Detergency

A need for qualification
A necessary innovation
A proven efficacy
Why better detergents’ performances?

• Techniques evolution
  – material
  – efficacy / time

• Biocide Directive
  – ecotoxicological data

• Biological data evolution
  – nv MCJ . . .
Definitions (1)

**Detergent** (Commission Recommendation 89/542/EEC)

- Products intended for washing and cleaning purposes or for use in connection with washing and cleaning process.... (washing machines; dish washers)
- The definition also applies to products used to prewash or whiten, fabric softeners or any product intended to be used for cleaning purposes...
**Definitions (2)**

**Detergency**

- Process by which soil is removed from a surface and undergoes solubilization or dispersion. Result of several physicochemical phenomena taking place at the interface of three phases: surface/soil/detergent.

- The phenomena are:
  - Wetting of surface.
  - Removal of soil from surface.
  - Avoiding re-deposition of soil on surface.
The detergent must come into contact with the surface so that ... ( \( F_a = \) adherence force )

- \( F_{\text{detergent/surface}} > F_{\text{soil/surface}} \)

- To lower the superficial tension of the detergent solution...
- ...and the interfacial tensions between aqueous bath, soil and surface
Basic principles
Detergency: 2 / Removal of soil

surface / soil + detergent

→

surface / detergent + soil / detergent

• The detergent solution wets the surface, is absorbed by it and lowers the surface’s attraction to allow the soil to separate itself from the surface.
Basic principles

Wetting

\[ \theta_1, \theta_2 \]

\[ \theta = \text{contact angle} \]
Basic principles

Surface tensions

- Glass: 177
- Teflon (PTFE): 18
- Mercury: 475
- Water: 72
- Water + Surfactants: 30
Basic principles
Detergency : 3 / avoiding re-deposition

• Chemical reactions
  – lipids undergo saponification
  – mineral soil undergoes solubilization
  – soil undergoes emulsification
  • Liquid soil = hydrophobic ; detergent solution = hydrophilic.

Surfactant potentials and émulsíateur des détergents
How can we better detergents’ performances?

- By understanding the mechanisms of detergency
- By apprehending what a detergent is made off and how it works
- By comparing performances
How can we better detergents’ performances?

• By having evaluation methods
  – easy and fast
  – reproductible
  – closely related to field problems
    • soil
    • surface . . .
Problems of Detergency

or

How can one eliminate soil made of unidentified compounds from an unspecified surface?
A strategy ...

To make water-soluble a substance which is not (or slightly) water-soluble...

...to gain efficacy when rinsing off.
Detergency

• Detergent action parameters
Detergent action parameters

- Soil nature
  - % Proteins
  - % Lipids
  - % Sugar
  - % Mineral
Detergent action parameters

• Soil state
Detergent action parameters

- Mechanical action
How can we eliminate soil?
with water?
a lot of water . . . ?
With detergent molecules... which ones...?
Which detergent molecule?

Hydrophobic carbonaceous chain

Hydrophilic part

Example
Lauric alcohol
ethoxylated
H.L.B. definition

H.L.B. (Hydrophilic-Lipophilic Balance) is a measure of the solubility of a substance in water and oil. A value of 0 indicates complete water solubility, while a value of 20 indicates complete oil solubility. Substances with an H.L.B. value between 3 and 8 are generally considered to be surfactants, which are effective at emulsifying oil and water.

The graph shows the distribution of oil-soluble and water-soluble substances across different H.L.B. values. D1 and D2 indicate specific points or substances within this spectrum.
Detergent molecule (D1)

Hydrophilic part

Hydrophobic carbonaceous chain

High H.L.B. : lack of tropism for organic soil

H.L.B. : Hydrophilic Lipophilic Balance
Detergent molecule (D2)

Hydrophilic part

Hydrophobic carbonaceous chain

Low H.L.B. : lack of affinity for water
Choice of Detergent and H.L.B. value

• A water-soluble protein can be masked by a lipidic matrix and therefore be non-water soluble. In the end, this protein is poorly eliminated by water.

• Protein solubilization is not the only paramount factor in choosing a detergent. One should also consider the hydrophilic/lipophilic nature of soil and surface.
Choice of Detergent

• Detergent’s other properties:
  – wetting effect or surfactant effect
  – emulsifying effect
  – dispersive effect
  – solubilizing effect

• Complementary properties due to pH
  – saponifying effect
Optimized elimination of water?

Once surface is dry... traces and deposits of minerals contained in water remain...
Elimination of water with wetting effect

Water is effectively eliminated. Traces and deposits are limited once surface is dry.
Wetting effect

Water

72 mN/m

Water, without the wetting effect or with a poor wetting effect, does not penetrate cracks properly.

Water + Surfactants

29 mN/m

Unaltered soil

Soil is attacked

Water, without the wetting effect or with a poor wetting effect, does not penetrate cracks properly.
Wetting effect

Water
72 mN/m

Water + Surfactants
29 mN/m

mN/m : Milli Newton meter

No contact with micro organism

Contact hence activity
Elimination of fat

Water cannot eliminate fat fixed to surfaces.
Elimination of fat

Emulsifying effect

Emulsified fat is eliminated with water.
Elimination of matter in suspension

Matter in suspension settles on surfaces.
Elimination of matter in suspension
Dispersive effect

Matter is dispersed and eliminated with water.
Elimination of ± soluble matter

Matter which is not well solubilized creates a film on surfaces.
Elimination of ± soluble matter

Solubilizing effet

Solubilized matter is easily eliminated.
Elimination of fat

Water cannot eliminate fat fixed to surfaces.
Elimination of fats
Saponifying effect

Fat undergoes saponification in very alkaline medium (sodium hydroxide, potassium hydroxide …) : pH >11 and is eliminated with water.
In theory . . .

• Soil is made of:
  – Sugar
  – Lipids
  – Proteins
  – Limestone
  – Dust
Reality is more complex !!!

- Basic sugar + Exopolysaccharides + Glycoproteins + Lipopolysaccharides...
- Lipids + Lipopolysaccharides + Lipoproteins...
- Proteins + Glycoproteins + Lipoproteins...
- Limestone + « protected » limestone
- Dust and other particules (iron, copper…)
How can one stimulate the detergent power of surfactants?

Enzymatic Power Assessment
Enzymology

• To assess the role of enzymes in detergency in addition to surfactants:
  – Enzymes’ ability to fraction slightly soluble matter into more water-soluble parts.
  – !: The difficulty lies in choosing the enzyme. The enzyme should not be too substrate-specific.
Enzymatic activity

Enzyme + Substrate $\rightarrow$ Enzyme + Hydrolyzed substrate or degradation products

- The enzyme is not consumed in the reaction.
- Its action continues on the substrate as long as environmental conditions are respected (temperature, pH...).
Enzymatic activity

Enzyme recognizes substrate, hence enzymatic activity
Enzymatic activity (2)

Enzyme does not recognize substrate. No enzymatic activity.
Protease action

Albumin + Protease

(only slightly soluble)

(Soluble)

Peptides of various weight
and in the end Amino acids

Control of the protease activity by electrophoresis
Electrophoresis principles

(1)

- Proteins to be analyzed are treated with S.D.S. (sodium dodecyl sulfate) which charges them negatively.
- The proteins are then set in an electrical field.
- The proteins move from the cathode (−) towards the anode (+).
Electrophoresis principles (2)
Electrophoresis principles (3)

- Their movement is slowed down by their size and/or their weight
- The biggest molecules are retained and slowed down more by the polycrylamide gel (ex. : human albumin)
- The smallest molecules are less retained and slowed down by the polycrylamide gel (ex. : albumin hydrolysis products : peptides)
Albumin is not hydrolyzed because there is no enzymatic action.

Albumin is hydrolyzed by protease and hydrolysis products are present.
Deposit of human albumin

Human albumin + Aniosyme DD 1
T0 + 15 mn

Human albumin + Aniosyme DD 1
T0 + 4 hours

Human albumin + Aniosyme DD 1
T0 + 8 hours

Human albumin + Aniosyme DD 1
Ultrasounds: 5 x 15 mn

Human albumin + Aniosyme DD 1
without protease

Absence of albumin hydrolysis

Products of albumin hydrolysis by protease

Proteasic activity of Aniosyme DD1 vis-a-vis human albumin
(Electrophoresis on gel)
Lipase action

Triglycerides + Lipase
(only slightly soluble)

Diglycerides + Lipase

Monoglycerides + Lipase
(Soluble)

Glycerol

Control of the lipasic activity by chromatography
Chromatography principles

Raw material or a substance to be analyzed is put on a surface (gel, paper . . .) which is then partially immersed in a tank of solvant.
Chromatography principles (2)

Solvent migrates by capillary action on and in the surface.
Chromatography principles (3)

The various compounds migrate according to their solubility in the solvent.
Chromatography principles
(4)
Chromatography principles (5)
Chromatography principles (6)
Chromatography principles (6)

Most solvant-soluble compounds

Less solvant-soluble compounds
Example N° 1 : A solvent efficient on lipids such as acetone or hexane is used

Most solvent-soluble compound : Triglycerides

Less solvent-soluble compound : Glycerol
Example N° 2: Water used as solvent

Most water-soluble compound: Glycerol

Less water-soluble compound: Triglycerides
Stain identification

The analyzed compounds are often invisible to the naked eye. Therefore an *indicator* (reagent ?) is sprayed on the surface...

... to color and make them visible.
Lipasic activity of Aniosyme DD 1 vis-a-vis triglycerides

(Chromatography in thin layer on silica gel)

- Under the solvent’s influence (Hexane ...), triglycerides migrate heavily, diglycerides migrate moderately and monoglycerides migrate slightly. Glycerol does not migrate at all.
Analysis of an unidentified compound by chromatography

Identified active ingredient A

Identified active ingredient B

Unidentified product X to be analyzed

Identified active ingredient C
After chromatography and *revelation*

A, B and C have migrated differently.

The unidentified X deposit has produced 2 stains which match B and C.

Therefore, X contains raw material B and C.
Lipasic activity of Aniosyme DD 1 vis-a-vis triglycerides

(Chromatography in thin layer on silica gel)

Absence of triglycerides

Products of triglycerides hydrolysis

Triglycerides deposit

Diglycerides deposit

Monoglycerides deposit

Glycerol deposit

- Triglycerides are mixed with Aniosyme DD 1. Lipase hydrolyzes the triglycerides in diglycerides, monoglycerides and glycerol.

- Chromatography demonstrates that there are no triglycerides left but rather the products of degradation of triglycerides.
Lipasic activity of Aniosyme DD 1 vis-a-vis triglycerides
(Chromatography in thin layer on silica gel)

• Aniosyme DD 1 without lipase cannot hydrolyze triglycerides. Non-hydrolyzed triglycerides are found but hydrolysis products such as diglycerides, monoglycerides or glycerol are not.
Lipasic activity of Aniosyme DD 1 vis-a-vis triglycerides

(Chromatography in thin layer on silica gel)

Absence of triglycerides

Products of triglycerides hydrolysis

Migration

Triglycerides + Aniosyme DD 1 without Lipase

Ultrasounds: 5 x 15 mn

Triglycerides deposit

Diglycerides deposit

Monoglycerides deposit

Glycerol deposit

Triglycerides + Aniosyme DD 1
T0 + 15 mn

Triglycerides + Aniosyme DD 1
T0 + 4 hours

Triglycerides + Aniosyme DD 1
T0 + 8 hours

Triglycerides + Aniosyme DD 1

Non-diglycerides Products of triglycerides hydrolysis
Amylase action

Glycogen + Amylase

(only slightly soluble)

Polyosides and in the end Glucose

(Soluble)
Amylase activity of Aniosyme DD 1 vis-a-vis glycogen
(Chromatography in thin layer on silica gel)

Glycogen deposit

Glucose deposit

Glycogen + Aniosyme DD 1
T0 + 15 mn

Glycogen + Aniosyme DD 1
T0 + 4 hours

Glycogen + Aniosyme DD 1
T0 + 8 hours

Glycogen + Aniosyme DD 1
Ultrasounds : 5 x 15 mn

Products of glycogen hydrolysis
We demonstrate:

- The specific activity of the 3 enzymes
  - Protease on human Albumin
  - Lipase on triglycerides
  - Amylase on glycogen

- Enzymes stability
  - In solution after 15 mn, 4 and 8 hours’ preparation

- Enzymatic non cannibalism
  - The lipases and amylases are active and are not destructed by the protease

- The stability in ultrasound bath
A High Performance Detergency

– by surfactants: demonstrated on organic soil (brain + animal fats) and on artificial monobacterial biofilm

– by enzymatic action: demonstrated on organic substrates (Human albumin - Triglycerides -Glycogen)