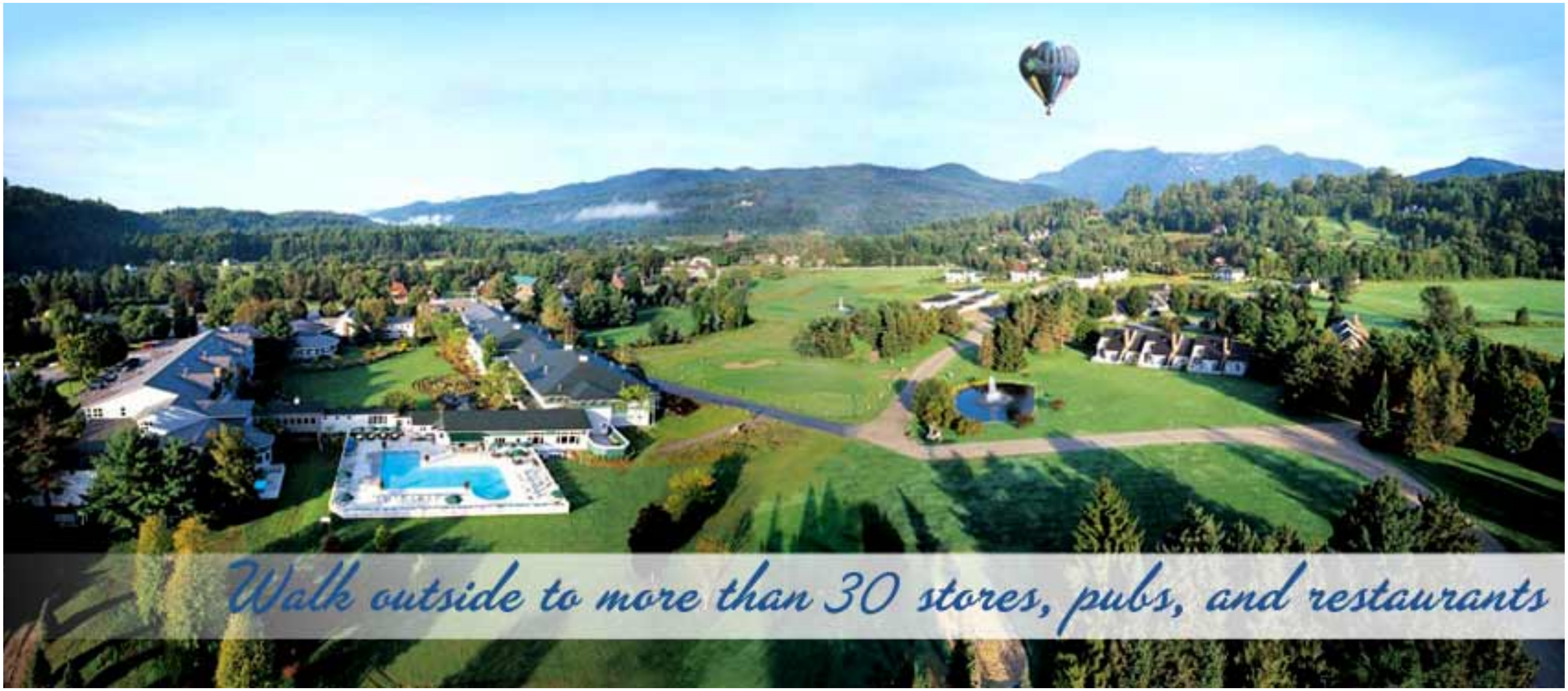

2013 Environmental Nanotechnology Gordon Research Conference

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Detection of carbon nanotubes in environmental and biological matrices using programmed thermal analysis

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Contributors

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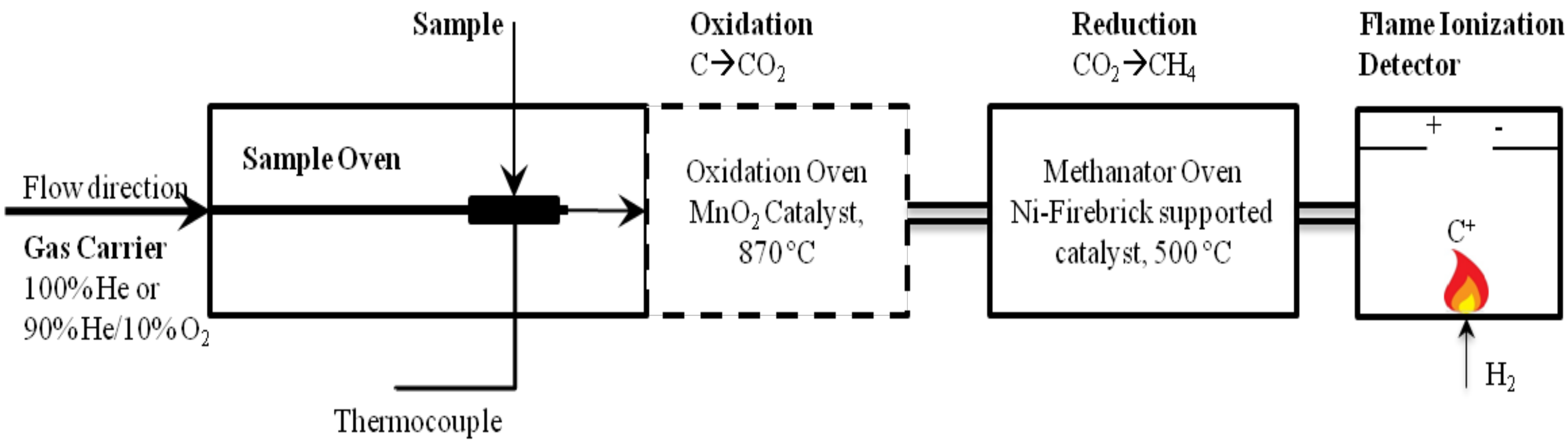
Carbon Nanotubes

- Optimization of analysis scheme
  - Many techniques exist
  - Here we optimize a thermal analysis method
- Objective is to remove organic carbon and "soot" signals in thermal analysis method
- Application to CNT analysis in rat lungs
  - Objective is to demonstrate spike recoveries in real lung tissue
- Future work – apply extraction and analysis in eco-toxicity testing and CNT release from products (e.g., thermal packaging)

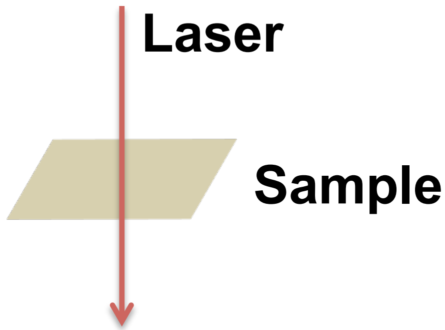
Elemental Carbon/Organic Carbon Analyzer



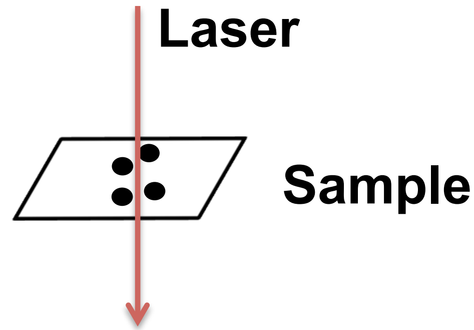
Sunset Laboratories Lab OC-EC Aerosol Analyzer

Manual OC-EC Cuttoff Required

Homogeneously loaded sample (e.g., air)



Heterogeneously loaded sample (e.g., CNTs)



Reasons using the laser transmission to separate OC from EC automatically for CNTs does not work:

- As oxidation occurs the CNTs are removed and the laser may or may not still be hitting the "sample."
  - This is even more pronounced when the CNTs are embedded in a matrix such as lung tissue.
- CNTs are generally too black for the laser to pass through, which results in a poor baseline for transmission readings.
- Typically, automatic correction will mistaken CNTs as OC or OC as EC.

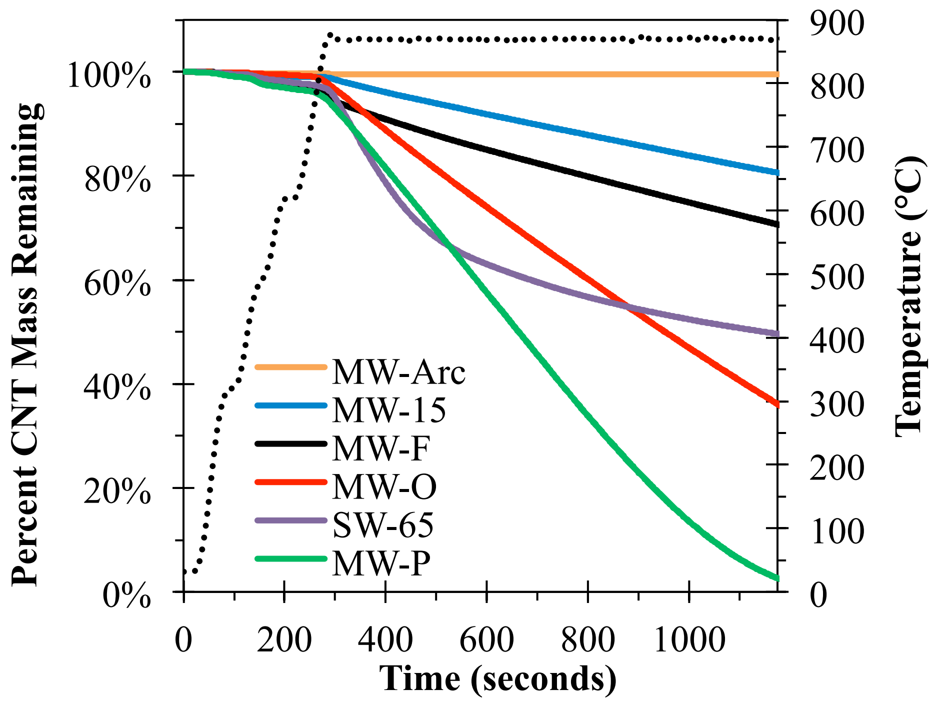
Not All CNTs are Equivalent – So we examined 15 different CNTs

Table 1. Properties of CNTs Used in This Study

CNT ID	CNT type	state	purity[a]	metal content[b]	outer diameter (nm)	inner diameter (nm)	length (μm)
MW-O	MWCNT	raw	>95%	<6%	20–30	5–10	10–30
MW-P	MWCNT	purified	>98%	<2%	20–30	5–10	10–30
MW-F	MWCNT	functionlized	>99.9%	<0.01%	20–30	5–10	10–30
MW-15	MWCNT	raw	>95%	<5%	7–15	3–16	0.5–200
MW-20	MWCNT	raw	>95%	<5%	10–20	5–10	0.5–200
MW-30	MWCNT	raw	>95%	<5%	10–30	5–10	0.5–500
MW-100	MWCNT	raw	>95%	<5%	60–100	5–10	0.5–500
MW-OH	MWCNT	functionalized	>95%	<1.5%	8–15	3–5	10–50
MW-COOH	MWCNT	fucntionalized	>95%	<1.5%	8–15	3–5	10–50
MW15G[c]	MWCNT	annealed	>97%	<1%	7–15	3–6	0.5–100
MW-Mitsui	MWCNT	raw	>98%	<1%	20–70	NA	NA
MW-arc	MWCNT[d]	raw	<50%	0%	5–10[e]	NA	NA
SW	SWCNT	raw	<50%	<10%	1.1	NA	0.5–100
SW-65	SWCNT	purified	<75%	<10%	0.8	NA	0.45–2

[a]CNT content reported by manufacturer. MW-P and MW-F calculated assuming no amorphous carbon remaining. [b]Metal content reported by manufacturer except for MW-F and MW-P determined using energy-dispersive X-ray spectroscopy and MW-15G using thermogravimetric analysis. [c]MW-15 annealed at ~2000 °C in UHP He. [d]Synthesized using arc method; all others are CVD. [e]Obtained from TEM images; all others reported by manufacturer.

Thermal Properties of CNTs

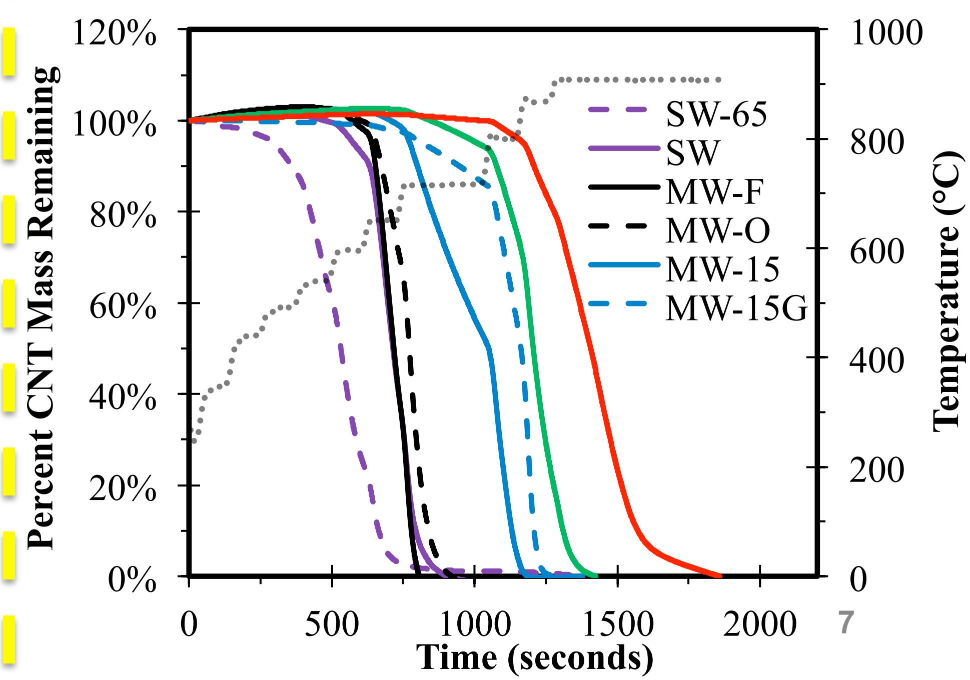


Inert conditions (100% He)

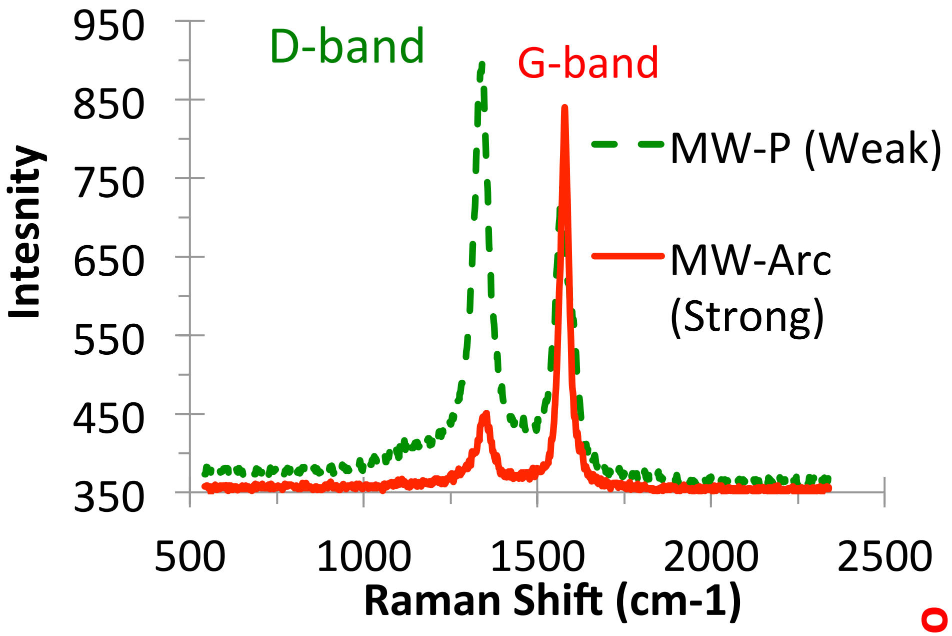
Conclusion: Surface oxygen groups allow some CNTs to burn in inert gas environment

Oxidizing conditions (90% He/ 10% O_2)

Conclusion: Not all CNTs "burn" at the same temperature

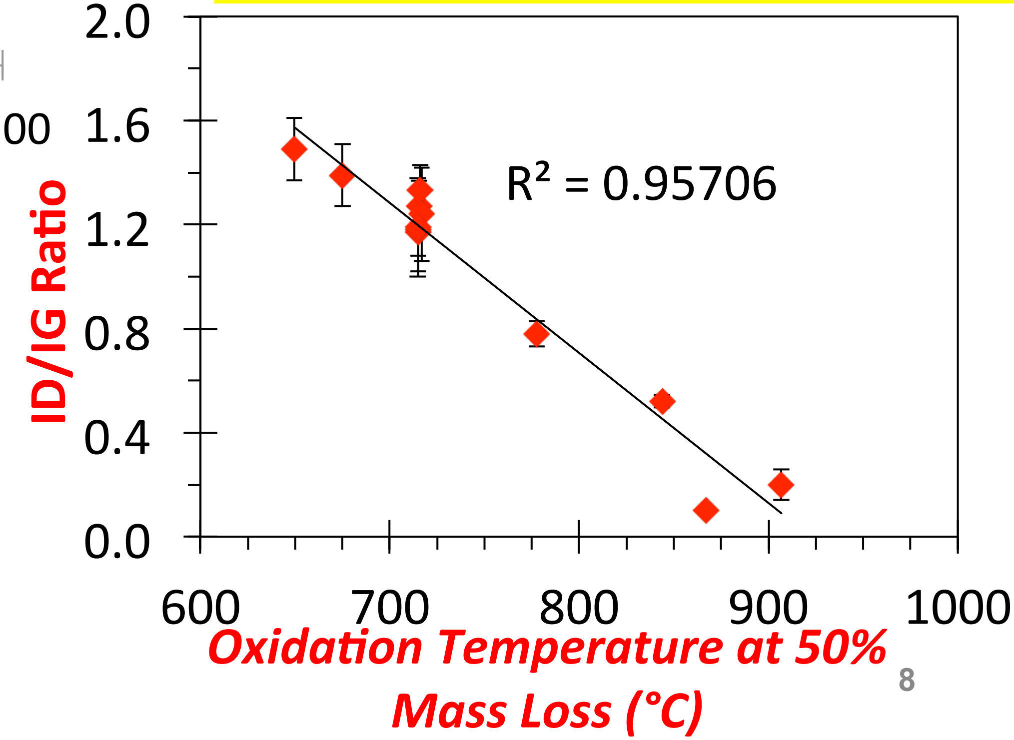


Oxidation "Capability" Can be Predicted from Raman Spectra

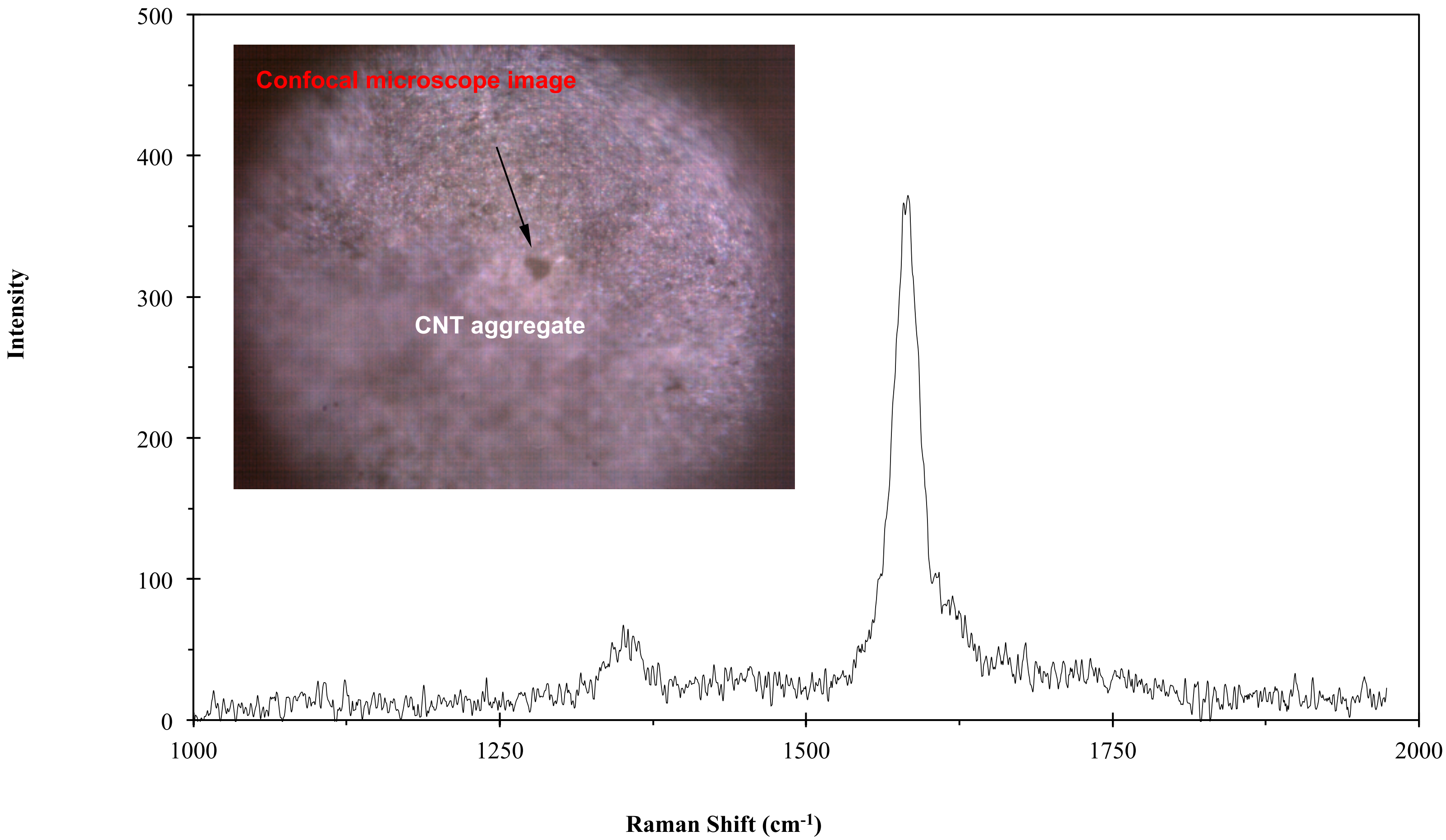


Raman spectra of two CNTs

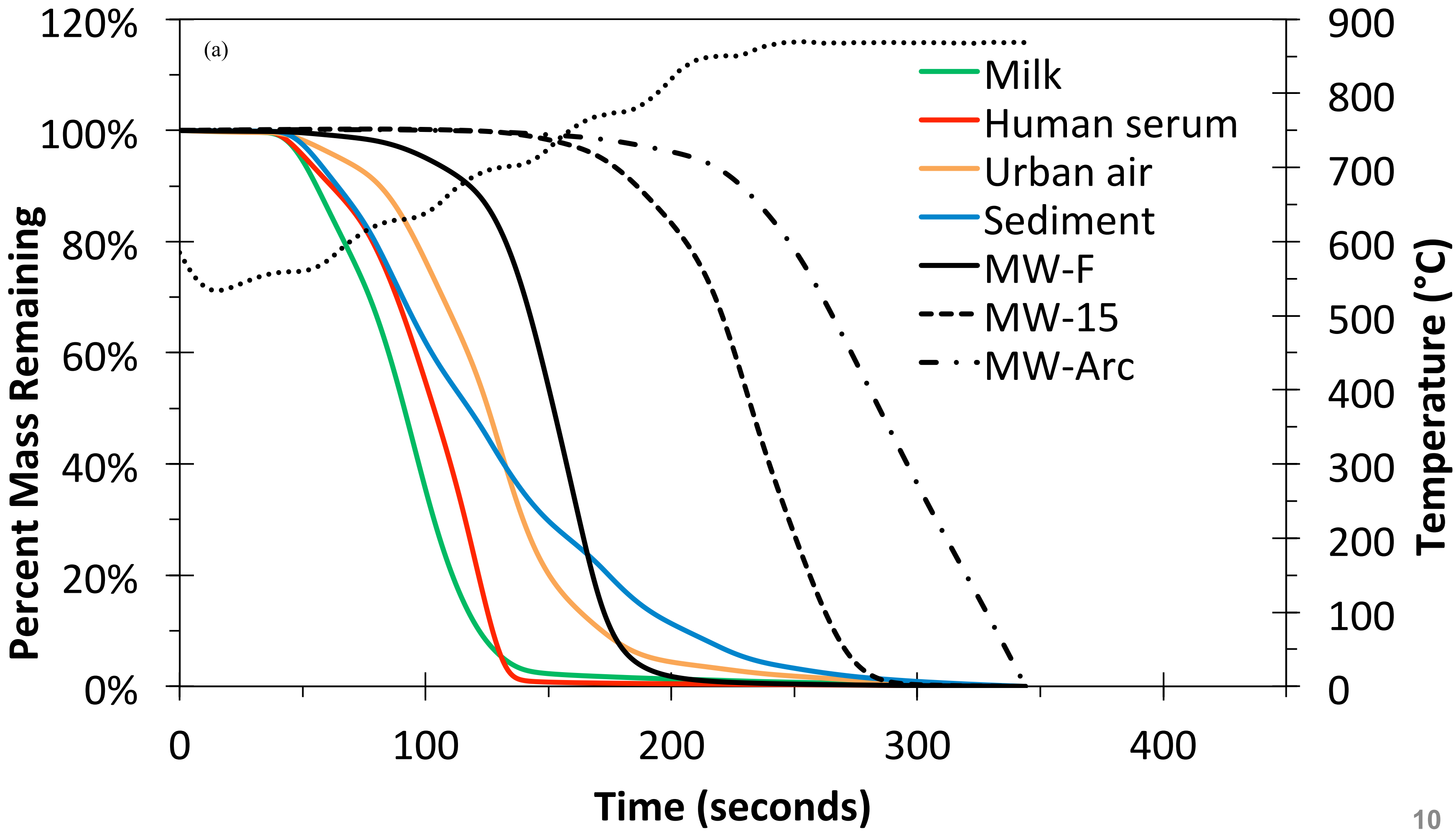
Correlation between oxidation temperature at 50% mass loss and the I_D/I_G ratio



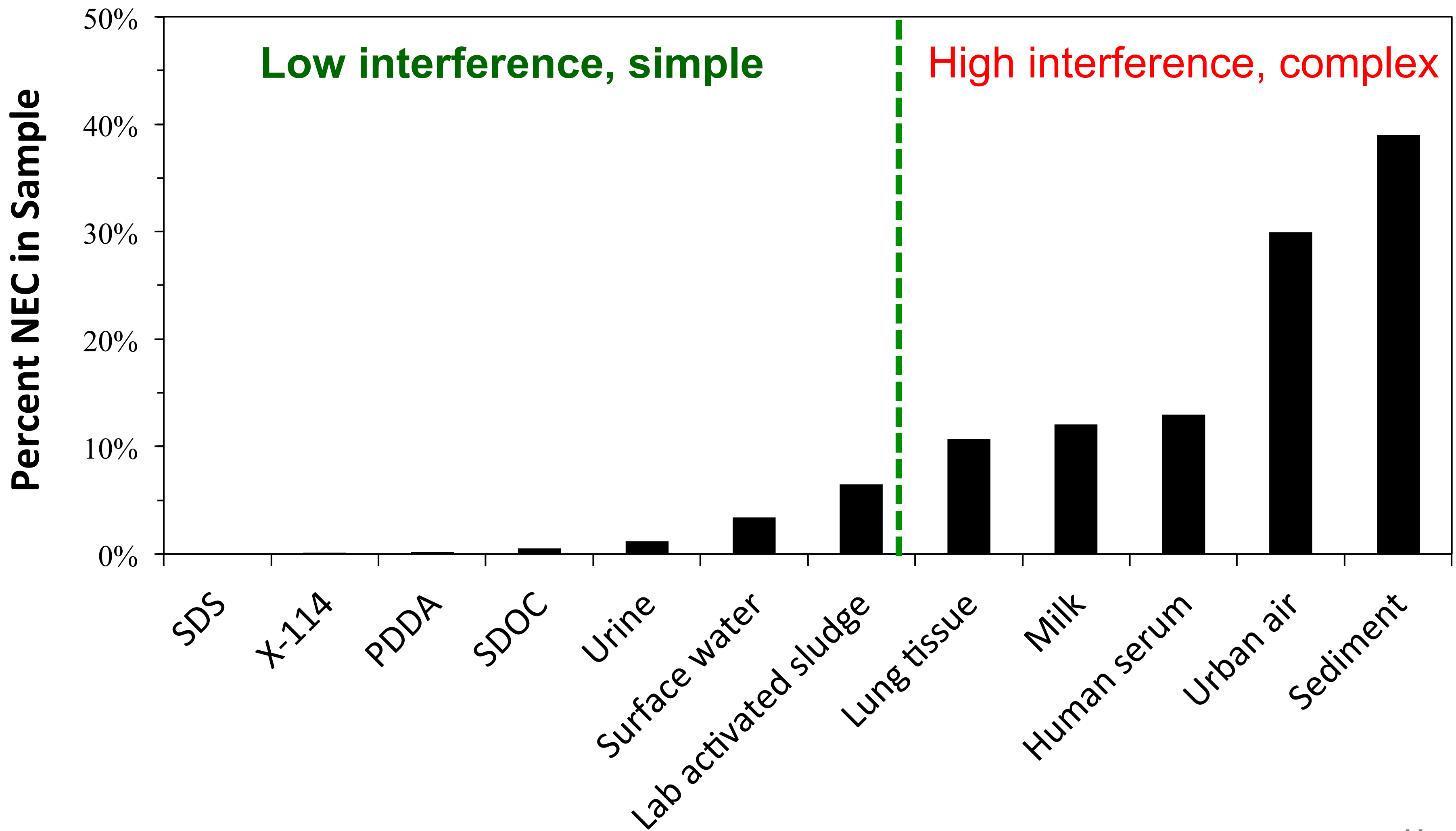
Raman spectrum of CNTs extracted from Cyanobacteria



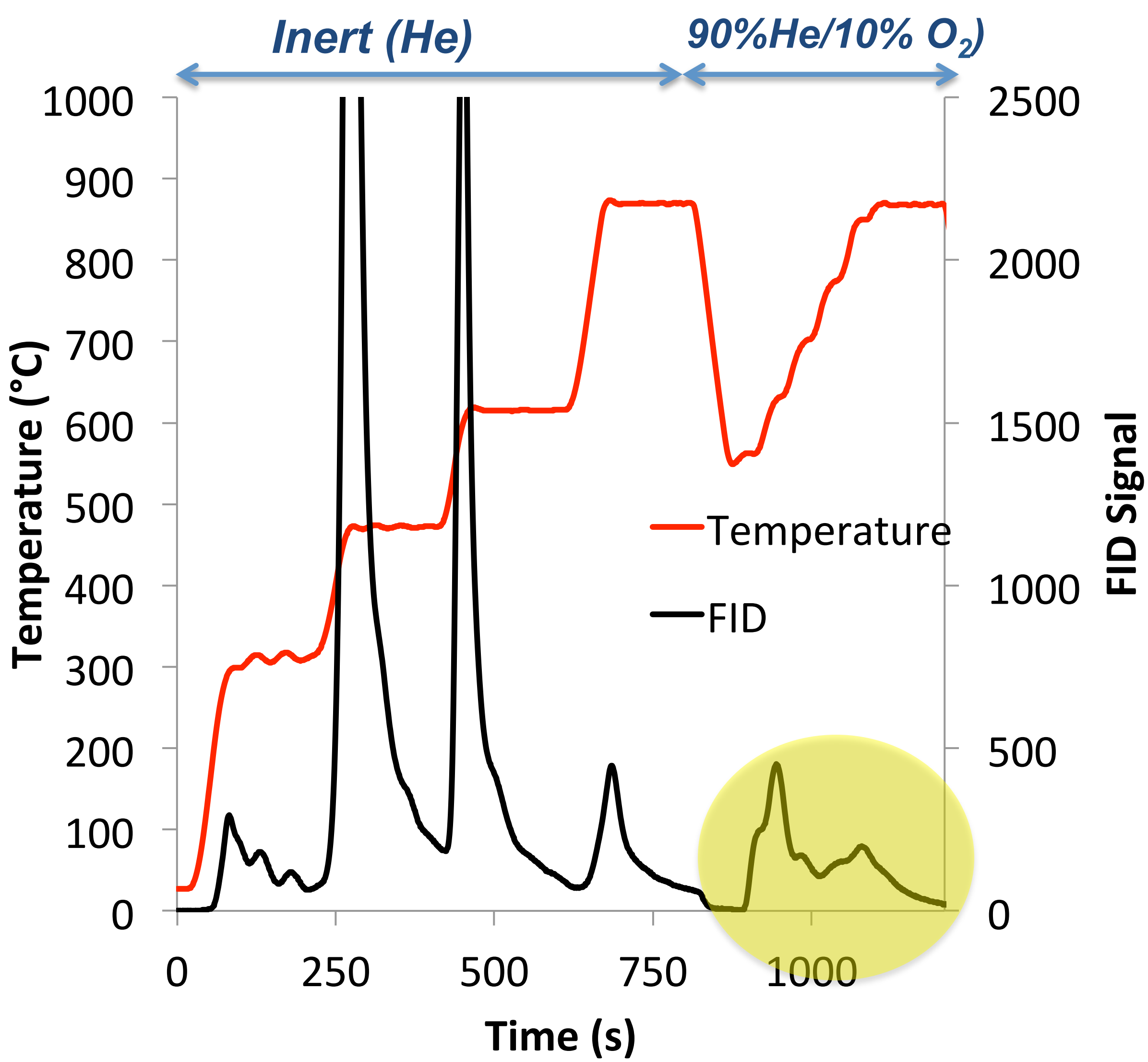
Organic materials produce non-elemental carbon (NEC) that causes interference where CNTs respond



Thermal Behavior of Various Environmental and Biological Matrices



Tissue Sample Example: Lung Tissue



Even a small amount of lung tissue (a few mg) cause significant charring (EC) response

Challenge with Thermograms

Organic material in tissue and environmental samples "char" and produce *responses* which could be interpreted as CNTs

Critical: Determine temperature dependency of various biological and environmental matrices to produce non-elemental carbon (NEC) as a function of the total carbon in the sample

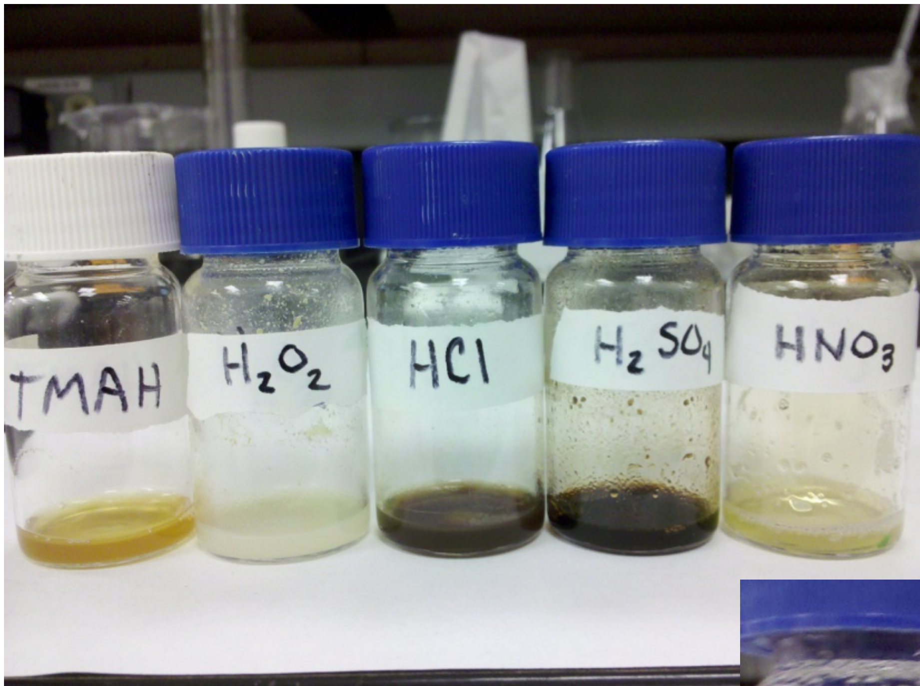
Challenge: How to digest away organic matrix without affecting CNT properties

Approach

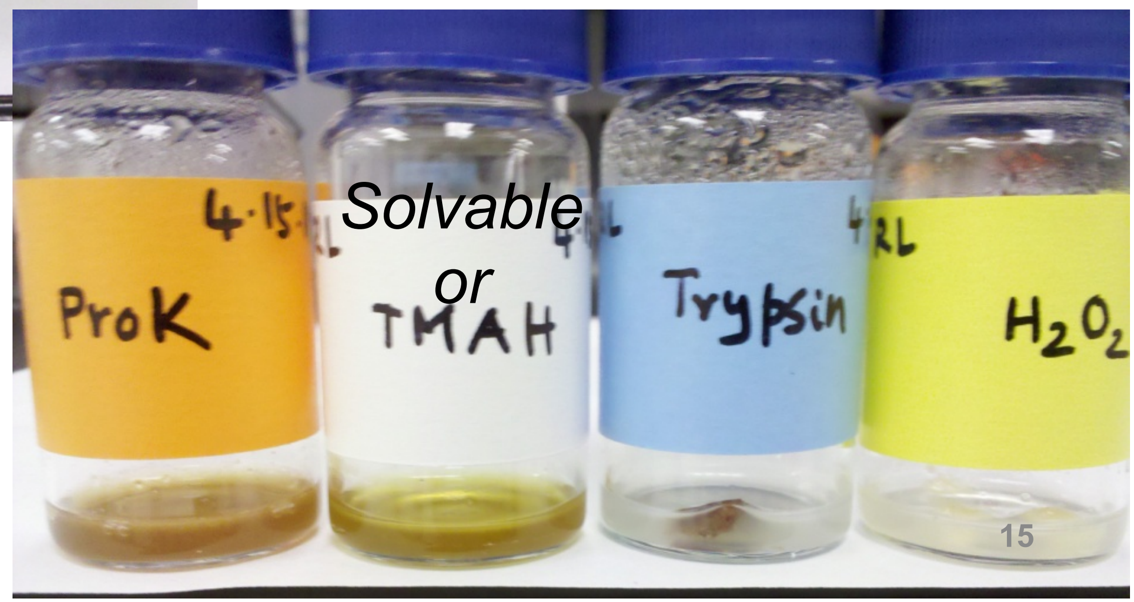
We investigated a variety of digestates using rat lung tissue

Experiments used Mitsui CNTs (CVD method) (Strong CNT classification)

Comparison of Different Chemical Digestion Methods



Color of rat lungs after chemical digestion



Black color results in highest NEC response

CNT Extraction from Rat Lung Tissue

Matrix	Acids		Acid/Oxidants		Oxidants[b]		Hydrolysis		Enzymatic
	HCl	HF	**HNO_3[a]**	H_2SO_4	H_2O_2	NaOCl	**TMAH[c]**	**Solvable[d]**	**Pro K[e]**
Rat lung			✓	✓	✓	✓	✓	✓	✓
CNT	✓	✓	✓				✓	✓	✓

Check mark:

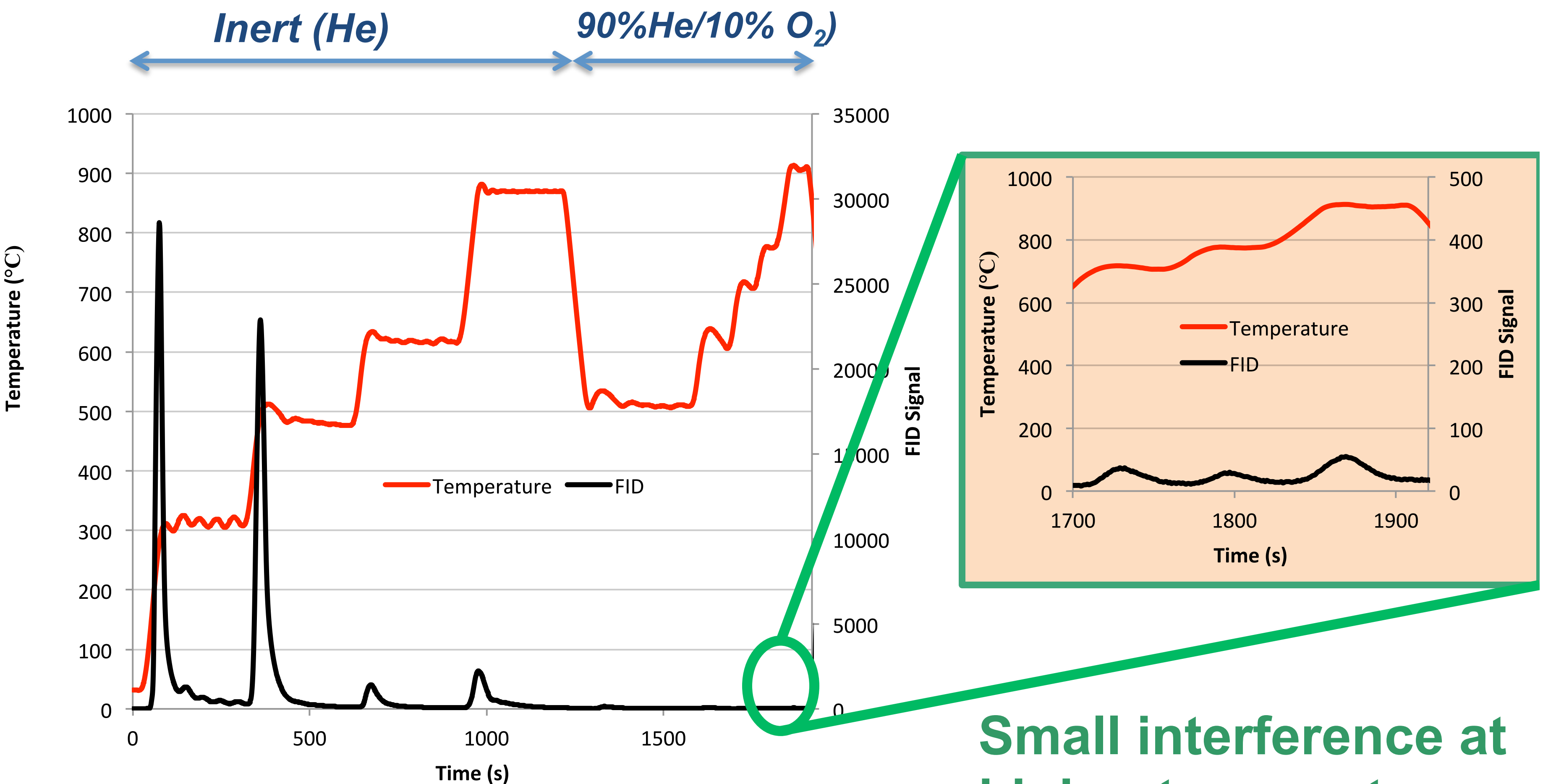
✓ **When rat lung tissue was successfully dissolved from solid to liquid** (for solid-liquid separation of CNTs)

✓ **When CNTs were not mineralized;** Subsequent TGA analysis showed many acids also lead to surface oxidation of CNTs which can change their thermal profile

Highlights of previous slide (Lessons Learned)

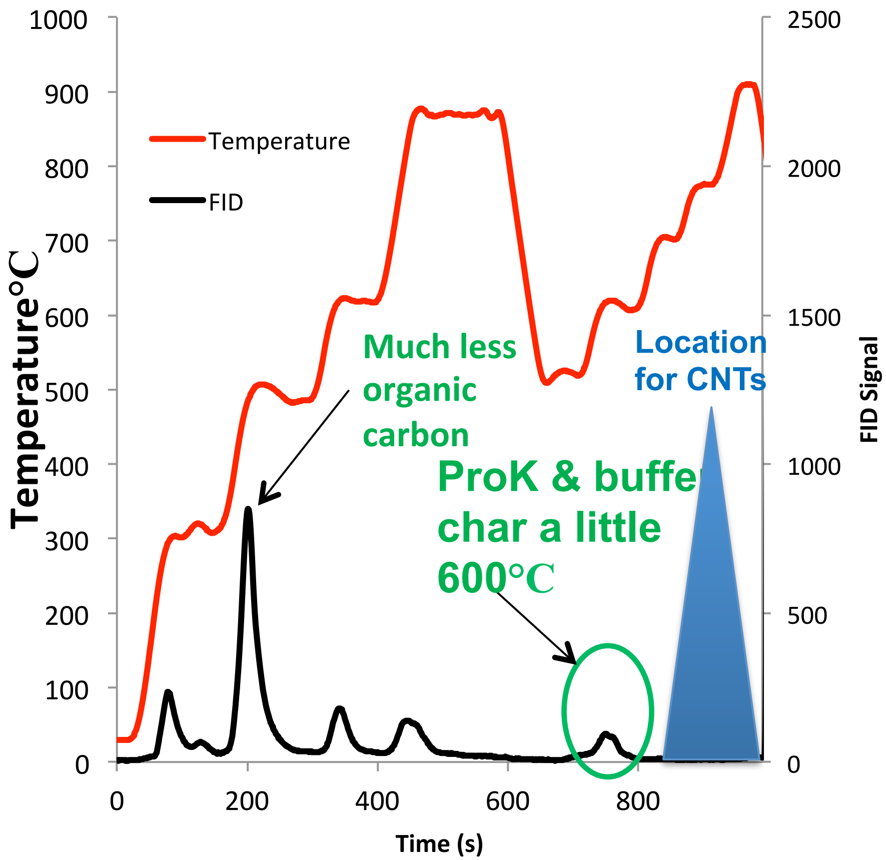
- Acids and oxidants are messy, hazardous to the analytical instrument, and some mineralize CNTs.
- Not all hydroxides are equal: TMAH and Solvable worked best, simple salt forms were not ideal (e.g., NaOH, KOH).
- A two-step digestion process involving Solvable (or TMAH) and Pro K is necessary to remove high-temperature stable carbon.
- Filtration is not an option due to sticking; centrifugation should be done at a minimum of 25,000 Gs for 20 min.

Thermograms of **Whole** Rat Lungs digested using Solvable (alone)

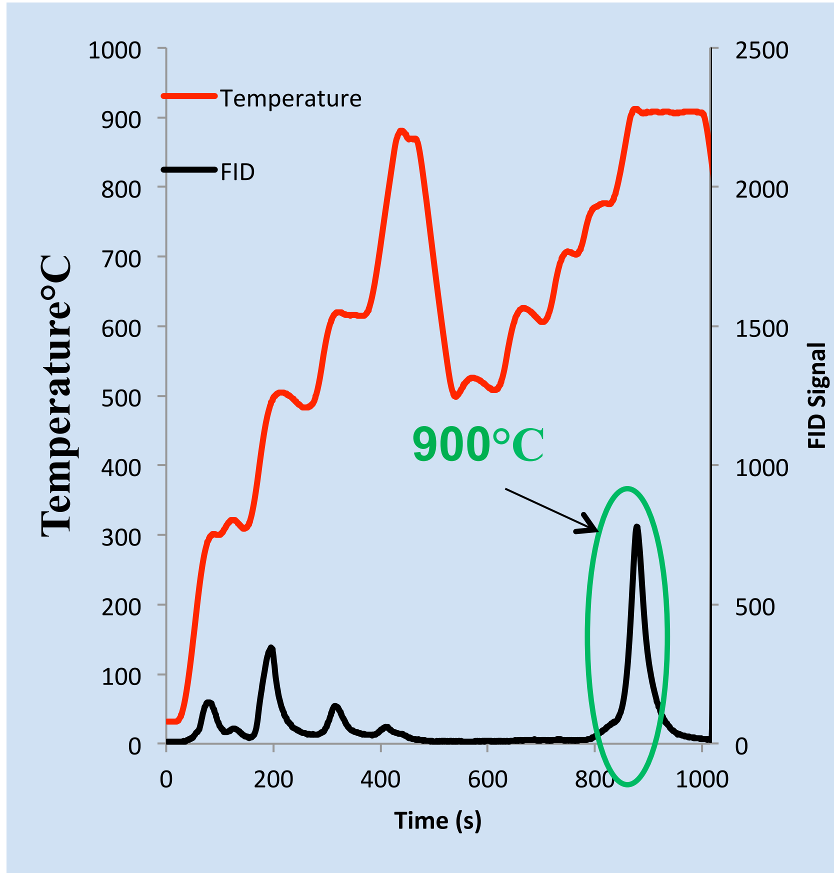


Small interference at higher temperatures where CNTs respond

Rat Lung Digested with Solvable + Pro-K reduces interferences more than Solvable Alone



Solvable and Pro K reduce charred OC & EC; EC does not interfere with CNTs



CNT Control – treated with Solvable and Pro K

Separation of CNTs from digested lung tissue

- **Need: a method to separate the CNTs from the digested lung liquid (5-15 mL)**
- Centrifugation or filtration ?
- Since we load samples onto quartz fiber filters (QFF) for TGA analysis we tried this first. Unfortunately, some sample passed through the filter because it was dispersed.
- Inducing aggregation of CNTs with salt allowed GFF to capture CNTs but they also stuck to the syringe and the filter housing.
- Centrifugation: Speeds below 25,000 Gs and times less than ~20 minutes did not produce good pellets, and some CNTs were left in suspension in the centrate. Washing with 1 M KCl in place of water enhanced the pellet stability.

Optimized CNT Extraction from Rat Lungs

DAY 1

Rat F-344
~250g

Rat Lung
Wet wt. ~1.0 g
Minced in Porcelain Crucible

Dry Lung 90C
Oven 2-4 hrs
Dry wt. ~0.2g

Solvable

Porcelain Crucible
Dry Lung
Add 5ml Solvable

1st Digest Dry Lung + Solvable
50C Over night

DAY 2

Transfer digested lung
Into 14ml tube, rinse crucible
With water, fill tube to top, mix

Spin at 30,000G 30min
Remove super

Add 60ul ProK +
200ul Buffer + 2.74ml water
In same 14ml tube, cover, 50C oven
Over night

DAY 3

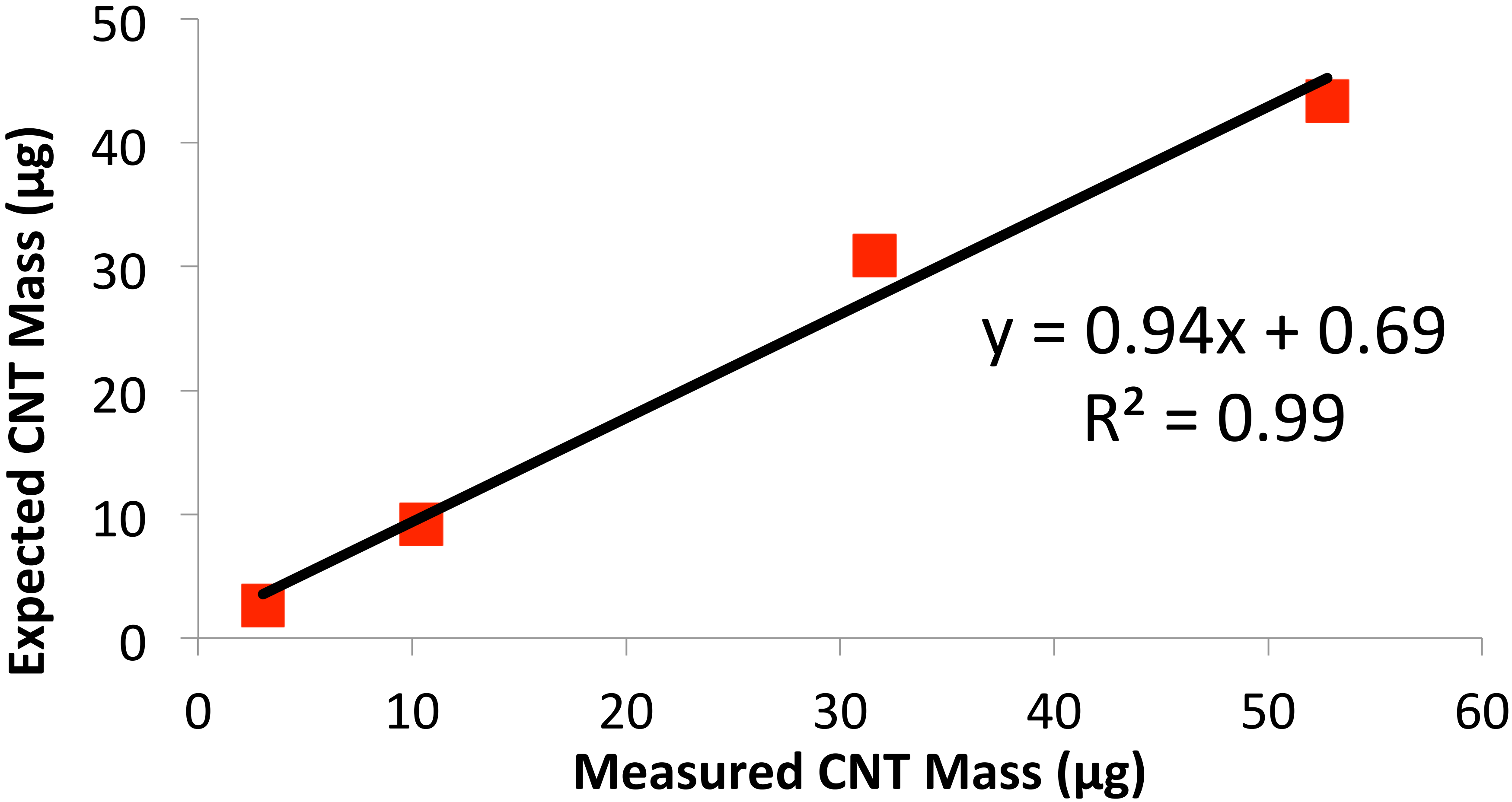
Transfer from 14ml into
2-3 1.5ml MCT

Spin 12,000G for 30min
Remove super

Combine 3 tubes
into 1 single tube
Rinse with water

Spin 12,000G 30min
Remove super
Pipette pellet onto membranes,
dry, store in foil; analysis ECOC

CNT Mass – Spike Recovery on Controls: CNTs treated with Solvable + Pro-K)



CNT Mass Recovery with Instilled Rat Lungs

- Rat lungs instilled with 3 μg of CNTs
- Digested using solvable + Pro-K and separated using previous protocol
- Residual material analyzed using programmable thermal analysis

- Controls (no lungs) for 3 μg of CNTs
  - Recovery: 3.30 μg ± 0.09 μg
- Instilled rat lungs for 3 μg of CNTs
  - Recovery: 2.80 μg ± 0.44 μg
  - Average recovery: 85%
  - Recovery was consistent
  - Losses attributed to extraction processes – CNTs lost during centrifugation & washing

CNT Summary

- Two distinct thermal classes of CNTs
  - Weak as described by an I_D/I_G ratio less than 0.75
    - Includes most MWCNTs and SWCNTs
  - Strong as described by an I_D/I_G ratio greater than 0.75
    - Includes MWCNTs with low defect density
- Hydrolysis and enzymatic digestion methods are most appropriate for CNTs and rat lung tissue

Future Work

- A robust CNT analytical method was developed
- Semi-conductor industry uses CNTs
  - Thermal packaging
  - Electrical conducting properties
- We plan to evaluate:
  - Workplace exposure (air) to CNTs
  - Release during mechanical wear of CNTs from thermal packaging

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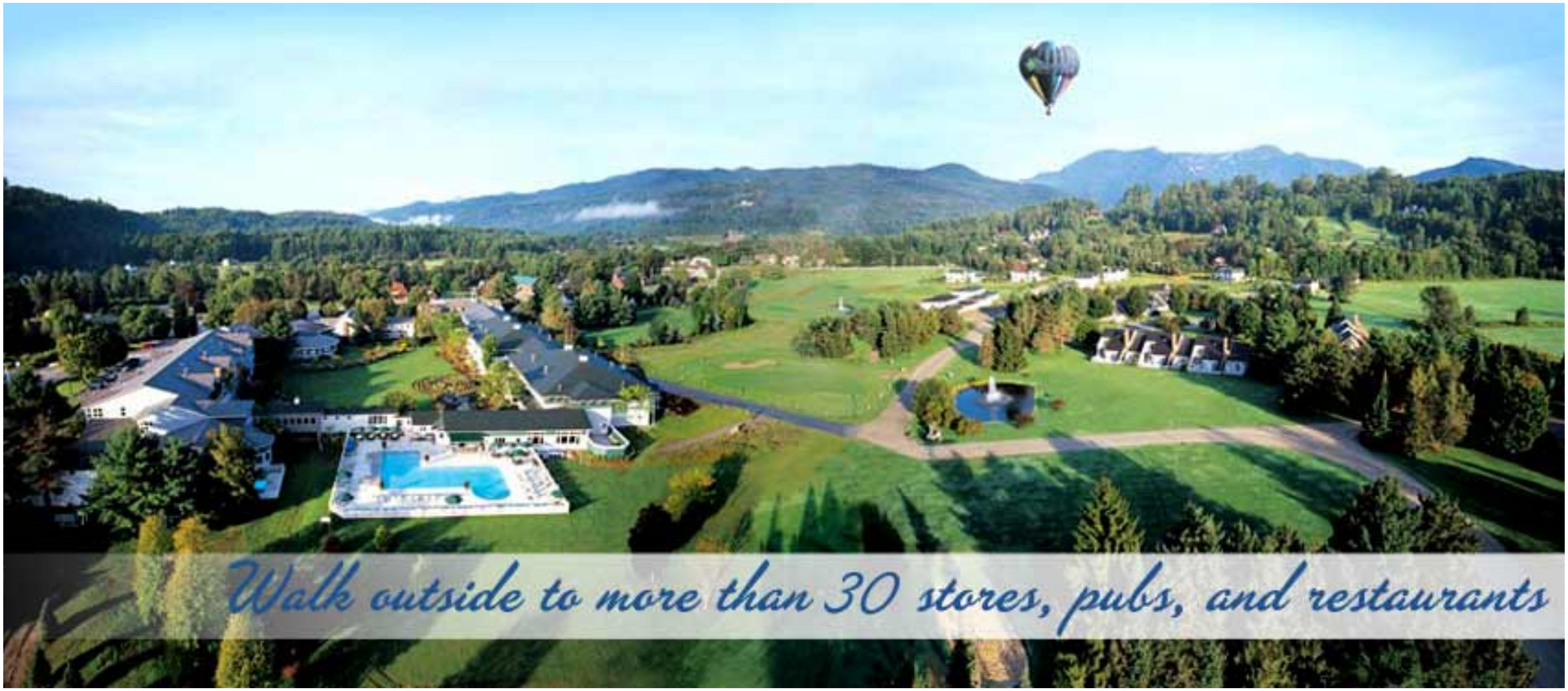

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