Chemical & Bioassay Analyses of Emissions from Biodiesel Fuel Combustion

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Research Objectives

- Study biodiesel and renewable diesel emissions:
  - Chemical characterization of toxics
  - Toxicity studies of emissions

Unregulated Toxic Emissions

- PAHs
- Alkyl PAHs
- Nitro-PAHs
- Selective reactive aldehydes

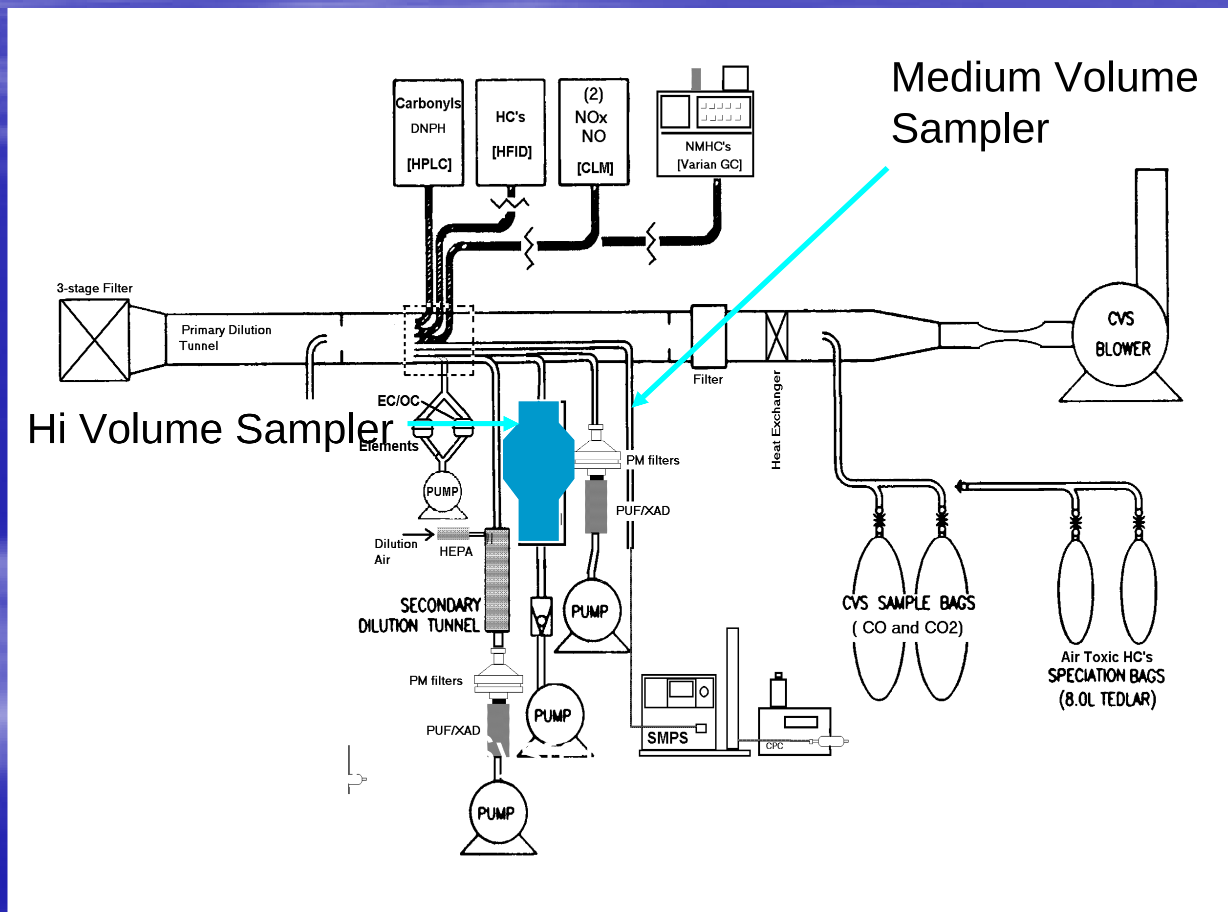
Toxicity Studies of Emissions

- Tests for markers of inflammation in human cells
- Tests for genotoxicity
  - Mutagenicity
  - Chromosomal Damage

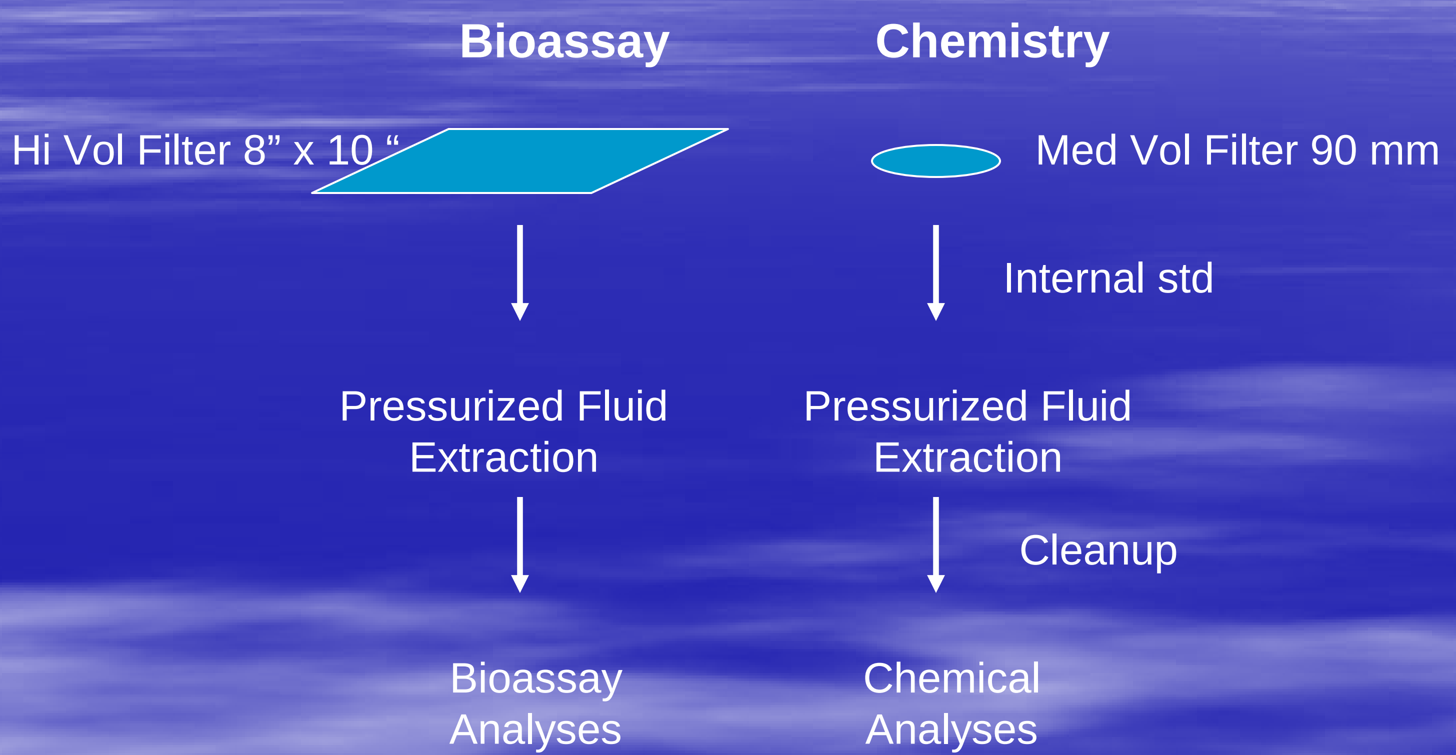
Test Vehicles

Vehicle/Engine	Engine Displacement (L)	Control Devices	Test Cycle	Fuels Tested
2000 Freightliner C15 Caterpillar	15	-	UDDS	CARB Diesel, Soy, Animal, and Renewable @ 20%, 50% and 100%
2008 Freightliner Mercedes Benz MBE 4000	12.8	DOC, DPF, EGR	UDDS	CARB Diesel, Soy, Animal, and Renewable @ 20%, 50% and 100%

Methods


Medium Volume Sampler
Hi Volume Sampler
Carbonyls
DNPH
[HPLC]
HC's
[HFID]
(2)
NOx
NO
[CLM]
NMHC's
[Varian GC]
3-stage Filter
Primary Dilution Tunnel
EC/OC
Elements
PUMP
Dilution Air
HEPA
SECONDARY DILUTION TUNNEL
PM filters
PUF/XAD
PUMP
PUMP
PUMP
PM filters
PUF/XAD
SMPS
CPC
Filter
Heat Exchanger
CVS BLOWER
CVS SAMPLE BAGS
(CO and CO2)
Air Toxic HC's
SPECIATION BAGS
(8.0L TEDLAR)

PM Filter Preparation for Analyses



PUF XAD Preparation for Analyses

Bioassay

PUF for Bioassay

Pressurized Fluid Extraction

Bioassay Analyses

Chemistry

PUF for Chem

Internal std

Pressurized Fluid Extraction

Cleanup

Chemical Analyses

XAD (Chem only)

Internal std

Liquid Extraction with Shaking

Cleanup

Chemical Analyses

Methods

Chemical Analyses

PAHs, Alkyl PAHs → GC/MS - EI

Nitro-PAHs → GC/MS -NCI

Methods

Bioassay Analyses

Genotoxicity

Human Cell Markers of Inflammation

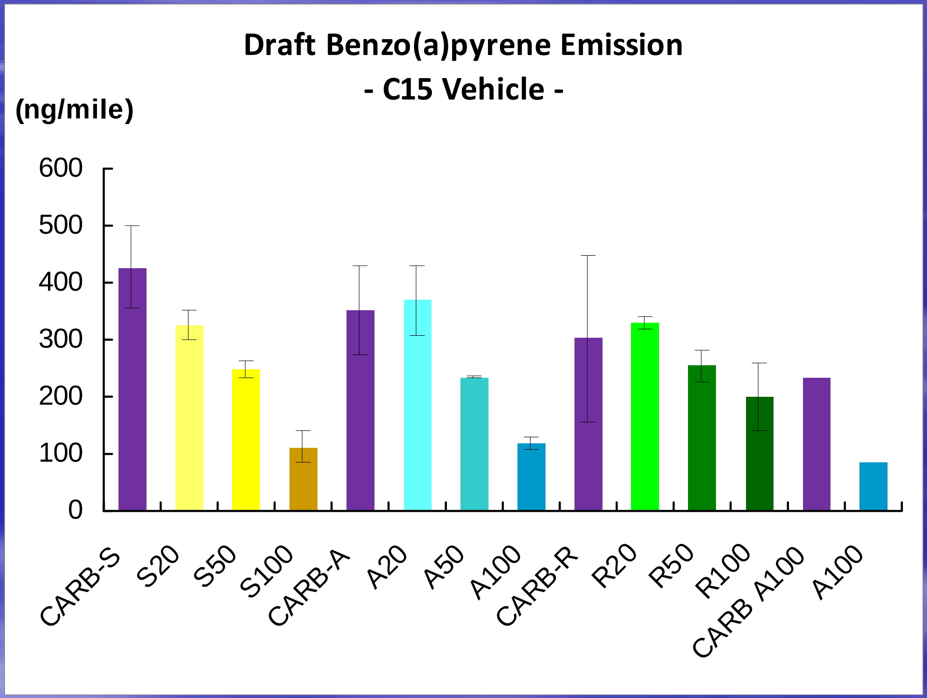
Chemical Analyses

- PAHs
- Alkyl PAHs
- Nitro-PAHs

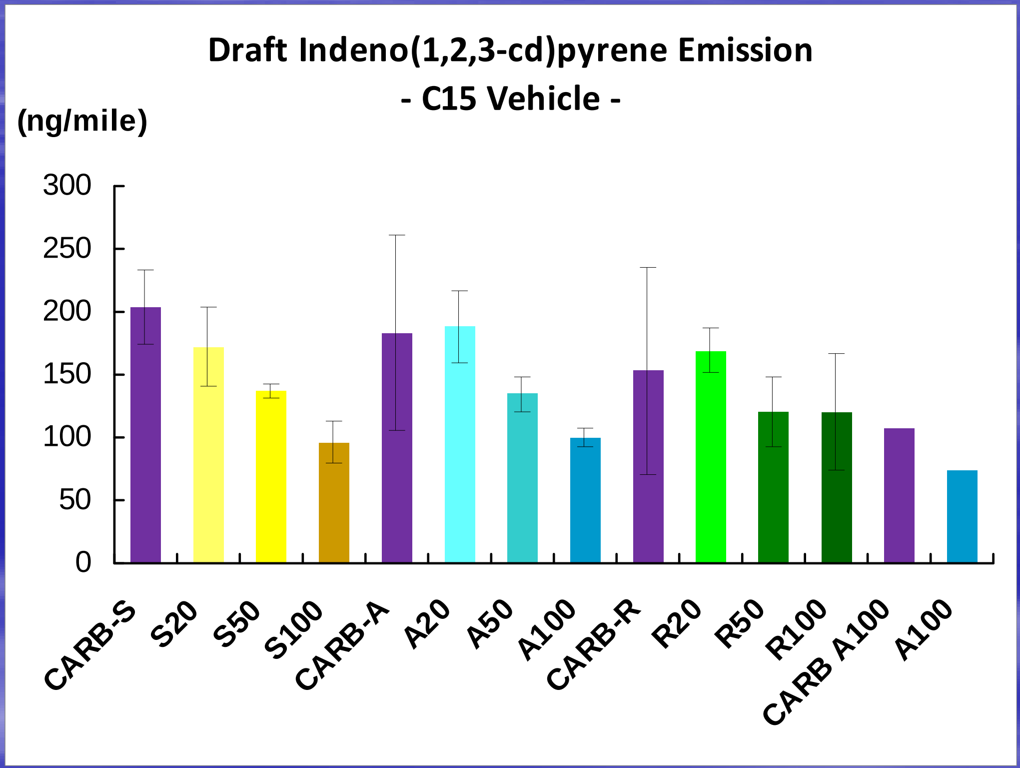
Compounds Analyzed

PAHs	Alkyl PAHs	Nitro-PAHs
Naphthalene	2-Methylnaphthalene	1N-naphthalene
Acenaphthylene	1-Methylnaphthalene	2N-naphthalene
Acenaphthene	2,6-Dimethylnaphthalene coelute	5N-acenaphthene
Fluorene	1,6-Dimethylnaphthalene	2N-fluorene
Phenanthrene	2,3,5-Trimethylnaphthalene coelute	9N-anthracene
Anthracene	3-Methylphenanthrene	3N-phenanthrene
Fluoranthene	2-Methylphenanthrene	2N-phenanthrene
Pyrene	9-Methylphenanthrene	3N-fluoranthene
Benz(a)anthracene	1-Methylphenanthrene	1N-pyrene
Chrysene+triphenylene	2-Methylanthracene	7N-BaA
Benzo(b+j+k)fluoranthenes coelute	2-Methylfluoranthenes	6N-chrysene
Benzo(e)pyrene	1-Methyl & 3-Methylfluoranthenes	6N-BaP+1N-BeP
Benzo(a)pyrene	4-Methylpyrene	
Perylene	1-Methylpyrene	
Indeno(1,2,3-cd)pyrene	7,12-Dimethylbenz(a)anthracene	
Dibenz(a,h)anthracene		
Benzo(g,h,i)perylene		
Dibenzo(a,l)pyrene		

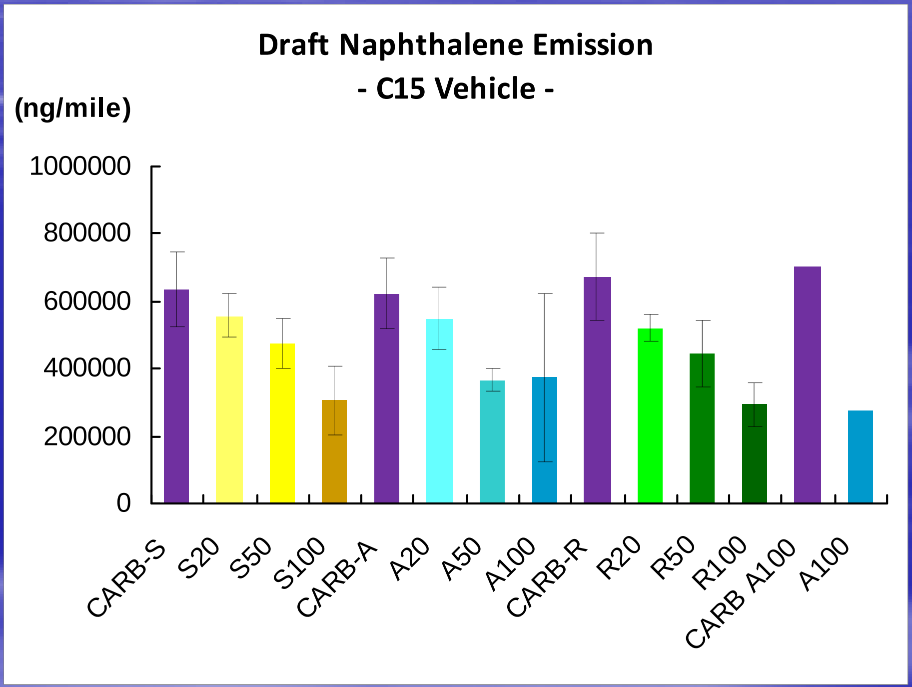
PAH Emissions
PM Associated PAHs
C15 Vehicle



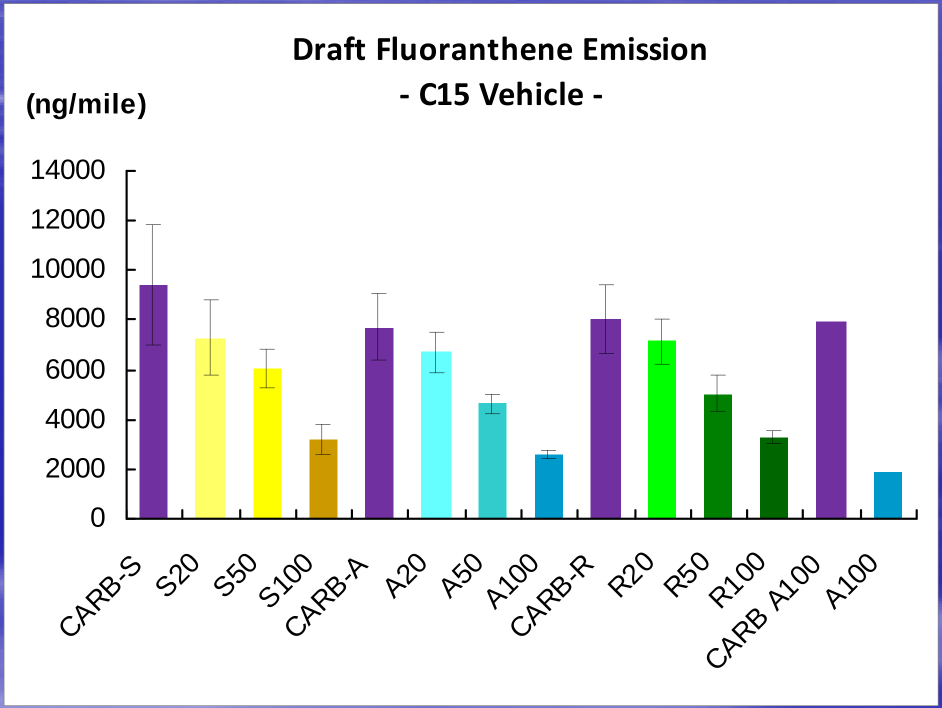
PAH Emissions
PM Associated PAHs



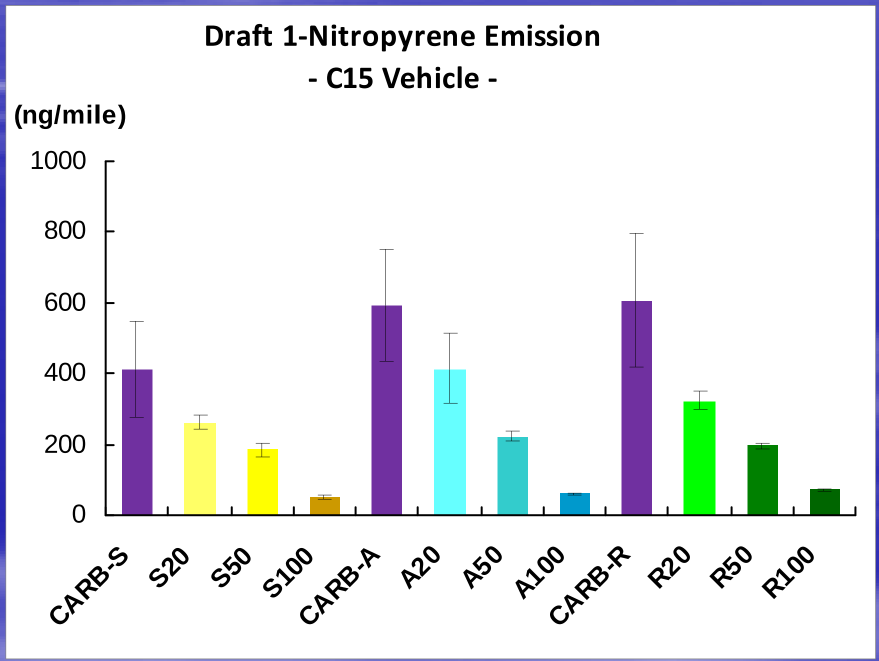
PAH Emissions
Vapor-Phase PAHs



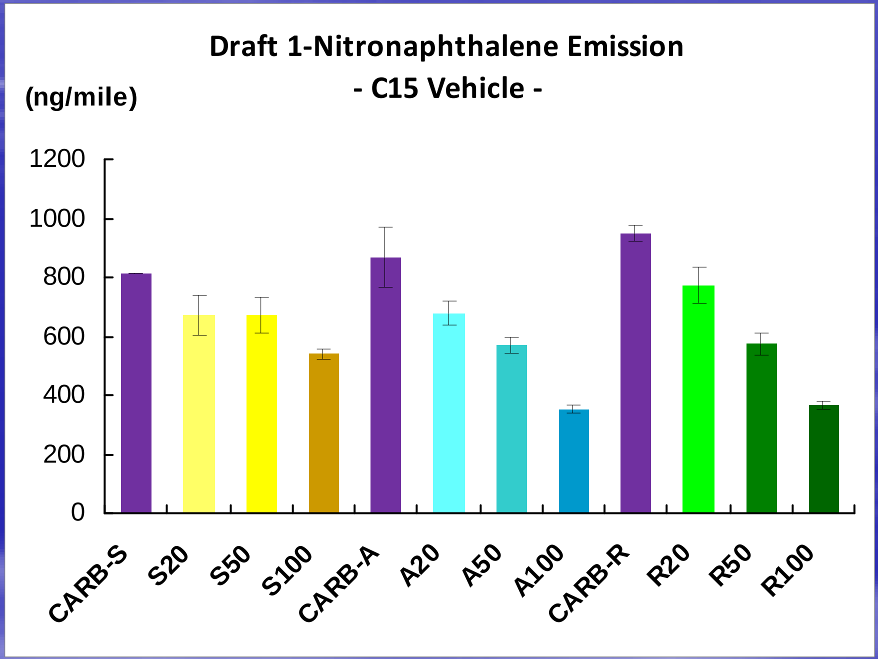
PAH Emissions
Vapor-Phase PAHs



PAH Emissions
Nitro-PAHs
PM Associated

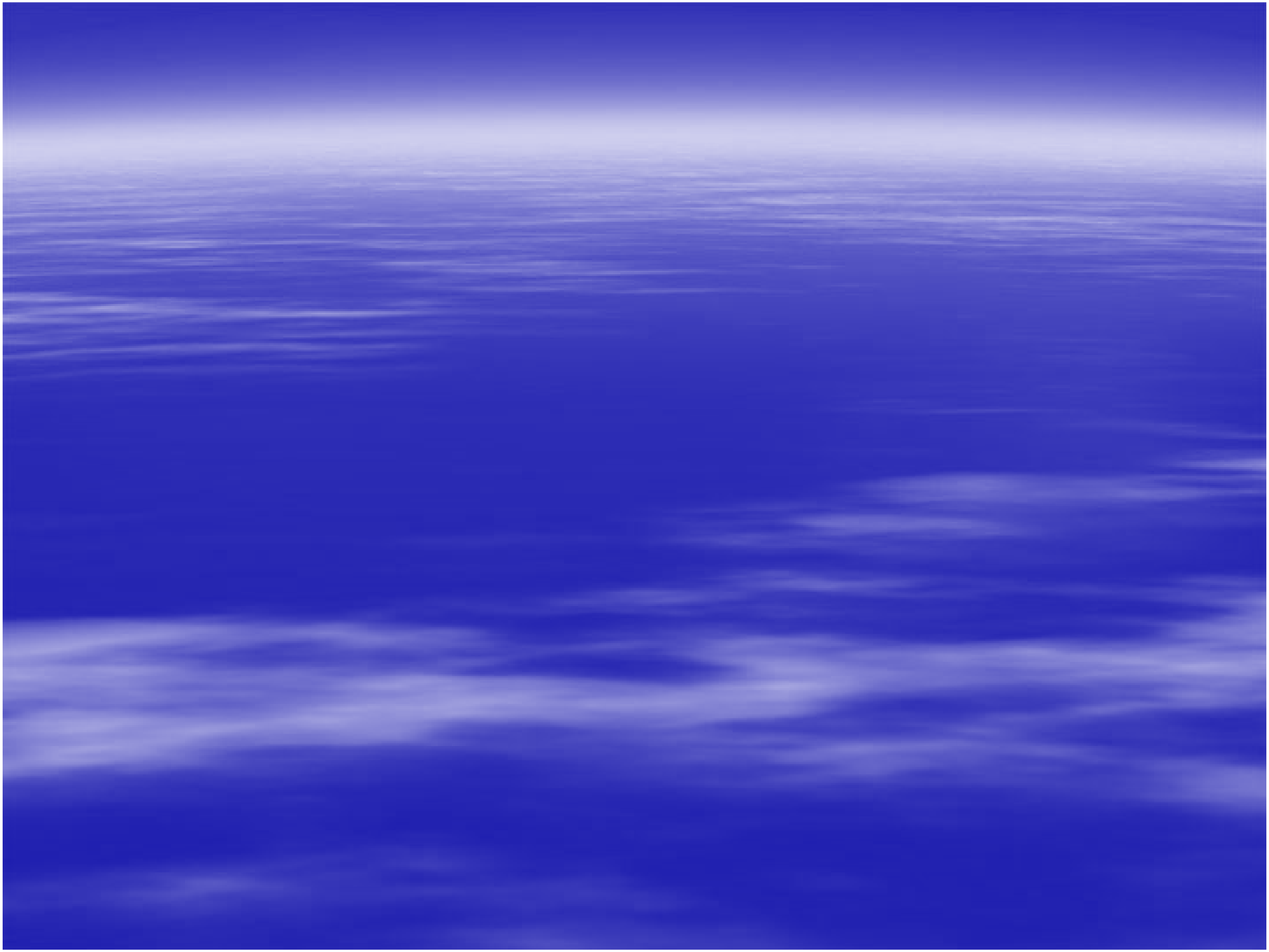


PAH Emissions
Nitro-PAHs
Vapor-Phase

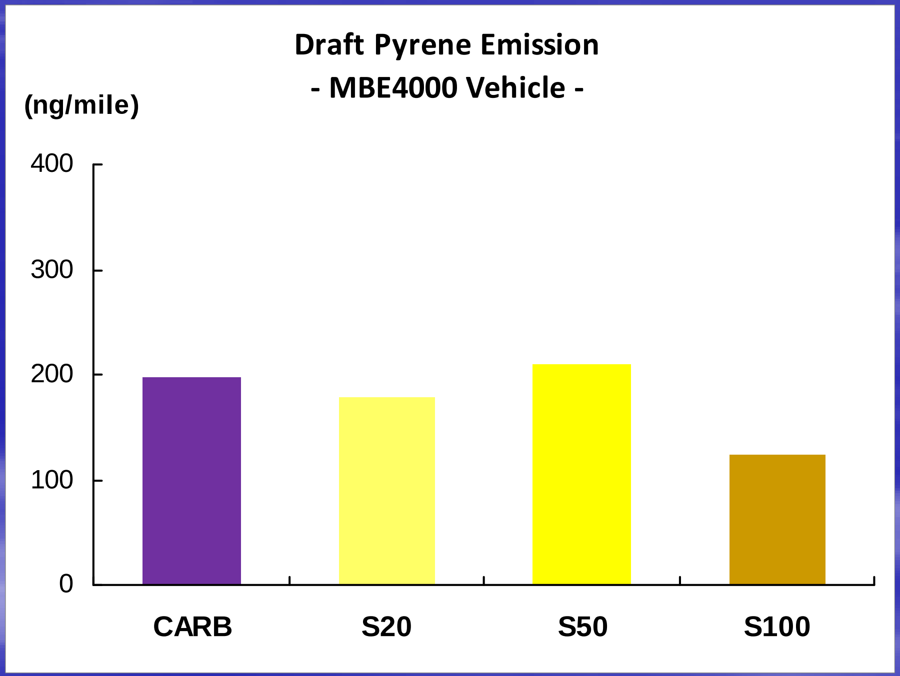


Summary PAHs
C15 Vehicle

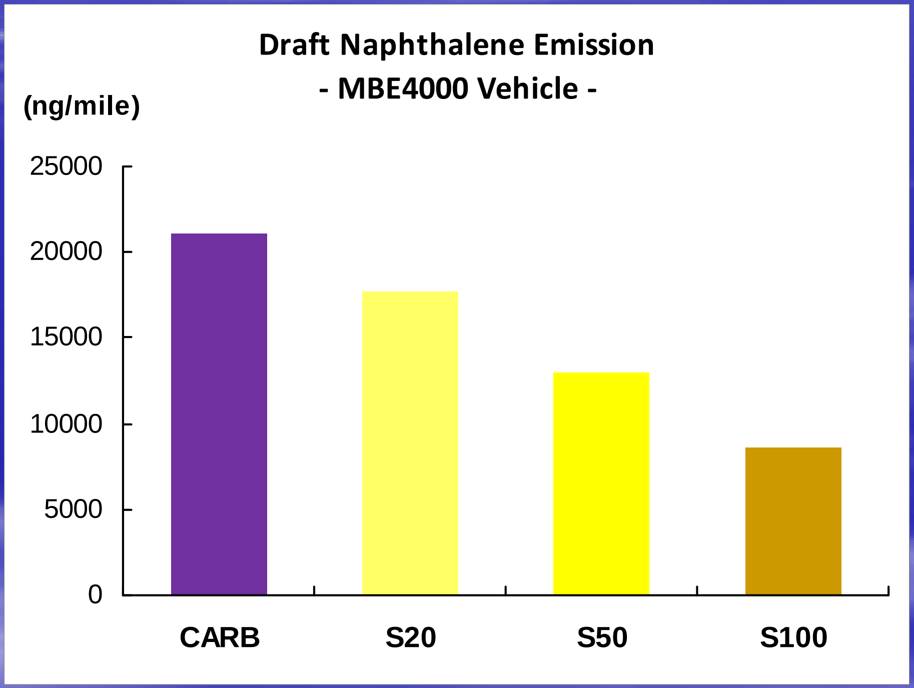
- PM and Semi-Volatile PAHs and Nitro-PAHs decreased with increasing blend level of biodiesels.



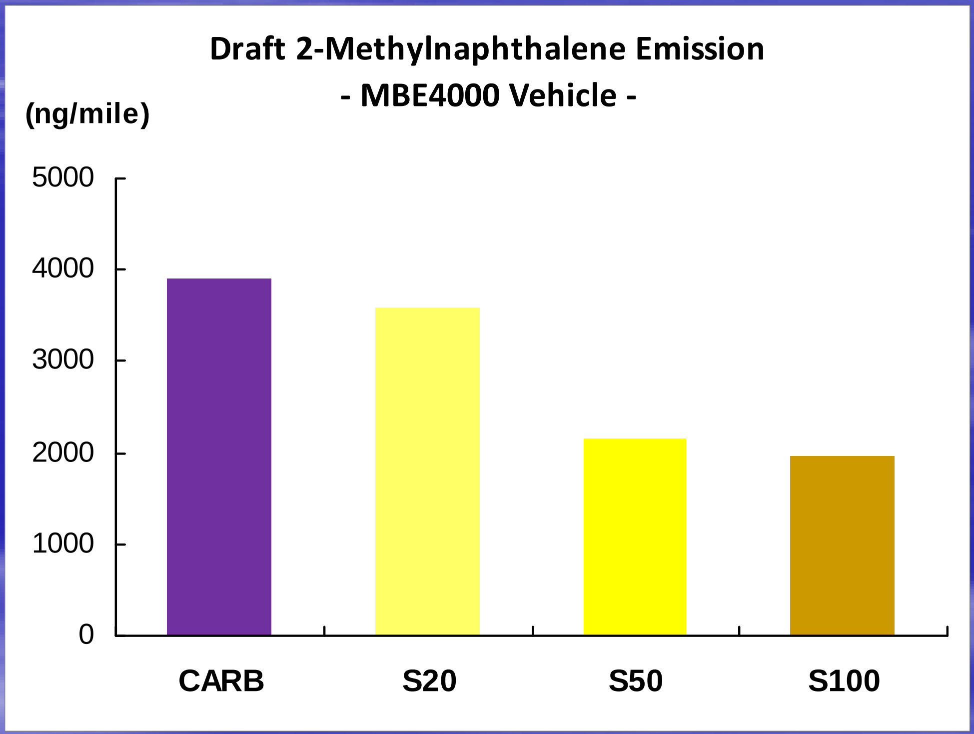
PAH Emissions
PM Associated PAHs
MBE 4000



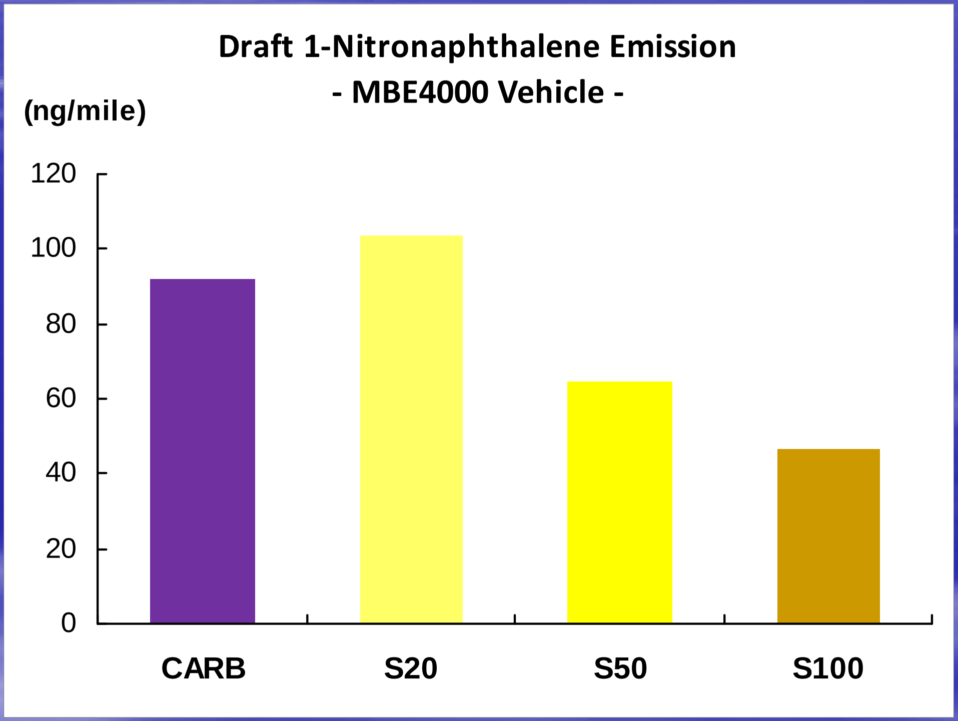
PAH Emissions
Vapor-Phase PAHs
MBE 4000



PAH Emissions
Vapor-Phase PAHs
MBE 4000



Draft PAH Emissions
Nitro-PAHs
MBE 4000



Summary PAHs
MBE 4000 Vehicle

- Low Levels of PM associated PAHs and Nitro-PAHs in CARB and Biodiesel Fuel Emissions
- Lower levels of Vapor-phase PAHs and Nitro-PAHs emissions

Reactive Carbonyls

Reactive Carbonyl Sampling

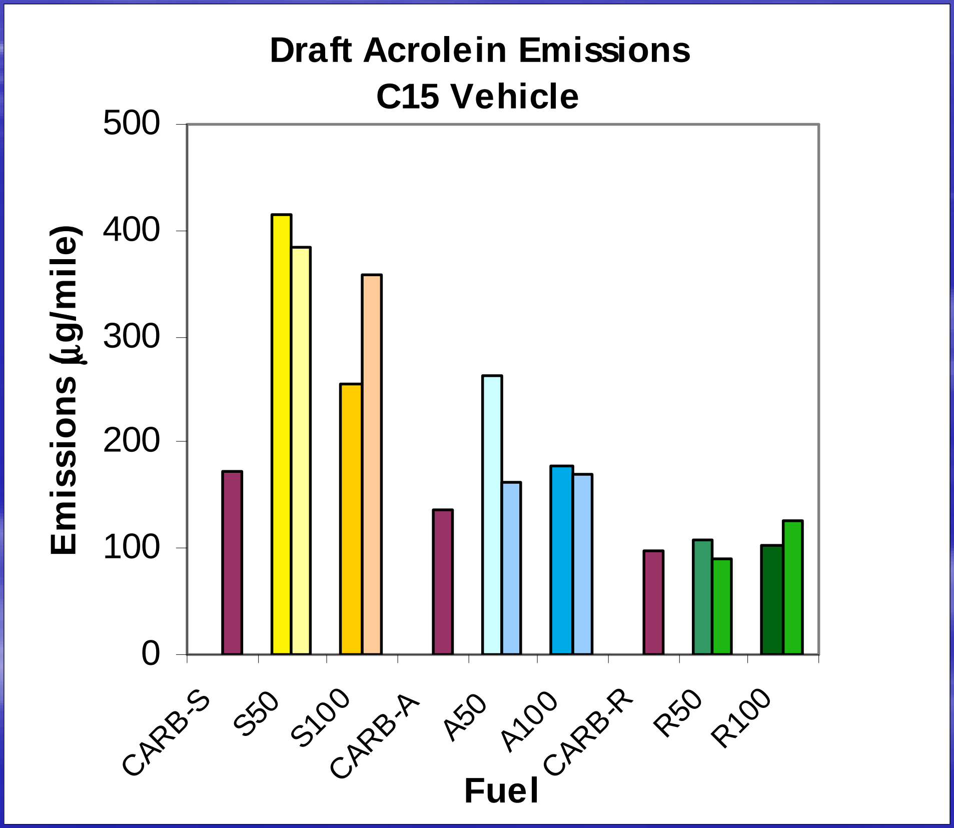
- Sampling from Dilution Tunnel to Mist Chamber
- Samples in parallel to Filtered/Charcoal dilution air
- Samples for single UDDS test cycle

Reactive Carbonyl Analyses

- Stable carbonyls formed through reaction with bisulfite
- Carbonyls liberated from bisulfite
- Free carbonyls derivatized by o-(2,3,4,5,6-pentafluorobenzyl)hydroxylamine (PFBHA*)
- Derivatives detected & quantitated by GC/MS - NCI

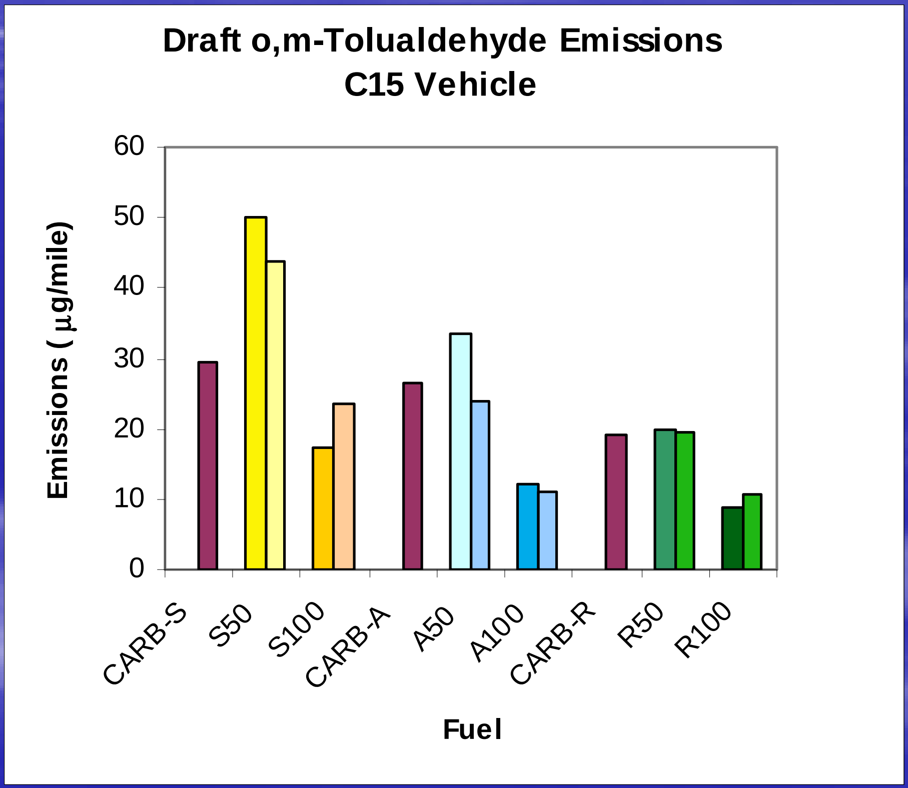
Carbonyl Emissions

C15 Vehicle



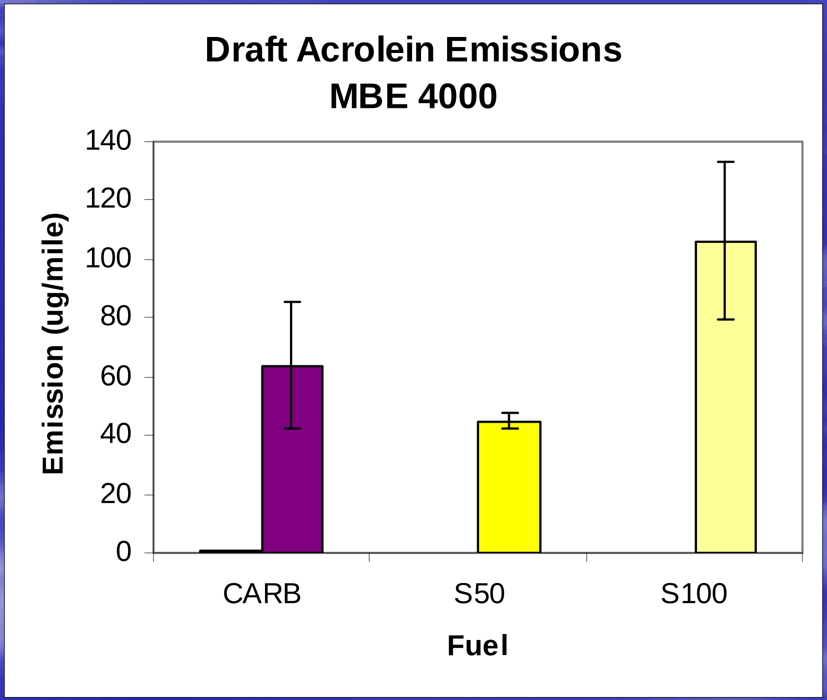
Carbonyl Emissions

C15 Vehicle



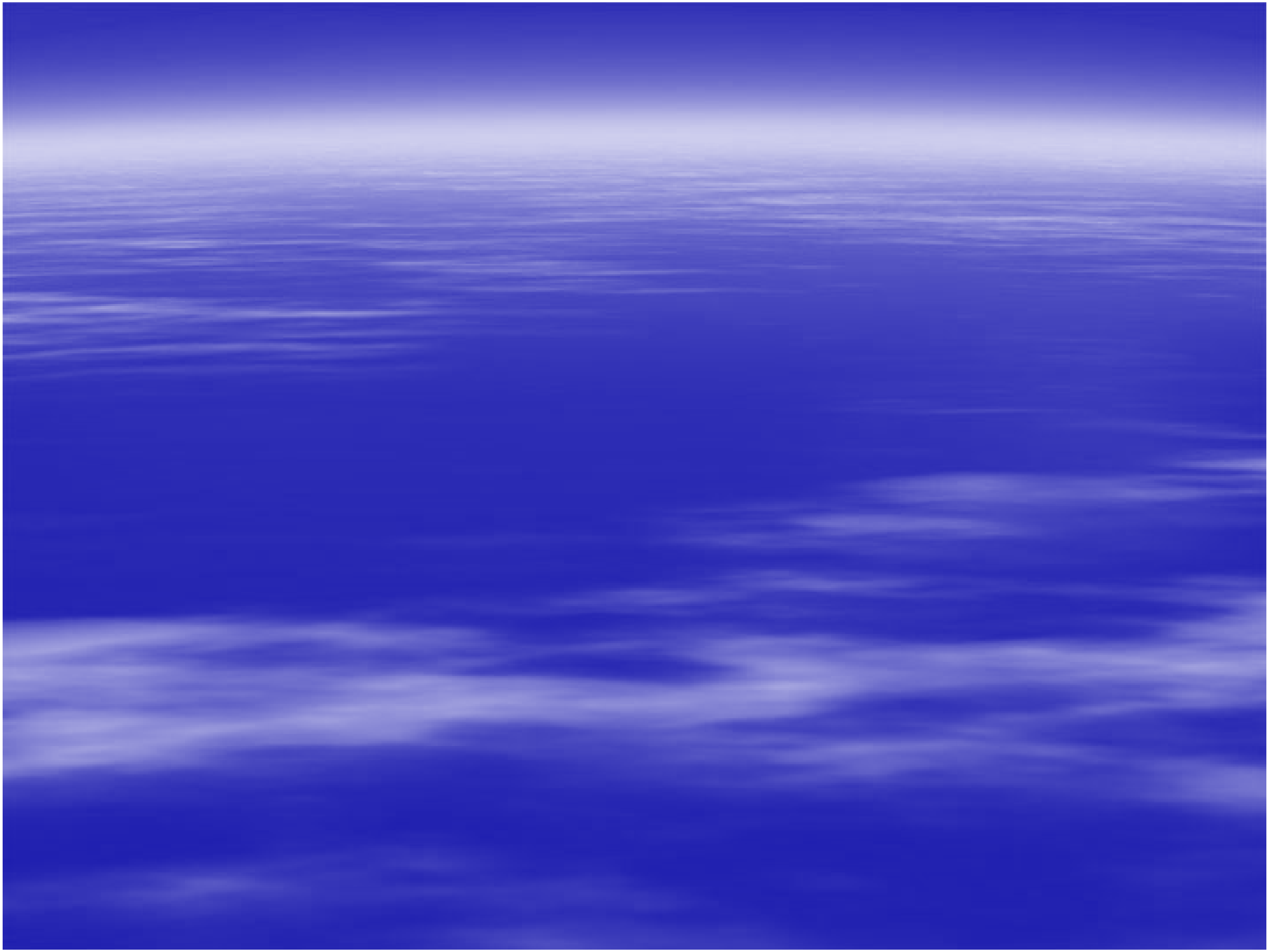
Carbonyl Emissions

MBE 4000 Vehicle



Summary
Reactive Carbonyls

- C15 Vehicle S50, S100 and A50, A100 were higher in certain carbonyls such as acrolein
- C15 Vehicle Renewable diesel no change over Carb
- MBE4000 Vehicle – carbonyls lower



Genotoxicity Tests

- Microbial
  eg. Ames Salmonella test
- Mammalian cell
  eg. Chinese hamster ovary (CHO)
- *In vivo*
  eg. Big Blue transgenic rodent

Genotoxicity Tests
Two Questions

- How consistent is it to hypothesized mechanisms of action for carcinogens?

- How does it compare to animal or human carcinogenicity tests?

Salmonella Tester Strains

- TA 98
- Frameshift mutation in the HisD gene coding for histidinol dehydrogenase
- Target site: series of 8 GCGCGCGC's

Salmonella Tester Strains

- TA 100

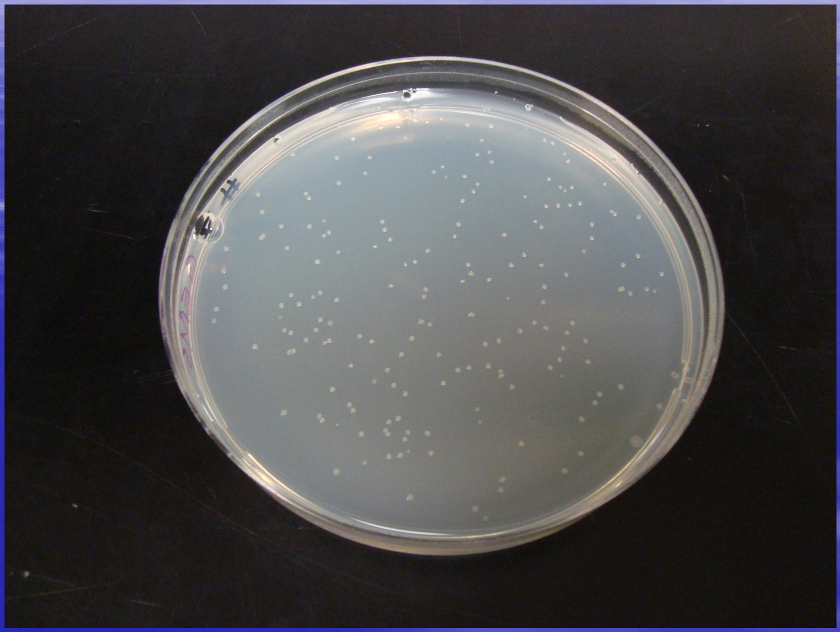
- Base-pair mutation in the His G gene coding for His biosynthesis
- Target site: GGG (proline) His dependent

Salmonella/microsome Test

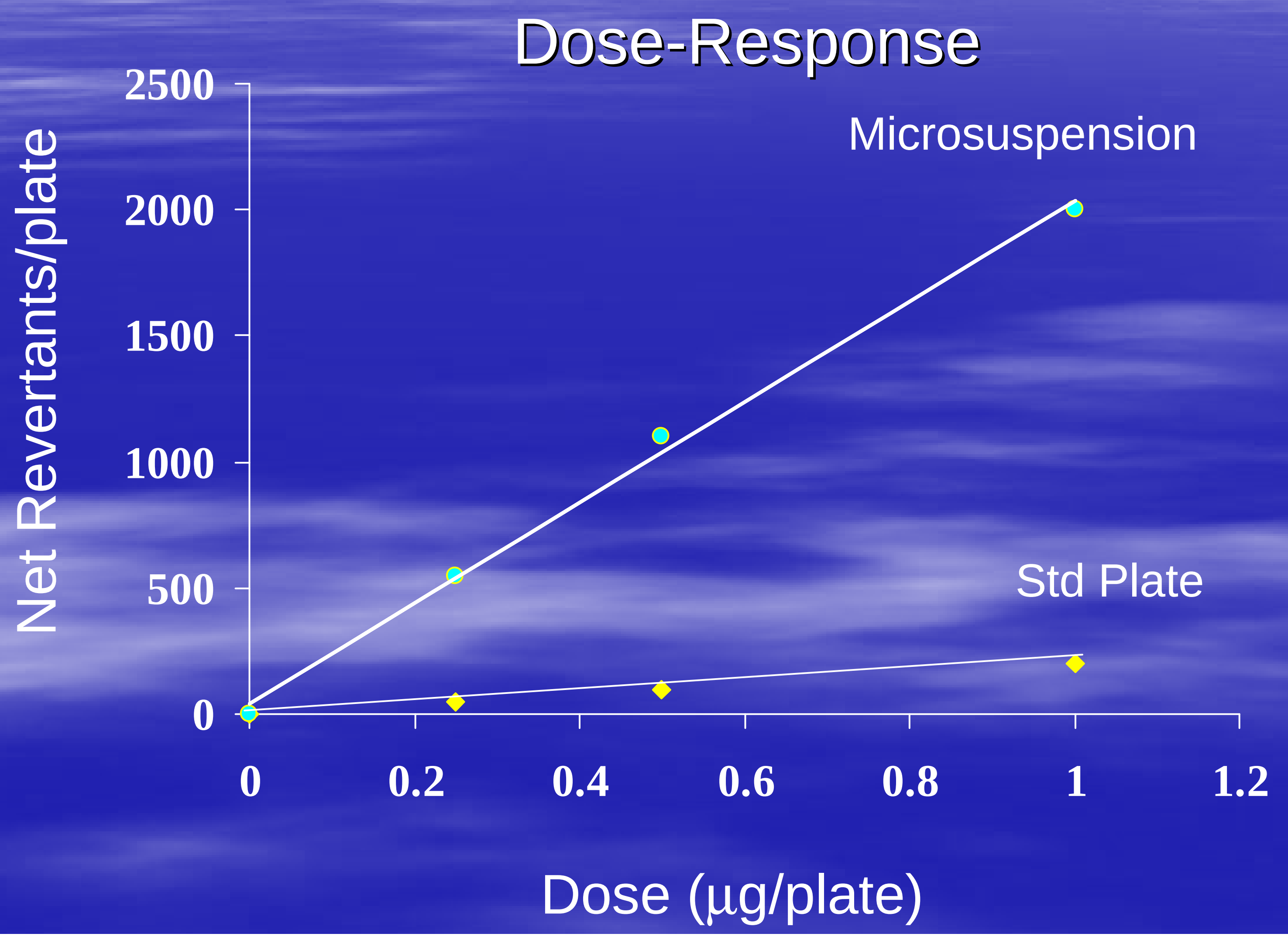
- A feature of the Test:
- Metabolic enzymes can be added to detect activation

Salmonella/microsome Test

- Metabolic enzymes needed for activation of certain compounds – eg. PAHs
- Enzymes from various tissues can be used – e.g. Lung, liver



Microsuspension



Urine Mutagenicity

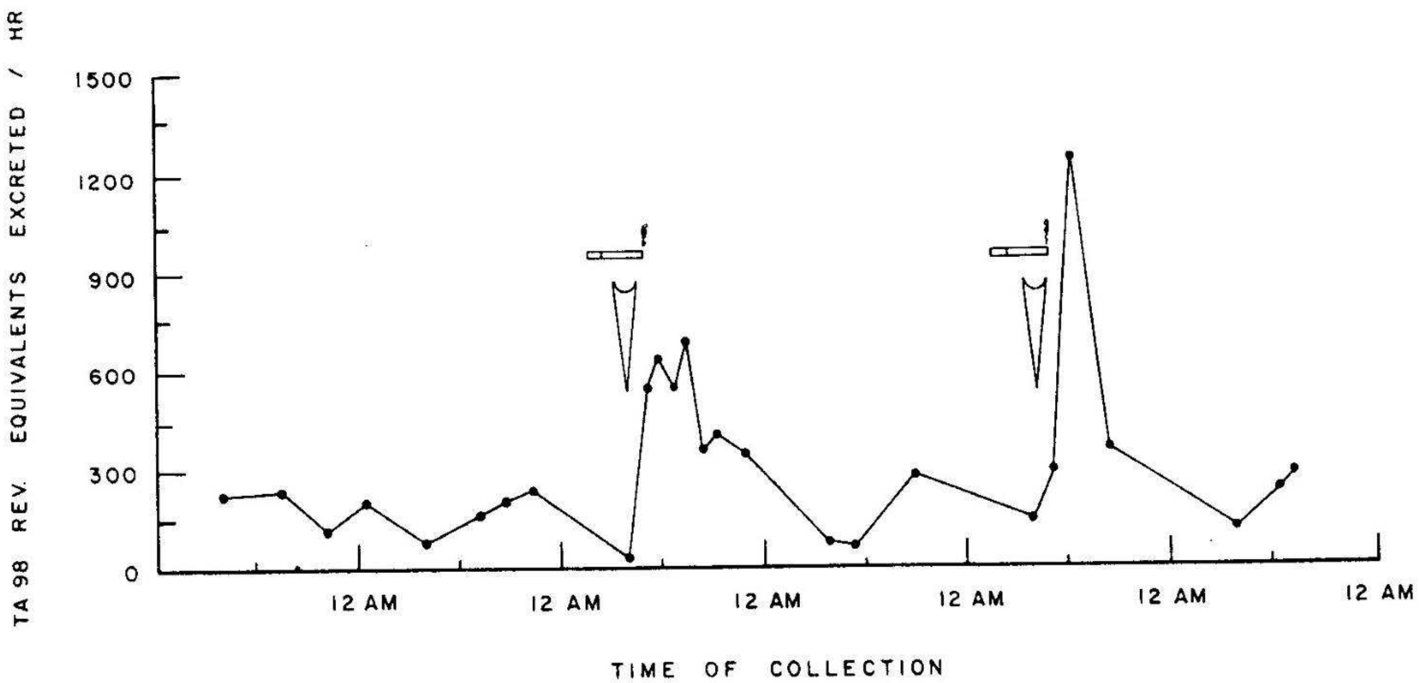
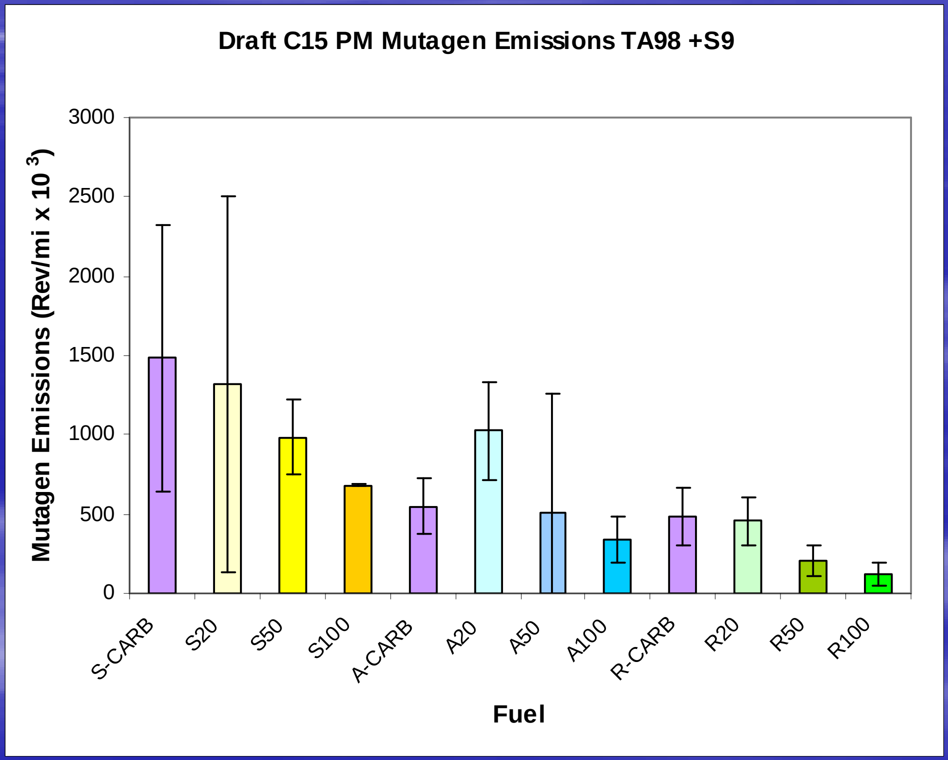


Fig. 1. The kinetics of mutagen excretion in the urine of an occasional smoker (smokes about 1 cigarette per week). A single filter-tipped cigarette (17 mg tar per cigarette) was smoked at the times indicated. TA98 revertant equivalents/h = TA98 revertants per ml urine multiplied by the total volume of urine donated and divided by the number of hours elapsed since providing the last specimen. TA98 revertants per ml of urine was determined from the slope of the linear portion of the dose–response curve for each urine specimen. Water extracted in an identical manner as urine was used as the control.

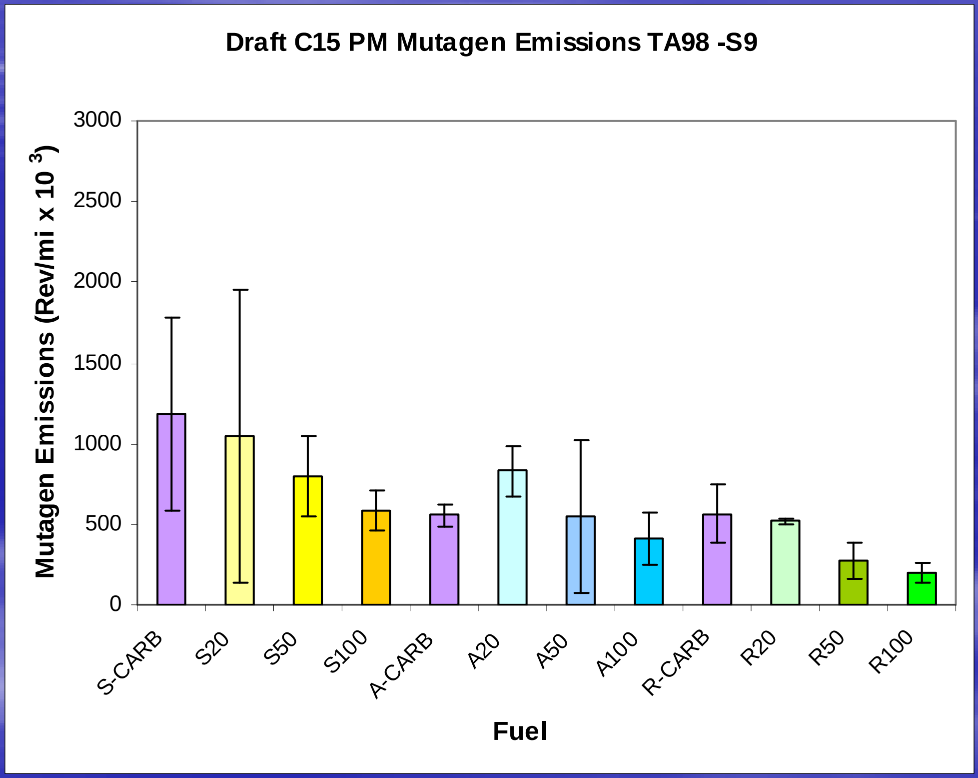
Mut Res 157:227, 1985

Mutagen Emissions TA98 (+S9)
C15 Vehicle PM

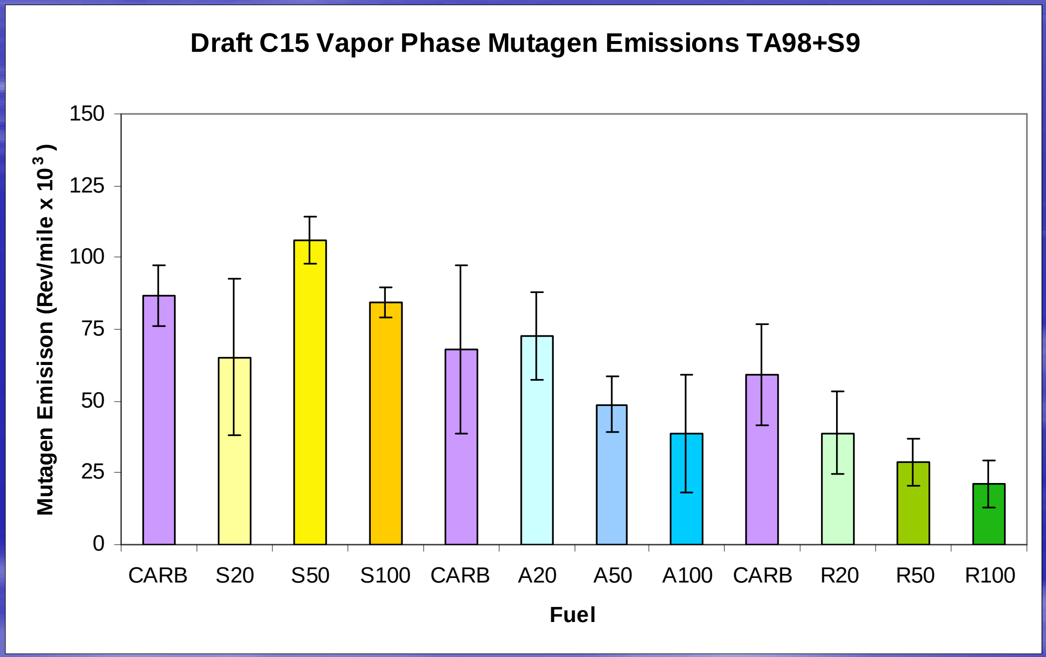


Mutagen Emissions TA98 (–S9)

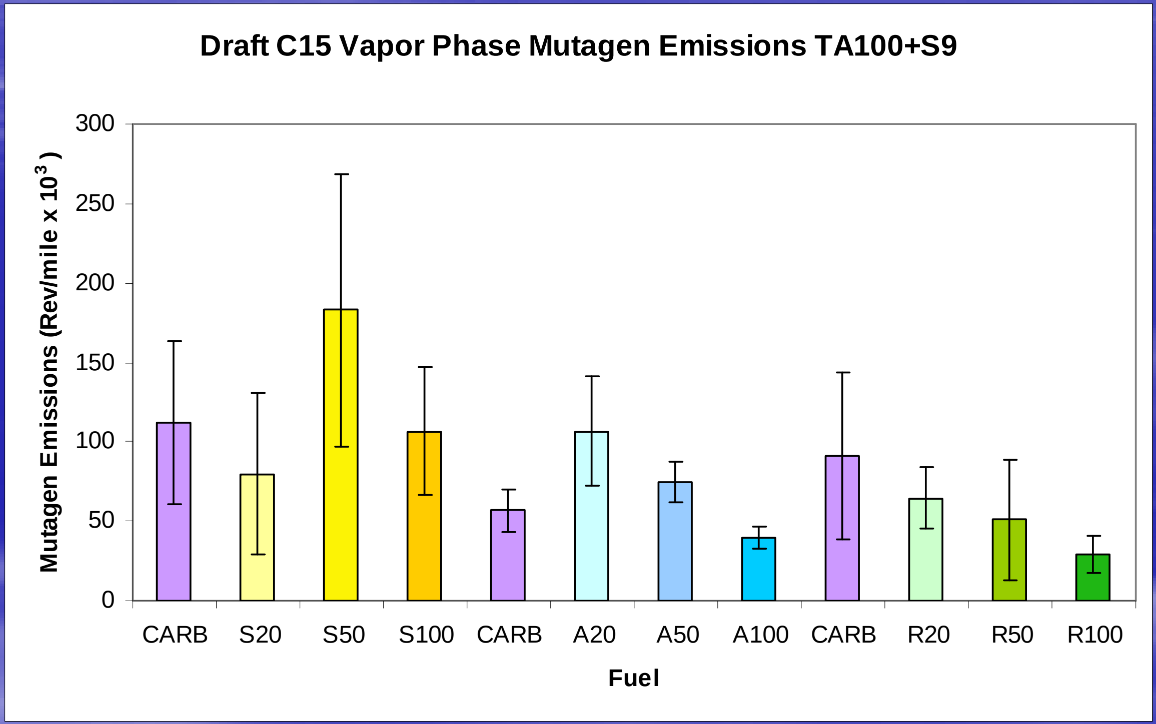
C15 Vehicle PM



Mutagen Emissions TA98 (+S9) C15 Vehicle VP

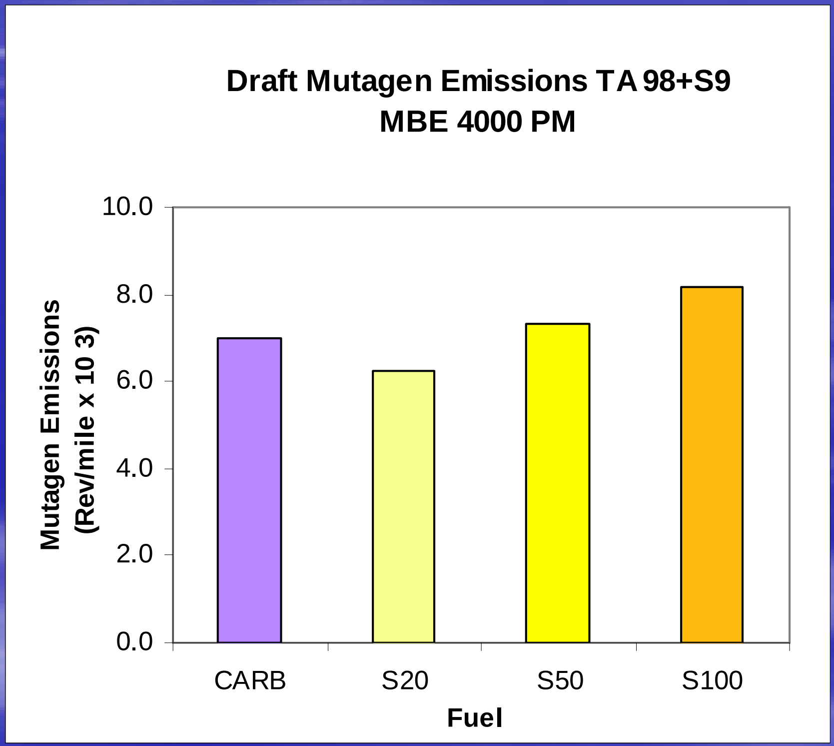


Mutagen Emissions TA100 (+S9)
C15 Vehicle VP



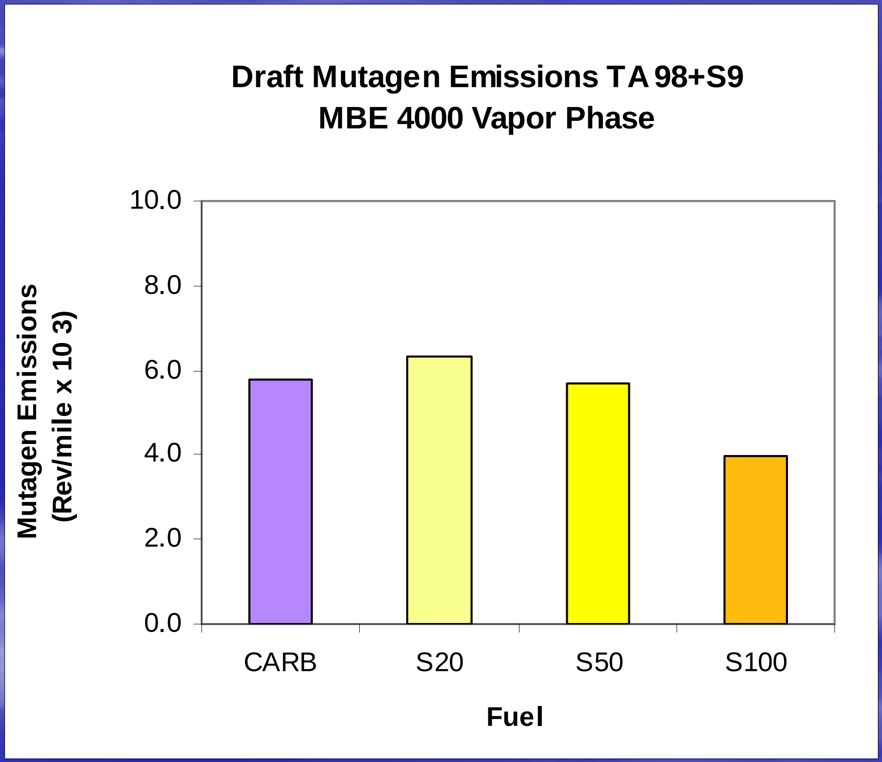
Mutagen Emissions

MBE 4000 Vehicle PM

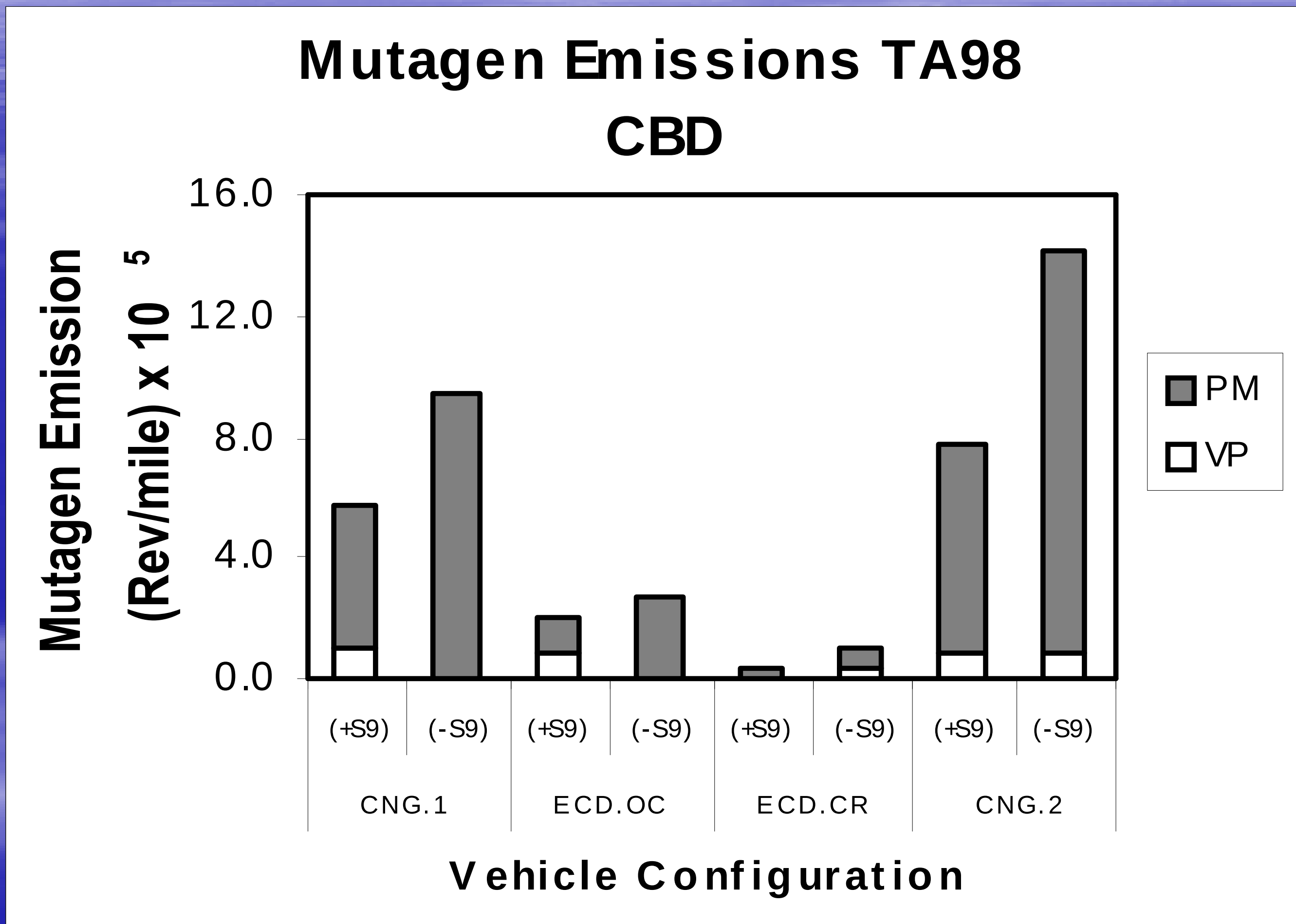


Mutagen Emissions

MBE 4000 Vehicle VP



Bus Emissions
Compressed Natural Gas



ES&T 39:7638, 2005

Summary

- For C15 Vehicle: Generally decrease in Mutagen emissions with blend level
- For C15 PM Samples TA98 (+ or – S9) more sensitive than TA100 for all fuels
- Vapor Phase samples lower mutagen emissions than PM TA100 slightly more sensitive
- MBE Mutagen Emissions considerably lower than C15

Summary

- Chemical and Biological tests were overall very consistent with each other regarding emission results for the fuels tested

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UCD – Etox

NESTE, SCAQMD, ARB

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