels

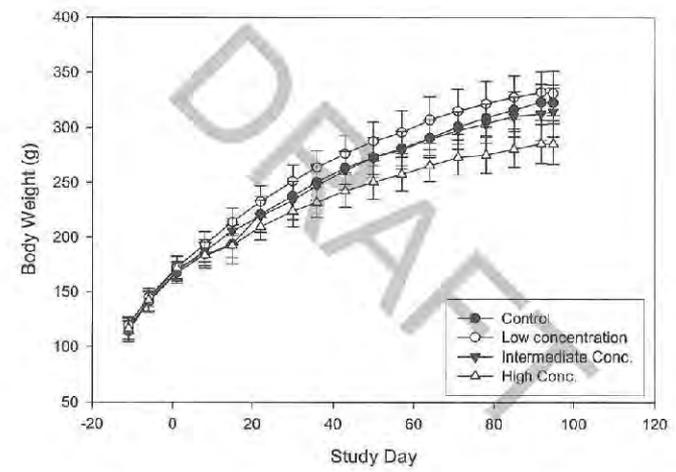
Group	Exposure Level	Number of Animals	
		Males	Females
Control	$0.02 \pm 0.10$	10	10
Low	$200.1 \pm 5.0$	10	10
Intermediate	$698.6 \pm 16.8$	10	10
High	$1988.4 \pm 48.1$	10	10

# Body Weights

Protocol 08013 Females  
**DRAFT**

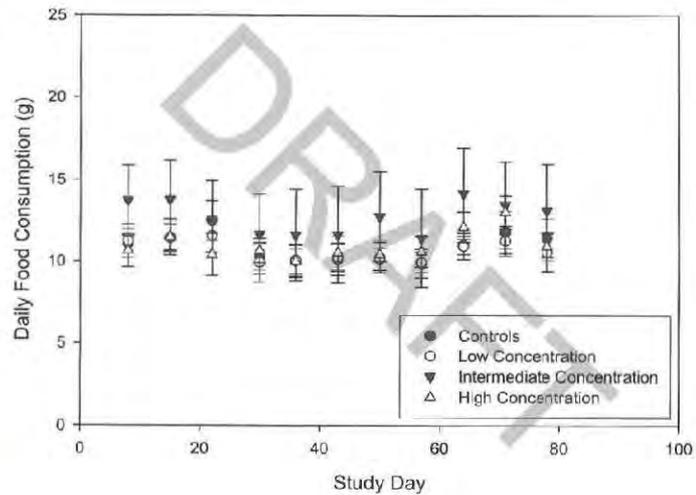


Male Body Weights (without outliers)  
**DRAFT**

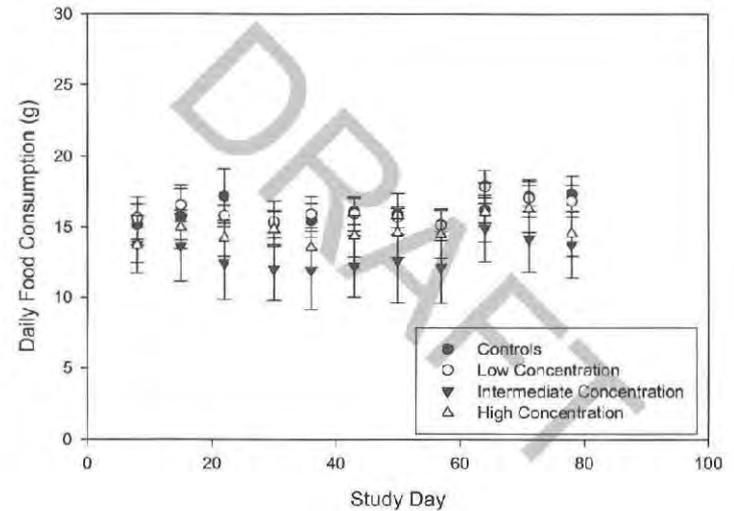


# Food Consumption

Protocol 08013 Average Daily Food Consumption, Females, mod  
**DRAFT**



Protocol 08013 Average Daily Food Consumption, Males, mod  
**DRAFT**



# Hematology

- ◆ There were no biologically significant or toxicologically significant changes in hematology or clinical chemistry values seen in any of the groups.

# Nasal Histopathology

Table 3 Incidence of selected findings in the noses of male rats (ten per group)

Group	Olfactory Epithelial Degeneration	Hypertrophy Hyperplasia Goblet Cells Nasopharyngeal Duct
Control	0	0
Low	0	0
Intermediate	9	10
High	10	10

Table 4 Incidence of selected findings in the noses of female rats (ten per group)

Group	Olfactory Epithelial Degeneration	Hypertrophy Hyperplasia Goblet Cells Nasopharyngeal Duct
Control	0	0
Low	0	0
Intermediate	8	9
High	10	10

# Lung Histopathology

Table 5 Incidence of Inflammatory Cell Infiltration in the lungs of male rats (ten per group)

Group	Multifocal Inflammatory Cell Infiltration	Focal Inflammatory Cell Infiltration
Control	0	0
Low	0	1
Intermediate	0	1
High	10	0

Table 6 Incidence of Inflammatory Cell Infiltration in the lungs of female rats (ten per group)

Group	Multifocal Inflammatory Cell Infiltration	Focal Inflammatory Cell Infiltration
Control	0	0
Low	0	0
Intermediate	0	3
High	10	0

# Kidney Histopathology

Table 7 Incidence of selected findings in the kidneys of male rats (ten per group)

Group	Increased Hyaline Droplet Tubular Epithelium
Control	0
Low	0
Intermediate	0
High	10

# Alpha 2 Microglobulin

Group	Exposure Level	$\alpha_{2\mu}$ - globulin Concentration ( $\mu\text{g}/\text{mg}$ )	
		Males	Females
Control	$0.02 \pm 0.10$	$49.43 \pm 20.87$	$0.16 \pm 0.05$
Low	$200.1 \pm 5.0$	$49.72 \pm 20.57$	N/A
Intermediate	$698.6 \pm 16.8$	$50.90 \pm 13.72$	N/A
High	$1988.4 \pm 48.1$	$35.65 \pm 8.49$	$0.15 \pm 0.01$

# F-T Jet Fuel Toxicity Assessment

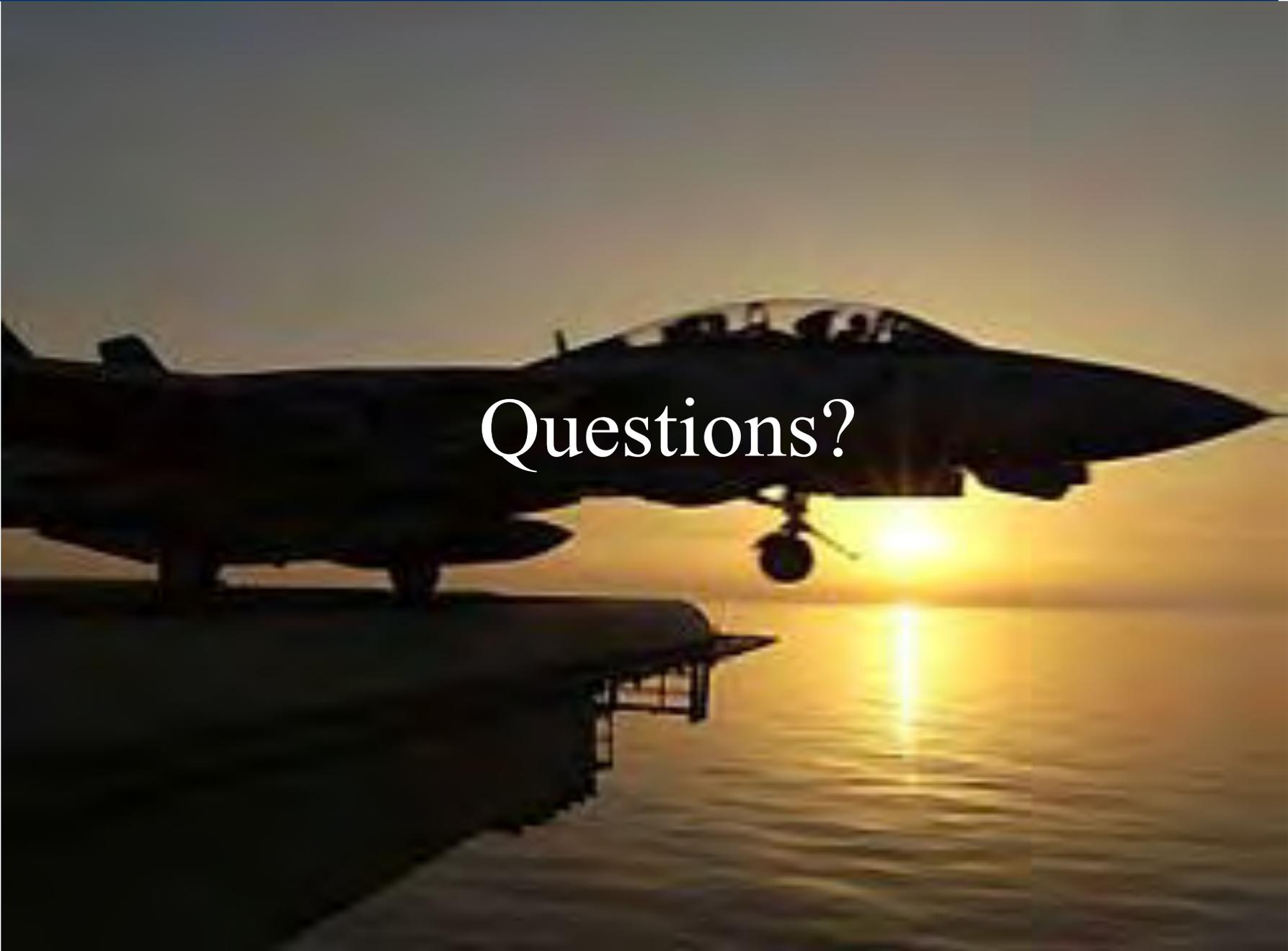
## Conclusions:

- **Dermal - animal protocol approved, start date TBD**
- **Genotox - No genetic mutations observed in vitro**
- **Inhalation**
  - **Acute - No clinical symptoms observed**
  - **2-week - Histological findings in:**
    - **lung: Inflammatory foci evident in highest doses**
    - **Olfactory epithelia: degeneration in highest doses**
    - **Kidney: hyaline droplet accumulation in all males exposed**
    - **Liver: Panlobular hepatocyte changes in all males exposed and females exposed to the highest concentration**
  - **90-day- Started 28 Apr 08**

# F-T Jet Fuel Toxicity Assessment

## **Next:**

- **Develop HHA**
- **Sensory Irritation Assay**
- **Genetic Biomarkers of Exposure**
- **Immunotoxicity Screen**

A silhouette of a fighter jet, likely an F-35, is positioned on the deck of an aircraft carrier. The scene is set at sunset, with the sun low on the horizon, creating a bright, golden glow that reflects on the water. The jet is the central focus, with its wings and tail clearly visible against the bright sky. The overall mood is contemplative and serene.

Questions?