

Advanced BioCatalytics Corporation

Yeast-derived Surfactant Synergists for Cleaning, Bioremediation and Agriculture

Presenter: Carl Podella

Co-Presenter: Michael Goldfeld

Advanced BioCatalytics

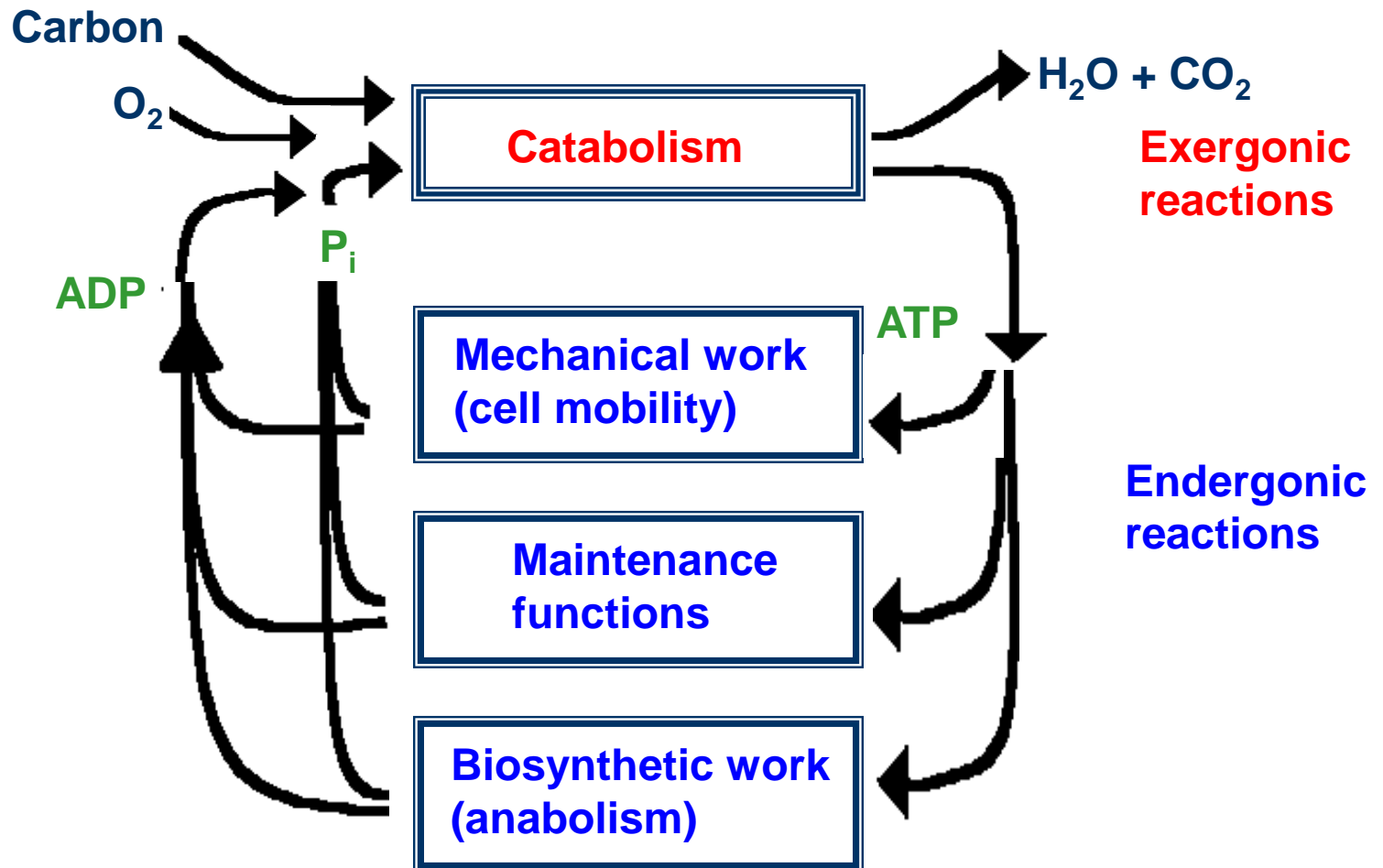
Pioneered the Use of Low Molecular Weight Proteins that:

- **Uncouple biochemical processes in bacteria**
- **Bind with surfactants to alter the functionality of most surfactants**

“UNCOUPLING FACTOR”

- **Stimulates uncoupling of oxidative phosphorylation in biochemical processes**
- **Enhances wastewater treatment**
- **Accelerates Bioremediation of hydrocarbon-contaminated soil**
- **Controls biofilm**

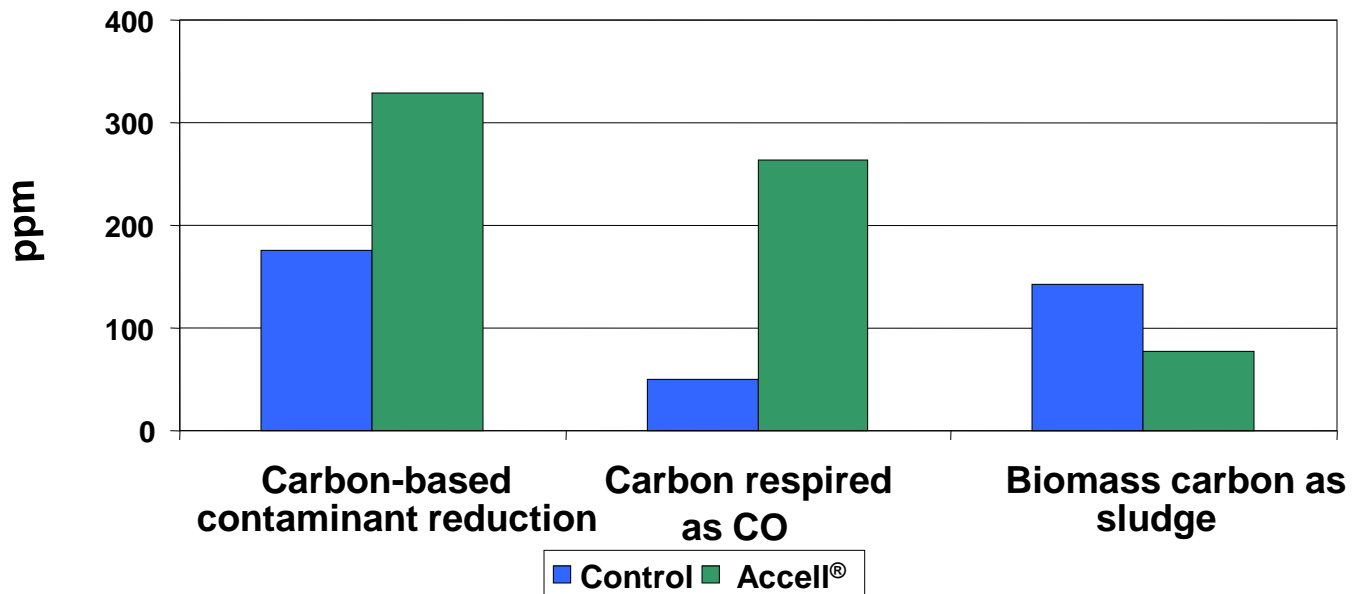
THE ROLE OF THE ATP-ADP CYCLE IN CELL METABOLISM



(Adapted from Atkinson and Mavituna, 1991)

MASS BALANCE RESPIRATION STUDY

Biodegradation of Tryptic Soy Broth in Bioreactor at 4 Hours



	Control		% Difference
Carbon -based contaminant reduction	175.3	328.6	87.5
Carbon respired as CO ₂	49.5	263.8	432.9
Biomass carbon as sludge	142.8	76.8	-46.2

TOTAL PLATE COUNTS - 4 HOURS	6.9×10^7	6.0×10^6
-------------------------------------	-------------------	-------------------

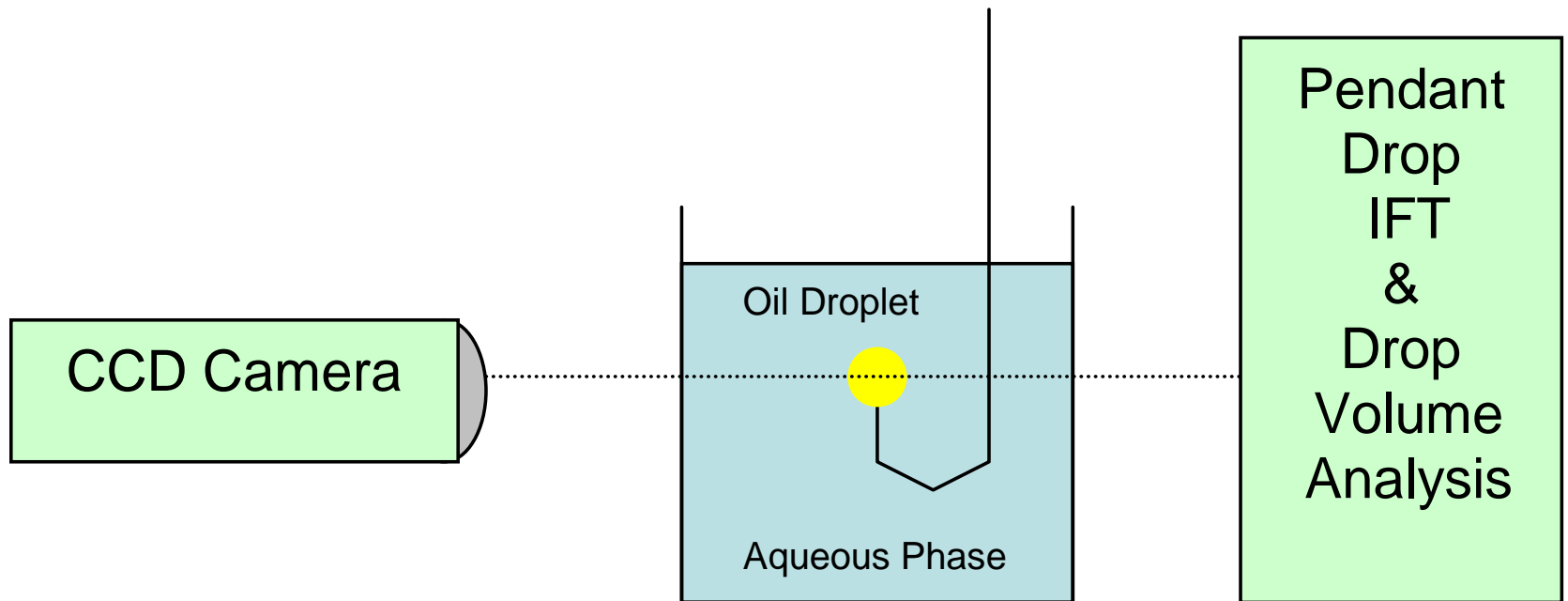
PROTEIN - SURFACTANT SYNERGIST (P-SS)

- **Low molecular weight stress proteins**
- **6,000 – 17,000 Daltons**
- **Derived from *Saccharomyces cerevisiae***
- **Highly stable over wide range of conditions**

EFFECTS OF P-SS ON SURFACTANTS

- **Reduces CMC values yielding greater cleaning power with lower surfactant levels**
- **Reduces surface and interfacial tension of surfactants**
- **Increases detergency by converting oil and grease to surfactant-like materials**
- **Allows creation of higher efficacy cleaners without harsh chemicals or solvents**
- **Has applications across a broad spectrum of surfactant types and end-use applications**

KRÜSS PENDANT DROP TENSIOMETER DSA10 WITH A KRÜSS DROP SHAPE ANALYSIS SYSTEM



ACCELL[®] EFFECT ON CMC, GREASE AND GREASE CONVERSION

Solution	CMC Pre-Grease Exposure	CMC Post-Grease Exposure	Grease Lost to Aqueous Phase	Grease Converted to Detergents	Percent Increase in Cleaning Power
10 ppm Surfactant (no proteins) in Distilled Water	442	442	1.5 %	0 %	N/A
10 ppm Accell [®] in Distilled Water	75	35	11.2 %	4.0 %	500 %

CMC: Critical Micelle Concentration (point at which detergency reached)

N/A: Not Applicable

PPM: Parts per Million (mg/l)

ACCELL[®] EFFECT ON CMC, GREASE AND GREASE CONVERSION

Solution	CMC Pre-Grease Exposure	CMC Post-Grease Exposure	Grease Lost to Aqueous Phase	Grease Converted to Detergents	Percent Increase in Cleaning Power
Microbial Sludge	N/A	67.8	4.2 %	0 %	N/A
10 ppm Accell [®] in Microbial Sludge	68.0	4.0	28.6 %	6.4 %	740 %
10 ppm 30K MWCO	70	9.0	24.4 %	6.1 %	710 %

CMC: Critical Micelle Concentration (point at which detergency reached)

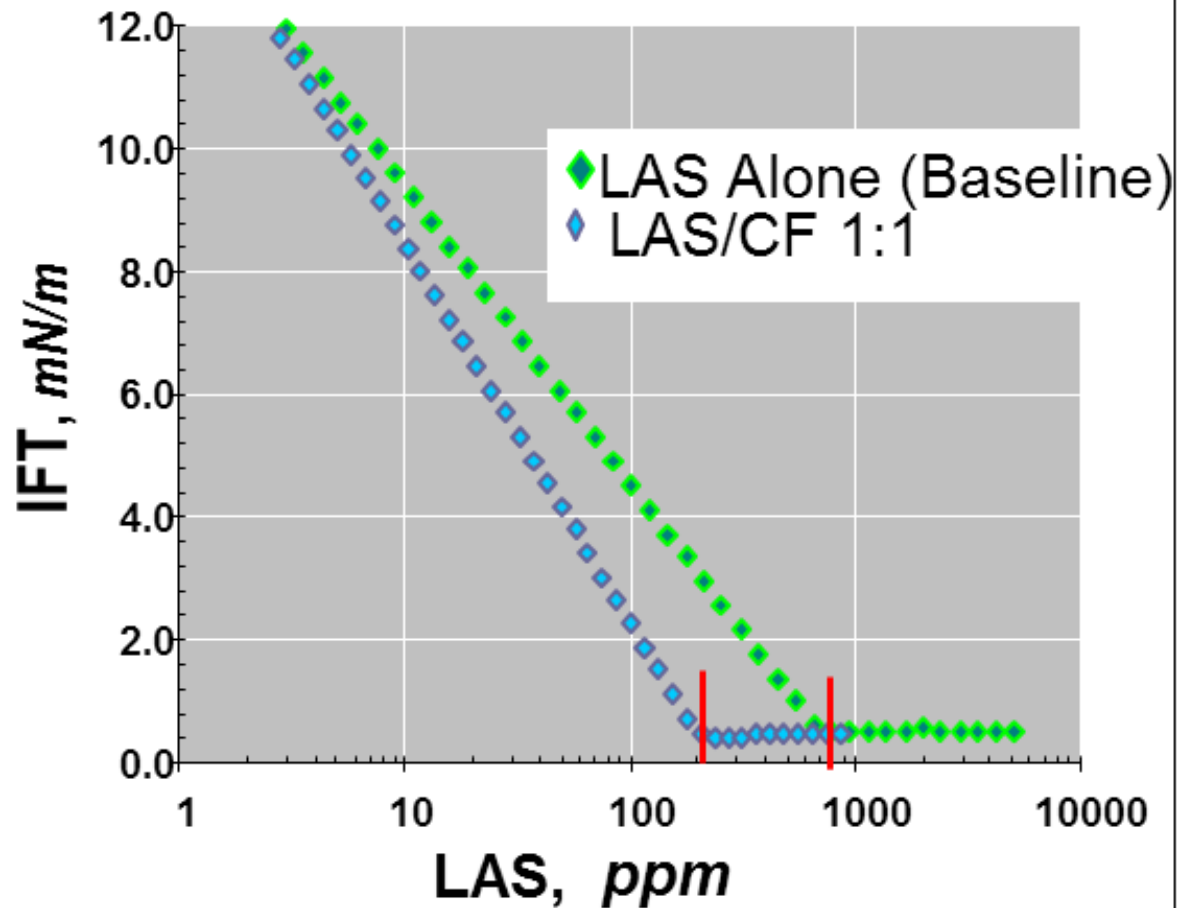
N/A: Not Applicable

PPM: Parts per Million (mg/l)

SURFACTANTS WITH & WITHOUT P-SS

CONDITION	CMC Based On Surfactant Concentration (ppm)	IFT at CMC With Canola Oil (mN/m)
Sodium Lauryl Ether Sulfate (SLES)	1240	1.76
Sodium Lauryl Ether Sulfate + P-SS	333	1.46
Linear Alkyl benzene Sodium Sulfate (LAS)	684	0.52
Linear Alkyl benzene Sodium Sulfate + P-SS	202	0.42
C9-11 Alcohol Ethoxylate 6EO	162	7.03
C9-11 Alcohol Ethoxylate 6EO + P-SS	73	6.31
C14-15 Branched Alcohol Propoxylate (4PO) Sulfate	105	0.93
C14-15 Branched Alcohol Propoxylate (4PO) Sulfate + P-SS	34	0.87

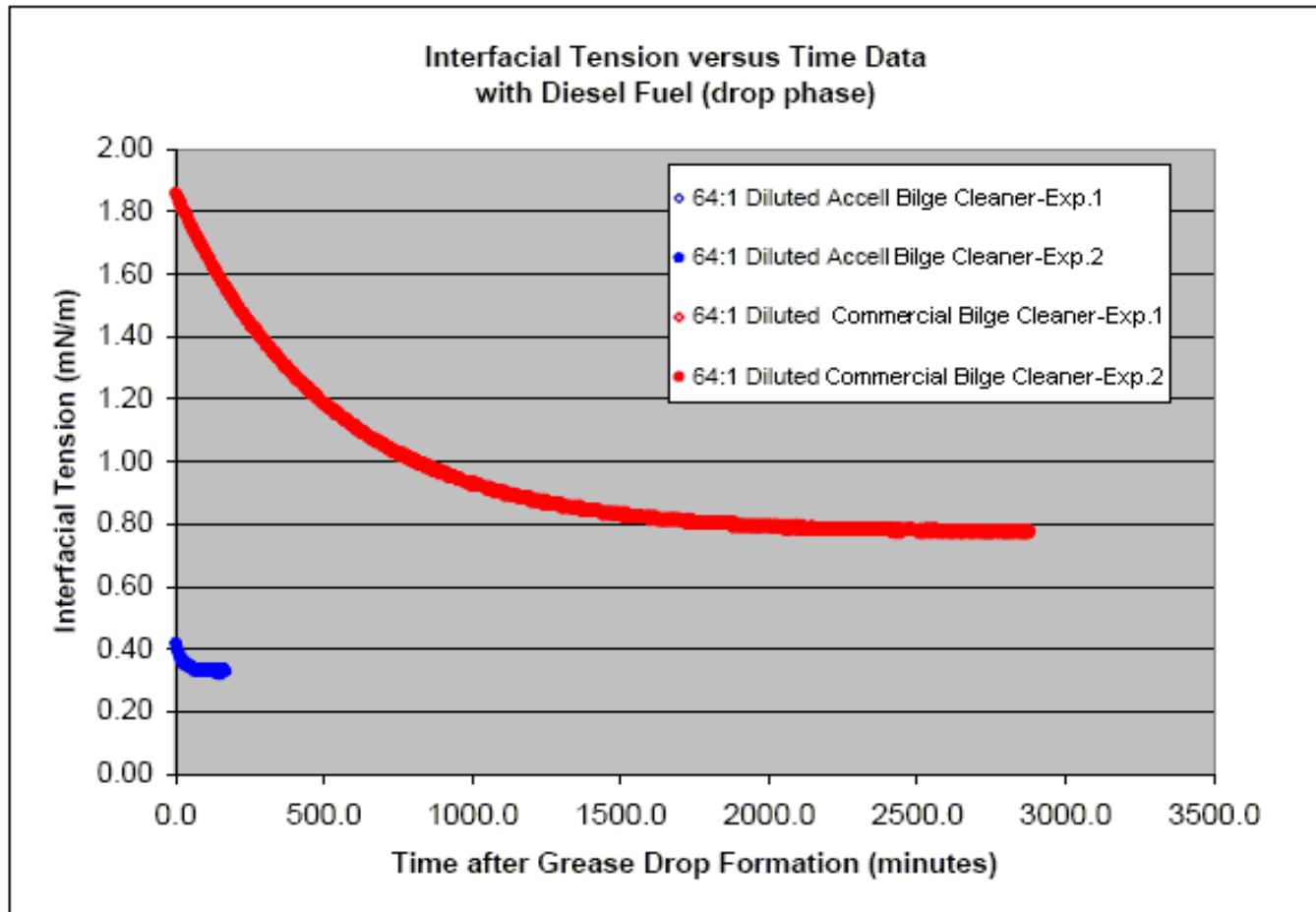
IFT & CMC in Bio-Soft D-40 (LAS, linear Na alkylbenzene sulfonate) Water / Canola Oil Interface



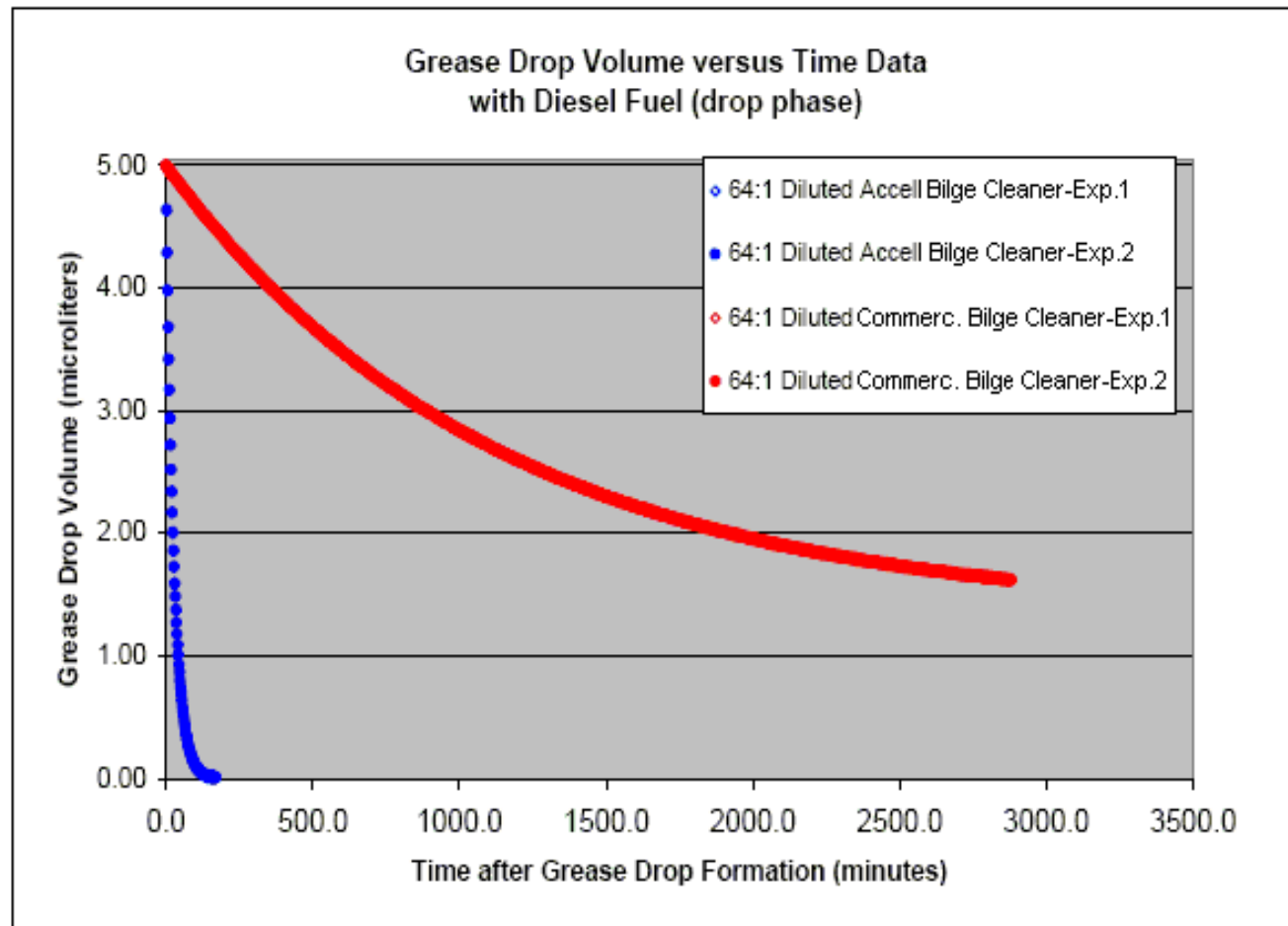
INTERFACIAL TENSIONS WITH DIESEL FUEL

Sample 2% Dilution	Test # 1 Ring Method IFT at 2% Conc. (mN/m)	Test # 2 Ring Method IFT at 2% Conc. (mN/m)	Test # 3 Ring Method IFT at 2% Conc. (mN/m)	Average Ring Method IFT at 2% Conc. (mN/m)
6 ABC 8-1	0.59	0.59	0.60	0.59
6 ABC 8-2	0.59	0.57	0.57	0.58
6 ABC 5-2	0.51	0.52	0.53	0.52
6 ABC 8-3	0.50	0.49	0.49	0.49
6 ABC 8-4	0.47	0.45	0.45	0.46
6 ABC 8-5	0.41	0.43	0.43	0.42
6 ABC 8-6	0.42	0.41	0.39	0.41
6 ABC 8-7	0.36	0.36	0.37	0.36

BILGE CLEANER FOR PLEASURE CRAFTS



BILGE CLEANER FOR PLEASURE CRAFTS

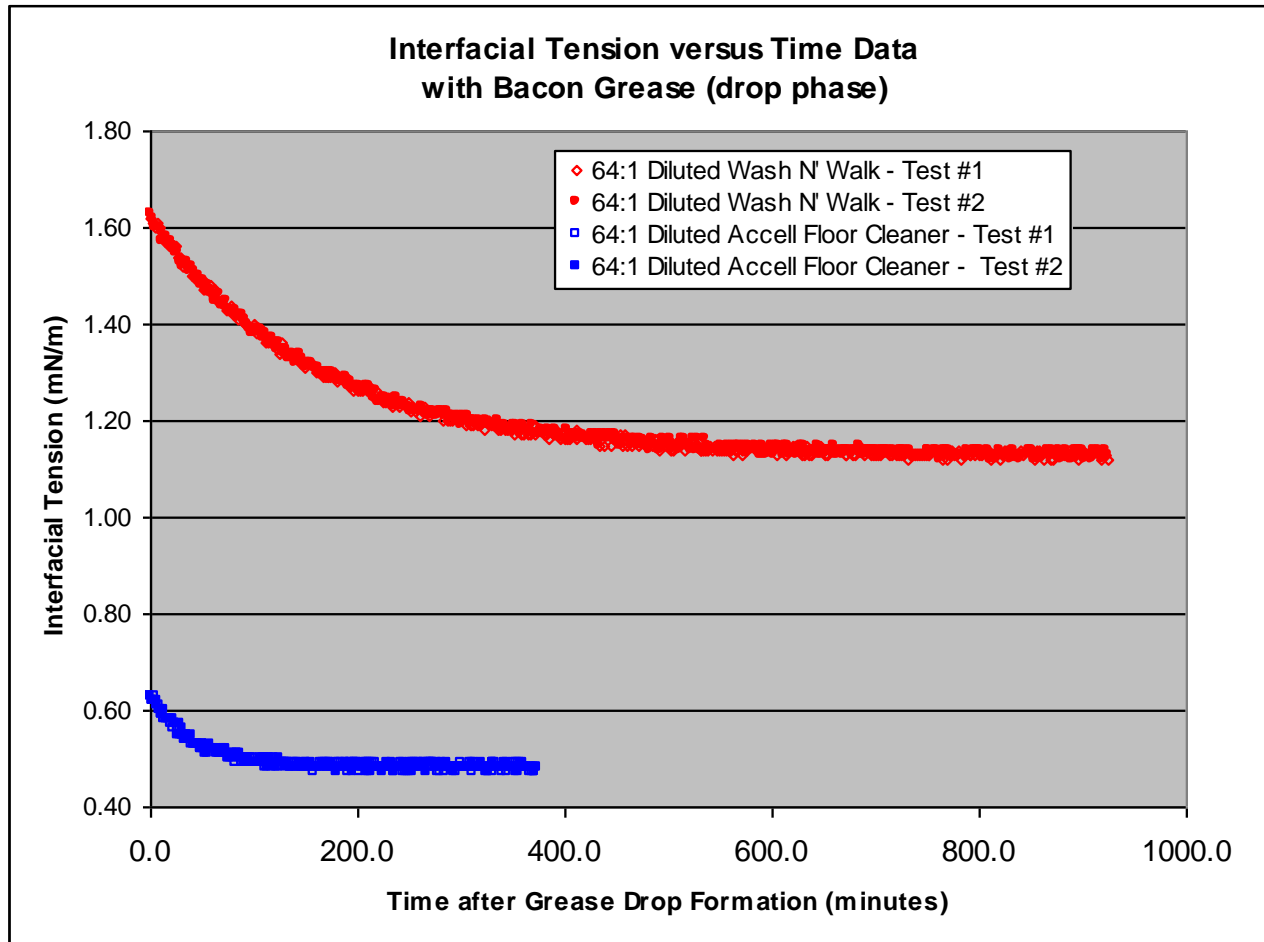






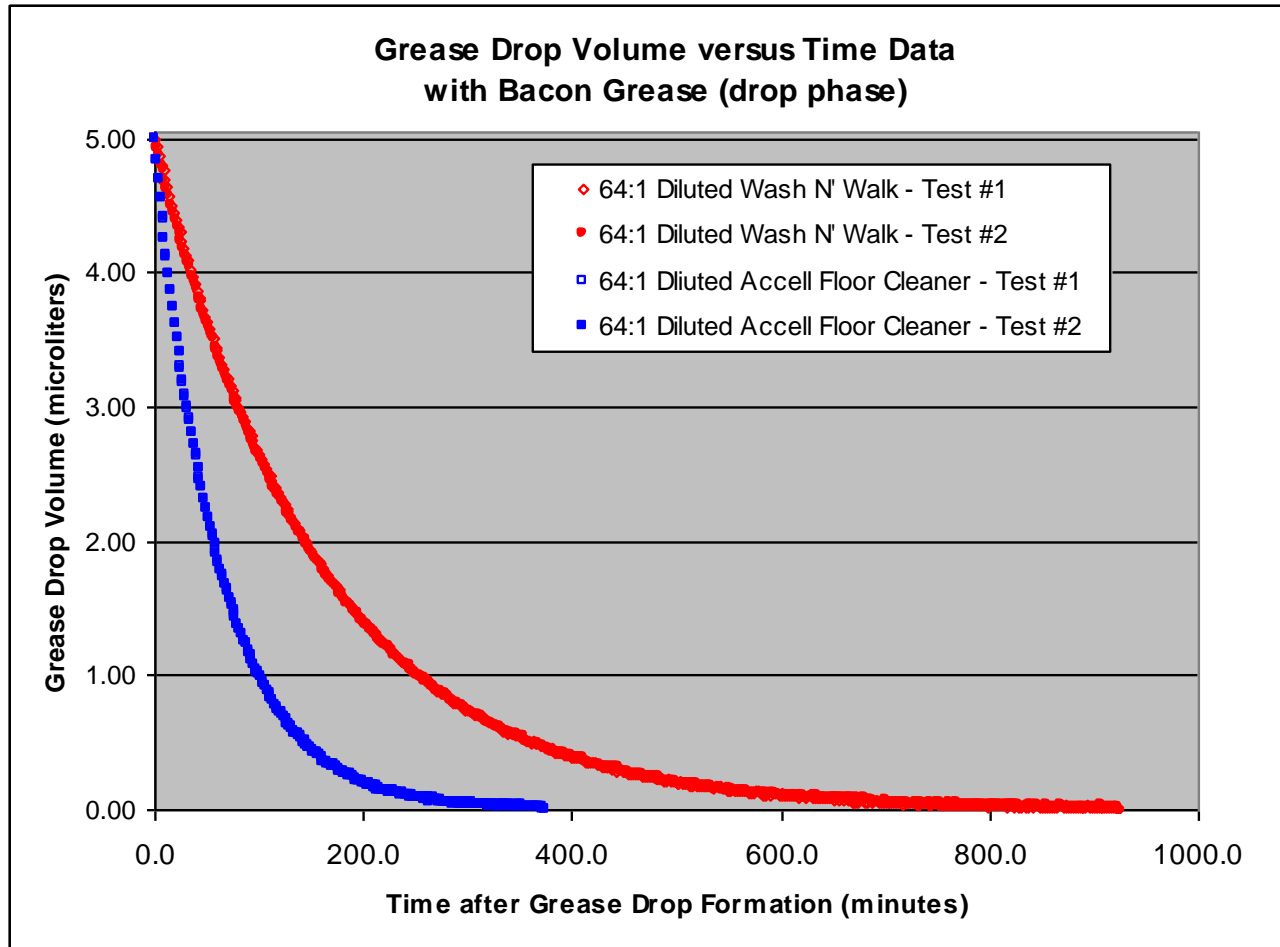
FLOOR CLEANER STUDY

Accell® Floor Cleaner vs. Ecolab Wash 'N Walk



FLOOR CLEANER STUDY

Accell[®] Floor Cleaner vs. Ecolab Wash 'N Walk



ACCELL[®] FLOOR CLEANER VS. ECOLAB WASH 'N WALK

Both Products in Dilute WAS ►	64:1 Walk N' Wash (Bacon Grease)	64:1 Accell Floor Cleaner (Bacon Grease)
► PRE GREASE EXPOSURE SOLUTION PROPERTIES		
Surface tension prior to grease drop exposure (mN/m)	19.71	26.52
► PROPERTIES AND EFFECTS DURING GREASE EXPOSURE		
Initial interfacial tension upon exposure to grease (mN/m)	1.63	0.63
Equilibrium interfacial tension with grease (mN/m)	1.13	0.48
Time frame for interfacial tension equilibration (minutes)	560	112
Grease drop volume after 2880 minutes = 48 hours (ul)	0.00 after 920 min	0.00 after 370 min
Time frame for grease drop volume equilibration (minutes)	920	370
► POST GREASE EXPOSURE SOLUTION PROPERTIES		
Surface tension of 5.0 ml retain after grease exposure (mN/m)	19.69	26.19
CMC after grease drop exposure (ppm)	60	21
► CALCULATED PROPERTIES BASED ON THE DATA ABOVE		
Concentration of the aqueous retain in terms of converted grease (ppm)	1000	1000
Percentage of the 5.0 ul grease converted to "surfactant-like" materials (%)	2.3	13.2

ACCELL[®] FLOOR CLEANER VS. ECOLAB WASH 'N WALK



SOME OTHER IMPLICATIONS

AGRICULTURE: ENHANCING WATER & PESTICIDE UPTAKE BY ROOTS & LEAVES

Currently, *ca.* **0.1%** herbicide applied reach the targeted structure in plant.

Adjuvants – mostly surfactants – increase uptake/efficiency of watering and pesticide applications, and reduce environmental impact.



EFFECT OF P-SS ON CMC VALUES

Surfactant	CMC, <i>ppm</i>	
	No P-SS	With P-SS
Lauramine Oxide	99.78	13.03
Na Lauryl Ether Sulfate	642.6	115.2
1:2 LO - SLES	146.1	7.72

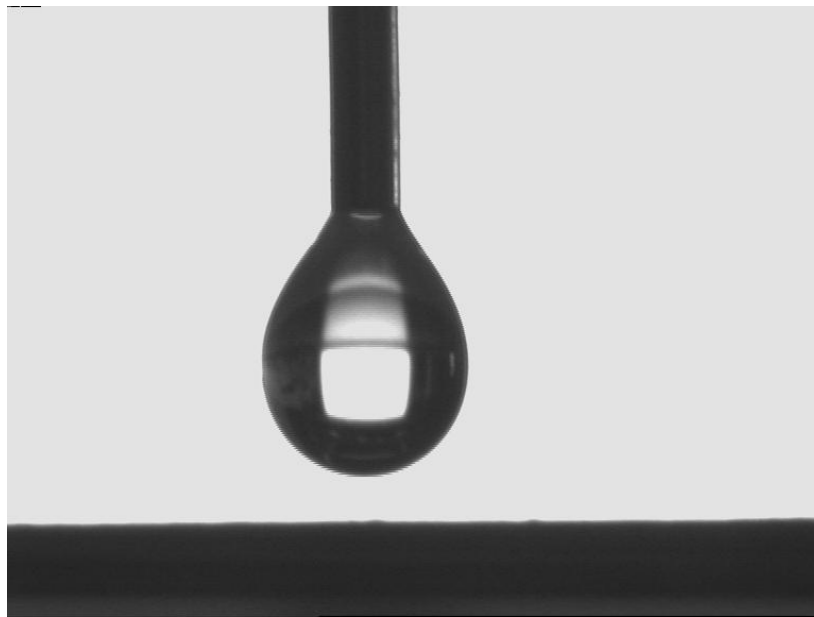
P-SS/SURFACTANT ADJUVANTS IN AGRICULTURE APPLICATIONS

80:1 Dilution in Water of	Leaf	Average Approximate Time to Complete Droplet Penetration (seconds)	Equilibrium Non-Penetrated Drop Volume (microliters)	Equilibrium Contact Angle (degrees)
Ethox. Tallow Amine	Cabbage	600+ (∞)	0.35	28.6
Ethox. Tallow Amine + P-SS	Cabbage	233	0.00	0.0
Lauramine Oxide	Cabbage	600+ (∞)	0.21	18.8
Lauramine Oxide + P-SS	Cabbage	194	0.00	0.0
Na Lauryl Ether Sulfate	Cabbage	600+ (∞)	0.18	18.1
Na Lauryl Ether Sulfate + P-SS	Cabbage	185	0.00	0.0
1:2 LO - SLES	Cabbage	600+ (∞)	0.09	12.5
1:2 LO – SLES + P-SS	Cabbage	159	0.00	0.0

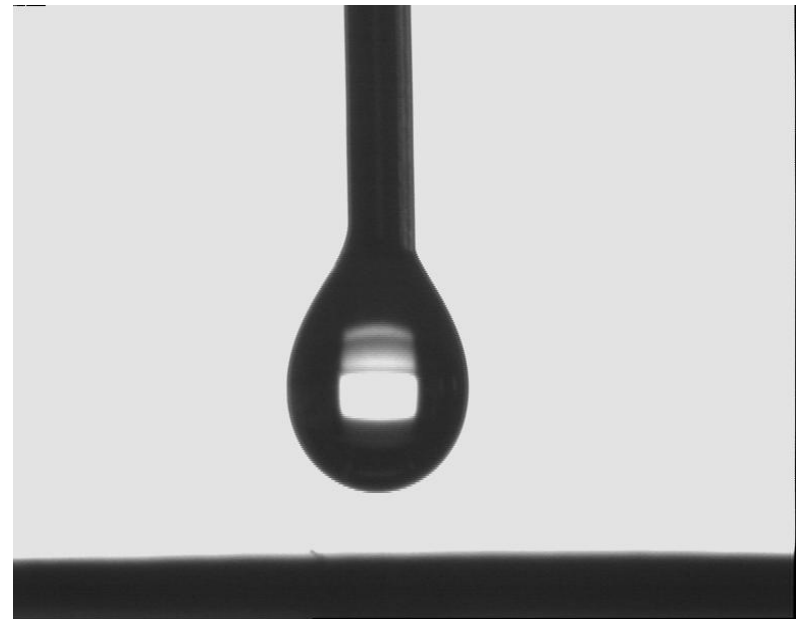
COMMERCIAL DETERGENT vs. ABC PRODUCT

Comparison of Wetting & Uptake for Two Aqueous Solutions on Tomato & Cabbage Leaves

0.25% NPE droplet on Cabbage Leaf



0.25% ABC droplet on Cabbage Leaf

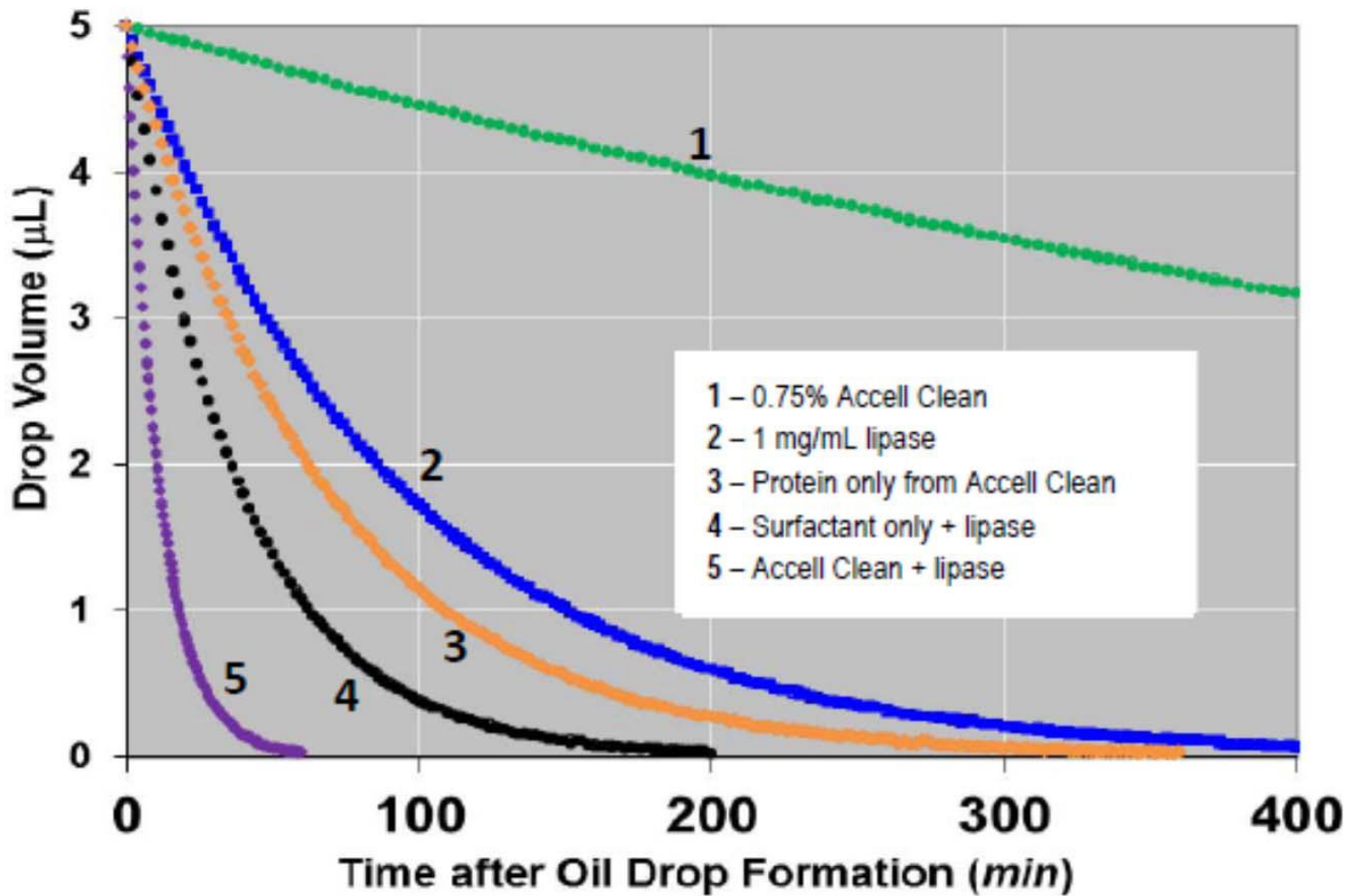


SURFACTANT AND ENZYME SYNERGIST

Statistics from 5.0 Microliter Prime Burning Peacock Oil Consumption and IFT Experiments	1.5% Accell Clean	1 mg/ml Lipase	0.75% Accell Clean	0.75% Accell Clean and 1 mg/ml Lipase
Initial IFT (mN/m)	1.54	16.62	2.03	1.98
Time of 50% Digestion (minutes)	348	66	605	7.9
IFT at 50% Digestion (mN/m)	1.34	10.41	1.77	1.24
Time of 99% Digestion (minutes)	2196	418	3816	50
IFT at 99% Digestion (mN/m)	1.13	4.52	1.49	0.62

ENZYME SYNERGIST

OIL DROP DIGESTION: VOLUME vs. TIME



CONCLUSIONS:

- **ABC's Surfactant Synergist Technology Enhances Surfactant Performance through Reduced Interfacial and Surface Tension and Critical Micelle Concentration Values**
- **Surfactant Synergist Facilitates the Formulation of Highly Efficacious Products without the need for Caustic Materials or Solvents such as Glycol Ethers or Alcohols**
- **Unlike Enzymes, the Surfactant Synergist Proteins are Highly Stable over a wide range of Temperatures and pH Values, providing Excellent Shelf Life**
- **ABC's Surfactant Synergist Provides Greater Efficiency at Lower Concentrations**
- **The Surfactant Synergist Technology is Compatible with the "Green Movement" because it is Fermented from Renewable Materials**