STABILIZATION OF FUNCTIONAL INGREDIENTS BY MICROENCAPSULATION: INTERFACIAL POLYMERISATION

by

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ABSTRACT

Perfume is an expensive ingredient for most laundry detergents. To target its delivery to the fabric fibres at the right moment after the wash, improve its performance and reduce costs, using perfume microcapsules is one of the technologies that have been developed. Old technology based on melamine-formaldehyde resins presents some safety and environmental issues and current microcapsules made by interfacial polymerisation techniques do not provide the desired performance. In this work it has been done a deep study of the interfacial polymerisation process focusing on the effect that the formulation and process conditions have on the final properties of the microcapsules produced.

The microcapsule walls have been characterized by SEM, TEM and FTIR. The encapsulation efficiency, release profile of the perfume from the microcapsules and their mechanical properties have also been measured. Microcapsules prepared at low temperature with a mix of trimesoyl and terephthaloyl chloride as organic monomers and diethylenetriamine, hexamethylenediamine and ethylenediamine as aqueous monomers showed good mechanical strength and low permeability which make them of industrial interest.

Microencapsulation of glycerol for its potential use in lipsticks and other cosmetic products has also been achieved. The use of a salt (magnesium sulphate) greatly stabilized the emulsion and permitted to form small and uniform microcapsules.

The process conditions selected may also be applied to encapsulate other oil-based or water soluble active ingredients for various industrial applications.

To,

my family and in memory

of my grandparents, Jose and Angela.

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NOMENCLATURE

Abbreviation	Stands for:
d32	sauter diameter
DETA	diethylenetriamine
DSC	differential scanning calorimetry
EDA	ethylenediamine
erf	error function
FID	flame ionization detector
FTIR	Fourier transform infrared
Fv	cumulative volume frequency
GC	gas chromatography
HCl	hydrochloric acid
HMDA	hexamethylenediamine
IFP	interfacial polymerisation
meq	milliequivalents
MF	melamine-formaldehyde
PDF or Pv	probability density function for drop volume
PMC	perfume microcapsule
PVA	polyvinyl alcohol
SC	sebacoyl chloride
SEM	scanning electron microscopy
TC	terephthaloyl chloride
TEM	transmission electron microscopy
TETA	triethylenetetramine
Trim	trimesoyl chloride
UF	urea-formaldehyde
UV	ultra violet

CHAPTER 1:

INTRODUCTION

The whole of science is nothing more

than a refinement of everyday thinking.

Albert Einstein

In the last century life expectancy has greatly increased due to the advances in nutrition and hygiene (Kinsella, 1992). The improvement of soap production made available for everyone a cheap method to disinfect and the development of synthetic detergents made them even cheaper and easier to use. Nowadays there are detergent compositions developed for specific uses (like dish, hand or laundry washing), but most of them are based on petro-chemicals and in the current world scenario with oil shortages and high prices their production is not sustainable. It is required to look for new product formulations and to reduce our dependence on chemicals.

The EU was aware of these problems and as part of the Sixth Framework Program for Research and Technological Development (FP6), a project titled "BIOSEAL" was launched. In this project several universities joined efforts with a commercial partner, Procter & Gamble (P&G), to produce the next generation of detergents. The main objective of the project was to compact the detergent formulation and to do so new technologies needed to be developed. The University of Birmingham was in charge of the microencapsulation of actives and the first active of interest identified by our industrial partner was perfume.

The main objective of detergents and other household cleaning products is to clean and disinfect, but customers usually perceive their action by the fresh odour left after their use. That "clean smell" is what gives them an idea of the performance of the product and influences the customers to choose product. The necessity to incorporate an odorant makes perfume one of the most important ingredients in the formulation, although it has not an active purpose.

Perfumes are mixtures of fragrant material extracts that collectively give a harmonious, pleasant and characteristic fragrance. Each individual component has different chemical and physical properties, making them difficult to stabilize in complex media of liquid detergents due to the interaction of the different chemical groups present in their molecules. Detergents and fabric softeners have perfume in their formulation, but only a very small percentage of it is really deposited on the fabric fibres, with the rest of it being wasted to the drain. To improve its stability in the aggressive detergent media and its deposition on the fabric fibres, perfume is required to be encapsulated.

Microencapsulation is a technique by which one material (normally active) is coated with another material or system, yielding capsules ranging from less than one micron to one millimetre in size. The technology has been widely used to encapsulate a large variety of materials, including inks (Wang *et al.*, 2008), agrochemicals (Martin *et al.*, 2010), flavours (Milanovic *et al.*, 2010), drugs (Galbiati *et al.*, 2011), phase change materials (Li *et al.*, 2011) and adhesives (Minami *et al.*, 2008), and new applications are found increasingly due to the development of new processes and the improvement of wall materials. The main purposes of using microcapsules are to isolate incompatible substances present in the same formulation and to control the release of the active ingredient encapsulated. This release can be due to the diffusion of the active through the wall material (sustained release over time), or it can be due to the breakage of the wall capsule (fast release). There are several ways to trigger the rupture of the shell by changing some of their environmental conditions, *e.g.*. chemical (pH or ionic strength) or physical (external light intensity or stress), and each of them is suitable for different final microcapsule uses.

Perfume microcapsules are solid particles with liquid cores, and they get entrapped within the fabric fibres providing a much more efficient use of perfume. Using perfume microcapsules it is possible to highly reduce the amount of perfume used in a formulation for the same final performance, saving money, chemicals and minimizing possible adverse effects caused by the perfume wasted when it is discharged to the environment (perfumes in high concentrations are usually harmful to water life for example). The use of perfume microcapsules also improves the lasting life of perfume on the cloths, making possible to release it much more slowly and keeping cloths fresher for much longer time. Microcapsules provide the option of releasing the perfume in the place and at the time where it is desired.

Microcapsules can be made using several techniques. They can be classified as chemical or physical methods, depending on the nature of the process used, but there are some methods that are based on both of them. Chemical methods are the ones in which a chemical reaction forms a solid shell surrounding the active (*in-situ* polymerisation, interfacial polymerisation, coacervation) while physical methods are the ones in which a change on a physical property created the shell (solvent evaporation, spray drying, fluid bed coating). Each of the techniques works with different materials and provides different properties to the final microcapsules. Perfume microcapsules should ideally be impermeable and have desirable mechanical properties and to date in-situ polymerisation and interfacial polymerisation techniques are the ones usually selected (Su *et al.*, 2006) to create shells with these properties to a certain extent. *In-situ* polymerisation is the technology that is currently in use in the market for encapsulating perfumes for detergents, but it requires the use of aldehydes (usually formaldehyde) to cross-link the polymer walls to improve their mechanical properties and to reduce their perfume permeability. However, formaldehyde is known to be carcinogenic and its concentration in

final products is highly regulated by law (Sumiga *et al.*, 2011), therefore an alternative formaldehyde-free technology is required. Interfacial polymerisation can be this alternative.

Interfacial polymerisation is a microencapsulation technique that has been in use for the last 50 years. It is based on the reaction between two monomers, each of them dissolved in a phase immiscible with the other, when both monomers meet at the interface they react and form a polymer. If an emulsion is created prior to adding one of the monomers in the system, the interface between the two immiscible phases will be the surface surrounding a droplet of the active and the reaction between the two monomers will make a polymer that will condensate on it quickly creating a microcapsule. The main advantages of this method in comparison with *in-situ* polymerisation process are that no aldehyde is used in the reaction (environmental regulation about aldehyde contents in final products is getting more and more strict) and that depending on the phases and methodology selected it is possible to encapsulate also water soluble actives, which is interesting to industry. On the other hand, microcapsules prepared with interfacial polymerisation techniques in the past had worse permeability and mechanical properties than the ones made with *in-situ* polymerisation. New perfume microcapsules based on interfacial polymerisation techniques need to be developed.

As indicated before it is also possible to use interfacial polymerisation techniques to encapsulate water soluble molecules and glycerol has been selected as a model active in this project. Glycerol is a basic chemical product used in many fields including in personal care products because of its humectant properties. Encapsulation of glycerol may help to improve its stability in such products, *e.g.* in lipsticks, leading to better products. For such application, the polymer shell should prevent glycerol from interacting with the rest of the components in

the product formulation and it should break to release the glycerol on the lips when lipstick is used on them.

Once glycerol microcapsules are developed it will be possible to study their use in other industrial applications, as glycerol is used in many fields (like food industry, personal and oral care products, pharmaceuticals and as chemical precursor of alkyd resins) and the increase in the production of biodiesel (from which glycerol is a by-product) over last years has provoked a huge increase in the production of glycerol, enabling a very cheap supply of it.

The aim of this work was to study the effect of the interfacial polymerisation process conditions on the final properties of the microcapsules produced and to produce microcapsules with the properties required for their use in industrial products. The two actives selected were: perfume for using the microcapsules in detergent formulations and glycerol for using the microcapsules in lipsticks.

A patent has been filled in European Patent Office with Number 10196327.0, on 21 December 2010, (pending to be granted) with the results obtained from this work.

An outline of this thesis is described below:

Chapter 2 describes an overview of the first active of interest: perfume, and it discusses its chemical properties and how they influence the encapsulation process. A general review of encapsulation processes is also described in this chapter. One of them, Interfacial Polymerisation, is selected to work with and it is described in detail including a review of the literature (historical development of the technique and mechanism of capsule formation) and

previous studies on perfume encapsulation. Due to the importance of emulsion formation in the process a section for describing the different types of emulsions, their stability and the different types of emulsifiers is provided. Finally a brief description of the different types of microscopy techniques available for characterising microcapsules and their limitations is presented.

Chapter 3 describes in detail the materials, techniques and experimental procedures used to produce and analyse the perfume microcapsules. This include a description of the perfume and the monomers used in the encapsulation process, the encapsulation process itself and the techniques used to determine: reactivity of the monomers with the perfume, reaction kinetics, leakage of perfume from the microcapsules, mechanical characterisation of single microcapsules, morphological and structural characterisation, particle size measurement and analysis of the wall chemistry.

Chapter 4 describes the microencapsulation of perfume using polyamide walls as well as some preliminary work done using polyester walls. The justification of the different experimental conditions used during the process is discussed. Results on particle size and size distribution are presented and the influence of stirring rate is illustrated and data fitted to theoretical models. The effect of different parameters, like temperature of reaction, surfactant concentration and aqueous monomer used, on the reaction kinetics is also studied. Finally some wall properties (chemistry and thickness) were measured and results using different experimental conditions were compared.

Chapter 5 describes the characterisation of the encapsulation process and properties of microcapsules relevant to potential applications: the loading and encapsulation efficiencies, the leakage of perfume from the microcapsules and the mechanical properties of single microcapsules (including a study of the viscoelasticity of the polyamide walls). The effect of the different experimental conditions on each of the properties is presented. At the end the formulation with the best properties is selected.

Chapter 6 describes the microencapsulation of glycerol with polyamide walls. This chapter includes an overview of the second active ingredient chosen: glycerol, a review of glycerol encapsulation and emulsion stabilization, the description of the process used to produce glycerol microcapsules and the analysis methods used in their characterisation. Some results on size distribution, stability of the microcapsules formed and encapsulation efficiency of 5 different formulations are also discussed. The formulations were made with different aqueous monomers, stirring rates and reaction temperatures.

Chapter 7 summarises the general conclusions and proposes recommendations for further development of the encapsulation processes.

CHAPTER 2:

LITERATURE REVIEW

Learn all you can from the mistakes of others.

You won't have time to make them all yourself.

Alfred Sheinwold

Summary

Nowadays perfumes are present in many daily articles. Their use in laundry detergents is very appreciated by customers, but presents a challenge to the manufacturers of detergents due to the low deposition of the perfume on the fabrics. Different technologies have been developed to improve their performance and the use of perfume microcapsules has demonstrated good results.

The most common microencapsulation methods are briefly described in this chapter. Interfacial polymerisation technique has been selected and a description of the procedure, including the mechanism of capsule formation, is presented. The main factors that influence the final properties of the capsules are identified.

The first step to prepare microcapsules using interfacial polymerisation techniques is to prepare a good emulsion, the emulsion formulation and stability is also discussed.

Different microscopy methods used to study the morphology, shape and size of the capsules and the thickness of the capsule wall are also presented.

2.1. Introduction

Humans have used perfumes and fragrance substances since the early days, at the beginning only for ceremonial purposes, but in our days these substances are available for everyday use and customers require their inclusion in the formulation of all type of products, from papers and inks to foods. But it is in the cleaning industry where the addition of odorants has a

capital importance as customers choose a product not only based on its cleaning effect but also on the smell that it leaves on the cloths or surfaces after use.

Perfumes are mixtures of different compounds with very diverse chemical groups and detergents are aggressive media that tend to interact with perfumes in the formulation. Besides, the main problem of the addition of free perfume to laundry detergents is that most of the perfume added is lost during the wash. During the last years many systems have been developed to prevent this interaction and to improve the deposition of perfume on the clothes (Aussant *et al.*, 2005). Encapsulating the perfume is one of the systems currently in use.

The current technology used to encapsulate perfume for detergents is *in-situ* polymerisation. But capsules formed with this process require the use of an aldehyde (usually formaldehyde) to cross-link the capsule walls and obtain good-quality microcapsules. Formaldehyde is a known carcinogenic chemical and when forming part of a polymer tends to dissociate with time and be released (Su *et at.*, 2006), which make it almost impossible to completely remove it from the formulation. The legislation on formaldehyde permitted concentration in final products is getting more and more strict (Sumiga *et al.*, 2011), therefore a new formaldehyde-free perfume microcapsule needs to be developed.

Interfacial polymerisation technique has been selected to make perfume microcapsules. This technique is well known and it has been successfully used to encapsulate agrochemicals (Hashemi and Zandi, 2001), oils (Soto-Portas *et al.*, 2003), flame retardants (Saihi *et al.*, 2006) and phase change materials (Su *et al.*, 2006).

2.2. Perfume

2.2.1. Introduction

Perfumes are mixtures of fragrant material extracts that collectively give a harmonious, pleasant and characteristic fragrance.

Humans have used mixtures of fragrance substances in the form of incense and perfume unguents for ceremonial purposes since the early days (Schreiber, 2005). Greeks and Romans used new materials and converted perfumes in luxury items and Arabs introduced the distillation, the procedure most commonly used still today to extract perfume oils from natural substances. The art of perfumery prospered during the Renaissance in Italy expanded to France in the XVI century, from that moment French perfumery has held a dominant position in Europe. At the end of the XIX century the first synthetic fragrance substances were produced and the modern perfumery began. The importance of perfumery has greatly increased since then and fragrances have begun to be used as ingredients in many products, like cosmetics, toiletries, soaps and household preparations, not only in fine products (perfumes and eau de cologne). In addition to French perfumery, US perfumery has become very important in the last years also.

2.2.2. Fragrant sources

Fragrance substances can be extracted from natural sources: plants and animals or chemically synthesised (Fahlbusch *et al.*, 2010; Surburg, 2006).

2.2.2.1. Plant sources

Plants have been widely used in perfumery as a source of fragrance oils and aroma compounds. These aromatics are usually secondary metabolites produced by plants to provide protection against herbivores, infections, as well as to attract pollinators. Plants are by far the largest source of fragrant compounds used in perfumery. The sources of these compounds may be derived from various parts of a plant. A plant can offer more than one source of aromatics, for example orange leaves, blossoms, and fruit zest are the respective sources of petitgrain, neroli, and orange oils.

The main plant sources are:

- Bark: such us cinnamon and cascarilla.
- Flowers and blossoms: The largest source of aromatics. The more used are: rose, jasmine, neroli, osmanthus, plumeria, mimosa, tuberose, narcissus, scented geranium, cassie, ambrette, vanilla, clove as well as the blossoms of citrus and ylang-ylang trees.
- Fruits: such as anise, coriander, caraway, cumin, litsea cubeba and juniper berry.
- Peel of citrus fruits: such as lemon, lime, orange and bergamot.
- Seeds: such as mace, angelica, celery, cardamom, tonka bean, carrot seed, coriander, caraway, cocoa, nutmeg and anise.
- Leaves and twigs: such as geranium, patchouli, petitgrain, lavender leaf, sage, violets, rosemary, citrus, hay and tomato leaf, spruce, fir and pine.
- Roots, rhizomes and bulbs: such as iris rhizomes, vetiver roots, rhizomes of the ginger family, angelica and costus.

- Woods: Very important to provide the base note to the perfume. Commonly used woods include sandalwood, rosewood, agarwood, birch, cedar, juniper, guaiac and pine.
- Resins: Valued since antiquity, resins have been widely used in incense and perfumery. Commonly used resins in perfumery include labdanum, frankincense/olibanum, myrrh, Peru balsam, gum benzoin, fir, galbanum, elemi, opopanax and pine.
- Herbs and grasses: such as tarragon, lemongrass, sage and thyme.

2.2.2.2. Animal origin

- Ambergris: It is a metabolic product excreted by sperm whales.
- Musk: It is a glandular secretion of a hornless deer in Central Asia.
- Civet: It is a glandular secretion of the civet cat.
- Castoreum: It is a glandular secretion of the beaver.
- Hyraceum: It is the petrified excrement of the rock hyrax.
- Honeycomb: Extracted from the honeycomb of the honeybee.

2.2.2.3. Other natural sources

- Lichens: Such as oakmoss and treemoss thalli.
- Seaweed: Distillates of some seaweeds, like Fucus vesiculosus, are rarely used due to their high cost and low potency.

2.2.2.4. Synthetic sources

Several semisynthetic products are obtained by chemical modification of a natural starting material. They include hydroxycitronellal from citronellal, citronellol from geraniol or citronellal, geranyl acetate from geraniol, and ionones and methylionones from citral.

Purely synthetic fragrance substances are produced from basic chemicals by complete synthesis. They can be divided into products that are identical to natural ones and products that do not occur in nature. Products identical to natural substances but obtained by chemical synthesis include benzyl acetate from toluene, phenethyl alcohol from benzene, menthol from thymol, and linalool, a product of acetylene synthesis. Other synthetic aroma chemicals have molecular structures completely different from those of natural products. They can be produced only by chemical synthesis, and often imitate the olfactory impressions of natural raw materials. Examples are 4-tertbutylcyclohexyl acetate (woody note, violet note), α-amylcinnamaldehyde (jasmine), 4-tert-butyl-α-methylhydrocinnamaldehyde (cyclamen), musk ketone (musk), and ethylene brassylate (musk). Many natural products will continue to be indispensable in perfumery. However, synthetic products are playing an increasingly important role in the perfumer’s assortment of raw materials because of their virtually unrestricted availability, constant quality, and generally steady price.

The majority of the world's synthetic aromatics are created by relatively few companies. They include: International Flavors and Fragrances (IFF), Givaudan, Firmenich, Takasago and Symrise. Each of these companies patents several processes for the production of aromatic synthetics annually.

The global market for flavours and fragrances was valued at US$ 12.6 billion in 2006 (IAL Consultants, 2007), from which slightly less than 50% correspond to fragrances ($6,224 million). From those, almost half ($3,075 million) correspond to the market of soap, detergents, household cleaners and air fresheners.

Last estimations of the sale values of the flavour and fragrance industry leaders are a bit higher, around $22 billion in 2010 (Leffingwell & Associates, 2011).

2.2.3. Perfume formulation.

2.2.3.1. Perfume notes

To prepare a perfume formulation different fragrant essences are mixed (Mata *et al.*, 2005; Schreiber, 2005). Due to the different odours and evaporation rates of the different essences the final perfume will have a designed odour that usually is not constant with time. More volatile compounds evaporate before showing their odour in the first moments while less volatile compounds will start smelling later. Depending on their evaporation rates, fragrance essences are divided into:

- Top notes: The scents that are perceived immediately after application of the perfume. They are small molecules that evaporate quickly. Green and citrus scents are in this group.
- Middle notes: The scents that emerge when the top notes dissipate. Most of the floral scents are in this group.
- Base notes: The scents perceived after the middle notes dissipate. They are usually not perceived until 30min of the perfume application. They are complex compounds with a low volatility. Woody and musky scents are in this group.

When different essences are mixed they don't behave as the pure essence, they are influenced by the rest of the essences in the formulation and interact with each other. In this way, the essences in the top and middle notes are influenced by the base notes and the base notes scents are modified by the middle notes essences present in the formulation.

2.2.3.2. ClogP

Another important factor in the formulation of a perfume is the hydrophobic or hydrophilic character of the essences present in it (Fahlbush *el al.*, 2010), especially if the perfume is going to be used inside another product like soaps, detergents and other household preparations. This character is measured by the ClogP ("calculated" logP), or logarithm of the partition coefficient of each essence in a mixture of two immiscible solvents at equilibrium. The system used is 1-octanol/water. The partition coefficient is the ratio of concentrations of the un-ionized compound between the two phases and the logarithm of this partition coefficient is called logP. Negative values of the ClogP mean that the compound is hydrophilic while positive values mean that the compound is hydrophobic.

2.2.3.3. Fixatives and solvents

Fixatives are used to equalize the vapour pressures (Schreiber, 2005), and thus the volatilities, of the raw materials in a perfume oil, as well as to increase the tenacity. Natural fixatives are resinoids (benzoin, labdanum, myrrh, olibanum, storax, and tolu balsam) and animal products (ambergris, castoreum, musk, and civet). Synthetic fixatives include substances of low volatility (cyclopentadecanolide, ambroxide, benzyl salicylate) and virtually odorless solvents with very low vapour pressures (benzyl benzoate, diethyl phthalate, triethyl citrate).

The only solvent used in fine perfumery is extremely pure ethanol that is diluted with water to the required concentration. Weakly odorous synthetic fixatives are used as solvents for alcohol-free perfumes, especially bazaar oils.

2.2.3.4. Perfumes for cosmetics, toiletries and household products

In cosmetics, toiletries, and household products, the perfume is usually of secondary importance for the effectiveness of the product; but it may, however, strongly influence the consumer's decision to buy a product and, in many cases, represents the only way of making the product's action perceptible to the consumer.

Although the model fragrance types are often created in the field of fine perfumery, chemical and physical aspects must also be taken into account in the development of perfume oils for toiletries and household products. For example, the perfume oil used in creams and white soaps must not cause discoloration; a perfume oil for aftershave lotion must be soluble in 50 – 60% alcohol; the fragrance used in a powdered detergent must be alkali resistant; and a fabric softener is expected to leave clothes with a pleasant odour; and even a household cleanser must have a pleasant and functional odour, although active chlorine places extraordinary demands on the stability of the perfume oil. Minimal perfume doses are also expected to adequately mask the often strong and unpleasant doors of products such as insecticides, floor cleansers, paints, and varnishes.

Fragrances for soaps must satisfy the following requirements: chemical resistance, low volatility of raw materials (*e.g.* geraniol and ionone), strength of odour in soap (*e.g.* citronellal and γ-decalactone), and adhesion to the skin (*e.g.* 1,2-furano-2,5,5,8a-tetramethyldecalin and

α-amylcinnamaldehyde). The development of perfume oils for this wide-ranging field requires special knowledge of perfumery, as well as information about the chemical and thermal stability of the perfumery materials used and the product being perfumed.

In the case of laundry detergents and fabric softeners the main problem of the use of perfume is that most of the perfume present in the formulation is wasted and leaves the washing machine with the washing water, only a very small percentage of the free perfume in the detergent is present on the cloths after washing, and most of it is evaporated during their drying. Research has been done and several technologies have been developed to increase the amount of perfume on the clothes and the effective time of the perfume on them. Some technologies use molecules that bind to the clothes on one region and to a molecule of perfume on other, increasing the "useful" part of the perfume. These molecules also decrease the volatility of the perfume and the odour lasts for longer time on the clothes. However the perfume has still a relative short life on the fabrics. The other technology that it is being used to increase the perfume deposition on fabrics and the life of the perfume is the microencapsulation of the perfume.

2.3. Microencapsulation

Microencapsulation (Thies, 1999) is a technique by which one material or mixture of them is coated with or entrapped within another material or system on a very small scale, yielding capsules ranging from less than one micron to one millimetre in size. Microcapsules are minute containers, normally spherical if enclosing a fluid, and have roughly the shape of the material which is encapsulated.

The substance that is encapsulated may be called the core material, the active ingredient or agent, fill, payload, nucleus or internal phase. The material encapsulating the core is referred to as the coating, membrane, shell, carrier, encapsulant or wall material.

Microcapsules have been widely used to encapsulate a large variety of materials including inks, agrochemicals, flavours, drugs, phase change materials and adhesives (Thies, 1999).

The main applications of encapsulation are:

- Protection of the active component from climatic effects and external damage (improving storage life).
- Conversion of a fluid active component (liquid or gas) into a dry "solid" system.
- Separation of incompatible components for functional reasons.
- Masking of the undesired properties of the active component.
- Controlled release of active components for delayed (time) release or long-acting (sustained) release, under influence of heat, pH, ionic strength, submersion in fluid, osmotic rupture, mechanical force, or any other possible mechanism.

When considering microencapsulation of a product required for a novel system, it is helpful to examine the following criteria (Thies, 1994):

- The characteristics of the active component and core medium of the product to be encapsulated.
- The performance required from the encapsulated product.
- The acceptable manufacturing cost for the microencapsulated product.

From these criteria, one can then consider the factors which may affect the choice of coating to be used for the capsule wall. The most important of these are:

- Ability of the coating to form films of appropriate physical properties with a suitable and convenient wall thickness.
- Appropriate chemical, physical and physicochemical properties of the coating to allow the use of convenient and appropriate methods for economical microcapsule production.
- Possibility of the surface hardening of the formed capsule wall surface, to give hardened microcapsules with a non-tacky surface, so that the microcapsules behave as a free-flowing fluid.

Factors such as those outlined above must be considered in relation to the choice of microencapsulation process. Factors relevant to the choice of wall material include the:

- Elasticity and mechancial strength of the wall film (capsules should be able to tolerate handling, but, for many applications, should rupture above a predeterminated pressure)
- Permeability of the wall film (which affects the economics and the storage life of the product)
- Melting point and glass transition temperature of the wall material (both factors will affect manufacturing conditions)
- Degradation properties of the wall material.
- Concentration and temperature range for which the coating material is sticky or tacky.

Many different methods for encapsulation are available, each one suitable for different applications and core materials and leading to different capsule properties. These methods are usually categorized into two groups: chemical methods and mechanical or physical methods (Thies, 1994; Thies, 2005).

2.3.1. Chemical methods

2.3.1.1. Coacervation

Coacervation consists of the separation from solution of colloidal particles which then agglomerate into separate liquid phase called coacervate. Coacervation consists of three stages: dispersion of the active material to be coated into an aqueous solution of a polyelectrolyte, deposition around the core material of coacervate formed by addition of an aqueous solution of another polyelectrolyte with opposite charge, and gelation of the coacervate.

Coacervation can be simple or complex. Simple coacervation involves only one type of polymer with the addition of strongly hydrophilic agents to the colloidal solution. For complex coacervation, two or more types of polymers are used.

Generally the core material used in the coacervation must be compatible with the recipient polymer and be insoluble in the coacervation medium.

The advantages of this method are:

- Excellent for coating hydrophobic liquids in small capsules.
- Highly developed process (widely used for carbonless copy paper)

- Forms excellent barriers.
- Fairly uniform coatings on irregular particles.
- Can form very thin, resistant good walls (<1μm)

The limitations of this method are:

- Core material must be insoluble in water.
- All coatings are based on gelatine chemistry.
- Aldehydes (used for crosslinking) can cause a concern.
- Coating may not wet the core particle.
- Particles often aggregate during processing.
- Process may take 16-24h and requires careful control.
- Expensive process.

2.3.1.2. Interfacial polymerization (IFP)

A multifunctional monomer is dissolved in the core material, and this solution is dispersed in an aqueous phase. A reactant is added to the aqueous phase, and polymerization of the reactant and monomer quickly takes place near the interface of the core droplets and the aqueous phase, forming the capsule walls.

The advantages of this method are:

- Excellent for coating hydrophobic liquids.
- Forms relatively elastic walls.
- Can coat droplets down to a few μm
- Tough walls for particles <50μm

- Can coat aqueous drops, with reversed phases.
- Simple process and fast.
- Has been scaled to large production.
- Inexpensive process, used for pesticides.
- Encapsulation process carried out at low temperature.

The limitations of this method are:

- Cannot coat solid particles (only if they are dispersed in a liquid phase).
- Polar liquids often cause trouble.
- Cores cannot be pH-sensitive.
- Walls are thin (1-2μm), not good diffusion barriers.
- Microcapsules >200-300μm are weak.
- Coating aqueous drops is much more difficult.
- Crosslinked walls: release is by pressure and shear.
- Monomers can react with the core material.

2.3.1.3. In situ polymerization

It is similar to interfacial polymerisation. The distinguishing characteristic of in situ polymerization is that no reactants are included in the core material. All polymerization occurs in the continuous phase, rather than at the interface of the continuous phase and the core material, as in IFP. Examples of microcapsules produced using this method include urea-formaldehyde (UF) and melamine-formaldehyde (MF).

The advantages of this method are:

- Smooth and thick walls formed.
- Excellent for coating hydrophobic liquids.
- Very simple process.

The limitations of the process are:

- Cannot encapsulate water soluble actives.
- Aldehydes (used for crosslinking) can cause a problem.

2.3.2. Physical methods

2.3.2.1. Spray drying

An emulsion/slurry is prepared by dispersing the core material, usually an oil or immiscible with water, into a concentrated solution of wall material until the desired size of oil droplets are attained. For the former case, the resultant emulsion is atomized into a spray of droplets by pumping the emulsion through a rotatory disc into the heated compartment of a spray dryer, where the water portion of the emulsion is evaporated, yielding dried capsules of variable shape containing scattered drops of core material.

The advantages of this method are:

- Fast drying of thermally-sensitive materials.
- Can handle a great variety of materials.
- Can form particles from 10μm.
- Very high production rates: up to 150,000 ton/year

- Equipment ready available.
- Low processing cost.

The limitations of this method are:

- Forms only matrix particles, not capsules.
- No thick protective wall.
- Wide size distribution of particles.
- Drying particles >100μm can require large equipment due to short residence time in the spray drier chamber.
- Loading of liquid cores often <30%.
- Suspended solids must be small.
- Possible dust explosion hazard.
- Loss of core material due to volatilization and/or degradation.

2.3.2.2. Fluid bed coating

Solid particles to be encapsulated are suspended on a jet of air and then covered by a spray of liquid coating material. The capsules are then moved to an area where their shells are solidified by cooling or solvent evaporation. The process of suspending, spraying and cooling is repeated until the capsules' walls are of the desired thickness.

The advantages of this method are:

- Excellent for irregular particles.
- Forms uniform walls.
- Works well down to 100-150μm.

- Coats cores which would dissolve in the coating.
- Forms excellent barriers from polymeric solutions.
- Can apply melted coatings.
- Thin coatings can be applied.
- Very thick walls are possible.
- Good production rates: 1,000-5,000ton/year
- Low cost.

The limitations are:

- Coats only solid core particles.
- Particles <100μm tend to aggregate.
- Coating viscosity should be <100-500cp
- Walls <15μm may have pinholes.
- Melted coatings difficult on small core particles.
- Suspended solids in coating may cause plugging.

2.3.2.3. Spay cooling/chilling

Spray cooling/chilling strictly speaking is not a true microencapsulation process, because a significant amount of the active material is located at the surface of the microcapsule or has direct access to the environment. The core material is dispersed in a liquefied coating or wall material, which is atomized. There is emulsification of the active compounds into molten wall materials, followed by atomization to disperse droplets from the feedstock. After that the

droplets are immediately mixed with a cooling medium and subsequently solidify into powder.

The advantages of this method are:

- Can handle solutions, suspensions, emulsions, …
- Can form particles up to 800 μm.
- Particles often spherical.
- Very high production rates.
- Low capital cost.
- Extremely low processing cost.

The limitations of this method are:

- Suspended solids must be small.
- No thick protective wall.
- Low viscosity needed for particles <30μm.
- Wide size distribution of product.
- All lines must be heat-traced.

2.3.2.4. Centrifugal extrusion processes

The core and the shell materials, which should be immiscible with each other, are pushed through a spinning two-fluid nozzle. This movement forms an unbroken rope which naturally splits into round droplets directly after clearing the nozzle. The continuous walls of these

droplets are solidified either by cooling or by a gelling bath. Generally capsules of a larger size, from 250 microns up to a few millimetres in diameter are produced.

2.3.2.5. Spinning disk

The internal phase is dispersed into the liquid wall material and the mixture is advanced onto a turning disk. Droplets of pure shell material are thrown off the rim of the disk along with discrete particles of core material enclosed in a skin of shell material. After having been solidified by cooling, the microcapsules are collected separately from the particles of shell material.

The advantages of this method are:

- Coats particles from 20μm to several mm.
- Excellent for melting coatings.
- Fast process: 10-30s, on disk 0.1-0.2s
- Coating viscosity can be >5,000cp (for large cores)
- Product size distribution same as for core particles.
- Coating can contain many suspended solids.
- Very high production rates.: up to 320,000ton/year
- Very low cost.

The limitations of this method are:

- Irregular core particles not ideal
    - Protrusions lead to thin spots in coating.

  - Extra coating required to fill “valleys”.
  - Best to granulate first, then coat.
- Cannot coat core particles <20μm
- Maximum coating thickness: 200-400μm (melts).
- Core particles are sometime off-centre.
- Volatile organic solvents lead to coating imperfections.

From the microencapsulation processes available only the interfacial polymerisation and the in situ polymerisation ones can provide a polymeric capsule wall with low permeability and the required properties to survive the handling and washing process but weak enough to break during the normal use of the cloths to release the perfume. There is a huge patent history in making melamine-formaldehyde encapsulation of perfume using in situ polymerisation and this is the current process used in many industrial applications, but as mentioned before this process requires aldehyde (formaldehyde) as a crosslinking agent and this compound has limitations if it is incorporated in final products because of environmental and health concerns. So far capsules produced by interfacial polymerisation (Danicher *et al.*, 1999; Kim and Park, 2007; Chu *et al.*, 2001) are more porous than those based on melamine-formaldehyde produced by in situ polymerisation (Hwang *et al.*, 2006; Su *et al.*, 2006b; Hong and Park, 1999), the latter of which have more smooth and compact walls, so the aim of this project is to develop new perfume microcapsules made by interfacial polymerisation in order to make them suitable to use in liquid detergents.

2.4. Interfacial polymerisation

2.4.1. Introduction

The use of polycondensation (or polymerisation) reactions to produce polymers of industrial interest started at the end of 1920s, when research was developed in DuPont laboratories. The first product of interest developed there was nylon in 1934 (Carothers, 1938), a polyamide polymer produced by the reaction of a diamine and a dicarboxylic acid. Nylon was a substitute of natural fibres like cotton and acquired high importance after World War II when it was extensively used by the American army in the confection of uniforms and parachutes.

The process to make nylon involved the condensation reaction between two different monomers at high temperatures (above 200°C) and in vacuum to obtain high molecular weight polymers (also called superpolymers). However working in those conditions were not easy and many monomers were not stable in them. This limited the possible use of different monomers to produce new polymers.

In 1951 a new method to produce these superpolymers at lower temperatures using high reactive monomers which react at the interface of two liquid phases in a system was developed (Magat *et al.*, 1955). In 1958, at the Meeting of the American Chemical Society in Chicago, P.W. Morgan described the principles of the Interfacial Polycondensation (Morgan and Wittbecker, 1959). At that same meeting, the preparation of different polymers using this method, for example: polyamides (Beaman *et al.*, 1959), polyurethanes (Wittbecker *et al.*, 1959) or polyphthalamides (Katz, 1959) was reported. In 1953, a method to encapsulate ink for carbonless copy paper using coacervation was developed (Green *et al.*, 1957) and

produced the first liquid filled microcapsules, which opened a new avenue to make microcapsules.

In 1964, Mackinney filed the first patent that describes the use of an interfacial polycondensation reaction between two different monomers for the formation of microcapsules. The process he used consisted of dissolving one of the monomers (acid dichloride or diisocyanate) in a water immiscible solvent, and another monomer (diamine or glycol) in water. The active to be encapsulated would be dissolved in one of the two phases, depending on its solubility, although he only gave examples of encapsulating organic soluble actives and fine solids suspended in the organic phase. An emulsion was then formed and the two monomers started reacting.

In 1969 Ruus filed a patent in which he made microcapsules by interfacial polymerisation following the following method: one monomer and the active material to be encapsulated are dissolved in a solvent and then they all are emulsified in an immiscible phase. When the emulsion is produced, additional immiscible phase containing the complementary monomer is added, and the reaction then begins. With this change in the process it was possible to control the capsule size much better and obtain more homogeneous capsules. He mainly worked with diacid chlorides, di- or poly- amines, and polyols.

In 1971 Vandegaer filled a patent very similar to Ruus' one. Vandegaer used the same method as Ruus to make microcapsules although he gave much more information about emulsifying agents and monomers. He made microcapsules with cross-linked shells to give them more resistance and barrier properties. To do that he used at least one polyfunctional monomer

(ideally one in each phase), like polyacid chlorides (trimesoyl chloride), polyols (benzenetriol), polyamines (tetraethylene pentamine, polyethylene imine, melamine). He used the method to encapsulate: pigments and dyes, pharmaceuticals, flavouring or perfuming agents, pesticides, herbicides and peroxides. In addition he developed a continuous process for making microcapsules.

Vandegaer's method to make microcapsules by interfacial polymerisation was adopted by industry (mainly agrochemical industry) and it did not change very much from then. The main change has been the improvement of the method to use it to encapsulate water soluble actives or suspensions of solids in water. Vandegaer spoke about that possibility but he only worked with inks and oils so that all the chemicals he used were suitable for working in the encapsulation of oils but not in these new conditions. The emulsifiers he used were not adequate to stabilize water-in-oil emulsions so that new emulsifiers had to be studied.

Other changes in the procedure were the formation of microcapsules with a polyamide shell and a polyurethane-polyurea-structured inner mass, where both polymers were formed during the encapsulation process (Heinrich *et al.*, 1983) to improve the mechanical resistance of the microcapsules, and the use of a solvent evaporation method with supercritical CO_2 (Benoit *et al.*, 2001). In addition new monomers have been studied as reactants (Argillier *et al.*, 2003; 2004).

The primary use of the interfacial polymerisation technique has been to encapsulate agrochemicals and their encapsulation with new emulsifiers has been reported by several

authors (Beestman, 1981, 1985 and 1994; Nesbitt *et al.*, 1984; Becher and Magin, 1986; and Benoff and Dexter, 1999).

With all these works done inside companies with patent protection, academic research has focus on understanding of the reaction and controlling all the parameters that affect the final properties of the capsules. There are many interesting works from which the following ones need to be mentioned:

1. Arshady (1989) wrote a review in which he compiled all the information about interfacial polymerisation technique previously obtained, discussed the theoretic mechanism of capsule formation and highlighted the main factors that affect the final properties of the capsules produced. He based his statements on previous work developed by other authors. It is a very complete guide about interfacial polymerisation, and a summary of it is shown in the next section (2.4.2).
2. Mathiowitz and Cohen (1989a and 1989b) carried out a very extensive work with respect to the preparation and characterisation of polyamide microcapsules. They studied different monomer formulations of capsules made at room temperature. For example, they used an organic solvent as core material and obtained capsules of 100-500μm in size and 0.8-3μm thickness. They studied the reaction kinetics, the structure of the polymer formed, its crystallinity and its porosity, and concluded that membranes made with linear polymers were more crystalline, more porous and had a higher degree of swelling than cross-linked ones, which were more stable thermally, revealed less structure from Differential Scanning Calorimetry (DSC) analysis, were less crystalline, swelled less and was less porous in structure. They also made double wall capsules by adding a different aqueous monomer at a later time and found that

they had very good thermal stability. The release properties of the capsules were also investigated and it was found that membranes made of cross-linked polyamides were much less permeable to the core material than membranes made with linear polyamides.

3. Janssen *et al.* (1993) developed mathematical models for capsule wall growth and studied the influence of the process conditions on the wall permeability of polyamide capsules. They produced their capsules using an extrusion method and capsules were big (500-1000μm). They found that an increase in the amine/chloride ratio and the use of diamines, in addition, to the triamines reduced the wall permeability.
4. Toubeli and Kiparissides (1998) studied the effect of the amine type and composition on polytherephthalamide membranes permeability. They produced membranes, not microcapsules, and found that the addition of diamines to the triamines resulted in the reduction of the membrane permeability, which was very pronounced when adding hexamethylenediamine (HMDA).
5. Soto-Portas *et al.* (2003) developed new polyamide microcapsules using Jeffamine as aqueous monomer (they also patented the invention, patent number: US2002158356), and produced capsules of 150μm at 28°C. Liquid paraffin was encapsulated and a capsule loading efficiency of 95% and a yield of 90% were obtained. When they added trifunctional monomers (DETA and Trim) they saw that the capsules had lower porosity and less permeable walls and that if the capsules were dried no paraffin leaked out.
6. Persico (2005) produced a PhD Thesis in which she described encapsulation of jojoba oil with polyamide. She produced capsules of 5-15μm and found that the best capsules she produced were made with HMDA as aqueous monomer and a mixture of TC and

Trim as organic monomers (with a molar ratio of COCl functions TC/Trim=3). Microcapsules made in those conditions had smooth and dense surface and low porosity walls. She found that the use of a cross-linking agent in the organic phase is much more efficient than in the aqueous one. She also found that the organic phase being encapsulated had a big influence on the organic monomers. For example, she managed to encapsulate toluene using an aliphatic acid dichloride, but she was not able to encapsulate jojoba oil with the same aliphatic monomer under the same experimental conditions. She concluded that the system behavior is strictly dependent on the type of organic phase. She also published a paper with her results (Persico et at., 2005).

2.4.2. Mechanism of capsule formation

The interfacial polymerization procedure consists of the reaction, at the interface of an emulsion, between two different monomers, one dissolved in the organic phase and the other one in the aqueous phase. Depending on the nature of these monomers, there are formed different wall polymers, as can be seen in Table 2.1.

Organic Monomer	Aqueous Monomer	Polymeric Shell Wall
Polyacid Chlorides	Polyamines	Polyamide
Polychloroformates	Polyamines	Polyurethane
Polyisocyanates	Polyols	Polyurethane
Polysulphonyl Chlorides	Polyamine	Polysulfonamide
Polyisocyanates	Polyamines	Polyurea
Polyacid Chlorides	Polyols	Polyester
Polychloroformates	Polyols	Polycarbonate

Table 2.1. Polymers produced from the reaction of different monomers. (Adapted from Arshady, 1999)

As it is possible to see in Table 2.1, the formation of polyesters and polyamides involves the reaction of polyacid chloride (-COCl) groups with polyol (-OH) or polyamine (-NH_2) groups respectively. During this reaction a bond is formed (-COC- or –CONC-) and a molecule of HCl is released to the media. The release of HCl provokes a change in the pH of the media which makes it possible to monitor the reaction kinetics with the use of a pH meter.

Arshady (1989) described the fundamentals of interfacial polymerisation reactions. The first step to make capsules by interfacial polymerisation is to prepare an emulsion of droplets with the active of interest in a continuous phase. These droplets will be the templates for the formation of the final microcapsules and the final capsules will have the same size as the original droplets. It is very important to prepare a stable emulsion prior to adding the monomers to start the reaction. Once the two monomers are in the system the reaction starts and the morphology and properties of the capsules formed are largely determined by the solubility of the polymer formed in the inner droplet phase. There can be two extreme situations:

1. The polymer formed is highly soluble in the droplets. Thus the polymer is entrapped inside the droplets. In this case the particles develop gradually into solvent swollen microspheres. A typical example of this situation is that of bisphenol A with phosgene.
2. The polymer formed is highly insoluble in the droplet phase. Thus the polymer precipitates at the interface and deposits on the droplets' surface forming a primary membrane around the droplets. Further polycondensation of monomers increases the thickness of the membrane and produces the final shell. Preparation of nylon microcapsules is a typical example of this case.

The process of capsule formation proceeds in three consecutive stages (Figure 2.1): (1) initial period of polycondensation, (2) formation of the primary membrane around the droplet, and (3) growth of this membrane to the final capsule wall.

1. Initial period of polycondensation: The two monomers meet each other and start reacting forming a precondensate that dissolves there. The reaction rate is usually instantaneous. The place where the two monomers meet will be determined by the partition coefficients of the monomers. Usually the solubility of the organic monomer in the water phase is negligible and the aqueous monomer is partitioned between the two phases. Therefore the initial reaction site is usually in the organic phase.
2. Formation of a primary membrane around the droplets: The process of polymer precipitation and formation of the membrane around the droplets is largely controlled by the solvency (or swelling power) of the medium for the polymer. The higher the solvency of the medium for the polymer is, the thicker and less porous the membrane is expected to be. However it is possible to modify the solubility of the polymer in the medium using additives such as monomers, stabilizers, salts, etc. For example it has been reported (Morgan and Kwolek, 1963) that the presence of quaternary ammonium salts in the polycondensation mixture can increase polymer molecular weights as a result of polymer swelling. But it has also been reported (McGinity *et al.*, 1981) that the same salts can be responsible for no capsule formation. Another factor that has a big influence on the final properties of the capsules is the rate of precipitation of the polymer during the initial polycondensation period. The precondensate precipitates when its concentration reaches a limit (usually very low but depends on the polymer formed and the medium). In general, the higher the rate of precipitation is, the less uniform (more porous and permeable) the membrane is. Precondensate precipitation

(that is, polymer formation) is proportional to the polycondensation rate during the initial polycondensation period. It is, therefore, possible to control the membrane permeability by factors which affect the rate of polycondensation such as monomer concentration, temperature of reaction and addition rate of monomers to the emulsion.

3. Growth of the membrane to the final shell: After the membrane is formed the two monomers are separated from each other and to continue with the polycondensation reaction one of the monomers must cross the membrane to react at the other side with the other monomer. Previous experiments on the formation of nylon and polyester films by interfacial polymerisation (Morgan and Kwolek, 1959) show that the primary membrane is always formed in the organic side, but the growth of the membrane takes place in the organic or aqueous phase depending on the nature of the wall formed: nylon membrane growth takes place in the organic side while polyester membrane growth takes place in the aqueous side. Corresponding to the membrane growing its permeability to the monomer is decreasing, as a result, the polycondensation rate diminishes and the reaction stops.

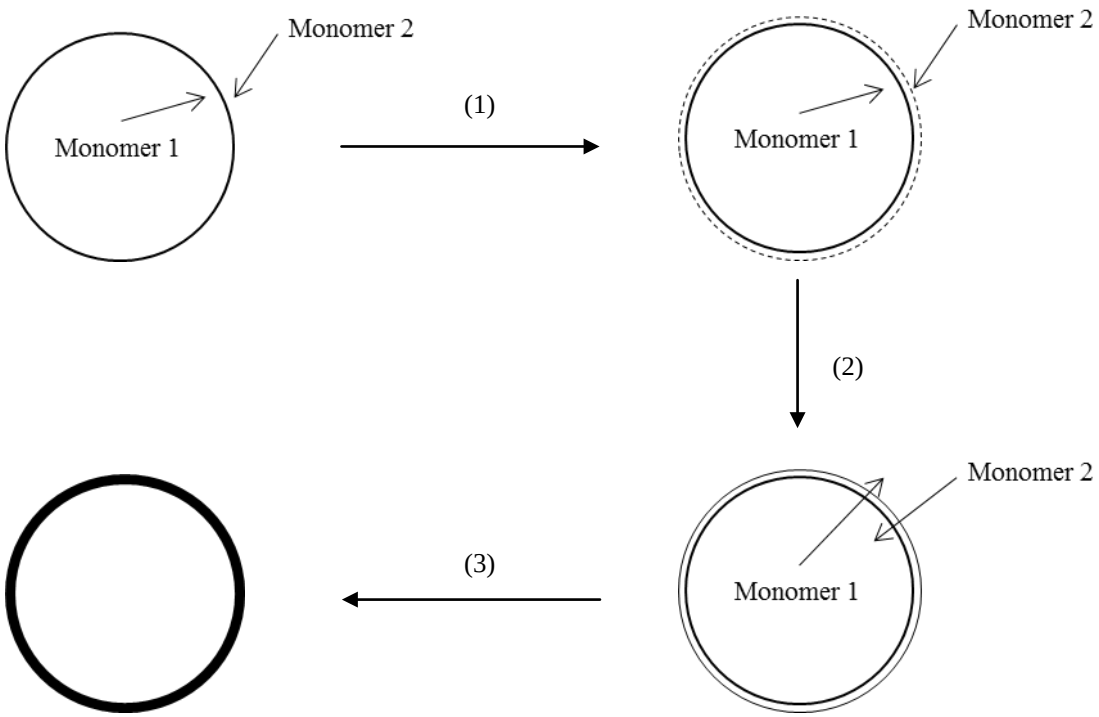


Figure 2.1. Process of capsule formation by Interfacial Polymerisation. (1)Initial period of polycondensation, (2)Formation of a primary membrane around the droplet, (3)Growth of the membrane to the final shell.

Arshady also suggested the main factors that control the course of capsule formation in interfacial polymerization:

- Concentrations and partition coefficients of the monomers.
- Volume ratio of the two phases.
- Type and concentration of additives.
- Type of stirrer and rate of stirring.
- Rate of polymerization and polymer molecular weight.
- Temperature of the polymerization mixture.
- Chemical structure and crystalizability of the polymer.
- Solvency of both phases for the polymer.

From all the factors stated the ones which have a bigger influence on the microcapsule size are the stirring rate and the surfactant concentration. Although the main properties of the microcapsules are given by the polymer formed (controlled by the monomers involved in the reaction), as showed before the properties of the capsules will be related to the initial rate of precipitation of the polymer forming the membrane, and this precipitation rate can be controlled by the temperature of reaction and the availability of aqueous monomer in the solution.

2.5. Perfume microcapsules

After the development of processes to encapsulate organic substances in 1960s, soon it was seen that perfume could be one of the products that could take advantage of the new technique to deliver odours to the right moment and place. Microcapsules of different perfumes were used in scratch and sniff products with a reasonably good success.

Perfume microcapsules made with polyamide walls were also incorporated into detergents to provide better perfume application to the clothes (Brain *et al.*, 1979), but it was difficult to obtain perfume microcapsules that maintain their stability during the storage in such aggressive media. The capsules that present better stability in detergents have been the ones made with aminoplast resins: urea-formaldehyde and mainly melamine-formaldehyde.

Capsules made with aminoplast resins have several advantages like low wall permeability and good mechanical strength, but they contain formaldehyde in the formulation, which is a known carcinogenic product. In the last years the laws have become more and more restrict with the presence of free formaldehyde in final products and companies have been making

efforts to reduce its level using new methods of production of the capsules and by adding new steps to remove this formaldehyde (adding scavengers).

There are currently commercial detergent products with perfume microcapsules in their formulation, *e.g.*, the fabric softeners Comfort (Unilever) and Lenor (Procter & Gamble).

2.6. Emulsions

Perfume microcapsules should have appropriate size, which is strongly determined by the size of the oil droplets since their wall thickness is generally very small (less than 1 μm). Therefore it is crucial to understand how the oil droplet size in an emulsion can be controlled.

2.6.1. Introduction

Emulsions (Shaw, 1992) are dispersions of one liquid in another in a form of droplets where the two phases of the emulsion are immiscible or partially miscible. The droplets are usually between 0.1 and 10μm in diameter. If droplets are smaller (between 0.01 and 0.1μm) they are called nanoemulsions. The liquid forming the droplets is termed internal or dispersed phase while the dispersant is termed continuous or external phase. The process of preparation of an emulsion is termed emulsification and the stabilizers added to the system are termed emulsifiers or surfactants. If the continuous phase consists of water and the internal phase of an organic liquid the term oil-in-water (o/w) emulsion is used. On the other hand if water is dispersed in an organic phase a water-in-oil (w/o) emulsion is produced. It is also possible to form oil-in-oil (o/o) or water-in-water (w/w) emulsions and in some cases oil droplets which hold water inside are observed to be dispersed in water (w/o/w) or the type (o/w/o) has been reported too.

Examples of natural emulsions (Shaw, 1992; Tadros, 2009) are cow's milk (o/w emulsion with 3.5% fat dispersed in the aqueous phase) and butter (w/o emulsion containing up to 20% water dispersed). The practical application of emulsions and emulsion technology is considerable, and includes foodstuffs, pharmaceutical preparations, cosmetics or agricultural sprays. A large volume of technological information on emulsions exists but much of it in private files.

The visual appearance of an emulsion reflects the influence of droplet size on light scattering, and varies from milky-white-opaque, for large droplets (>1μm), through blue-white, then gray-translucent to transparent, for small nanoemulsion droplets (<0.01μm).

2.6.2. Stability

An emulsion is stable if coalescence of the droplets is prevented by a sufficiently high energy barrier (Shaw, 1992). In general, the energy barrier is built up by the film of emulsifier that forms at the surface of the droplets. The uniform dispersion of the droplets in the external phase may be destroyed reversibly by sedimentation or irreversibly by coalescence. Sedimentation (or creaming) results from a density difference between the two phases. Droplets concentrate at the bottom (sedimentation) or the top (creaming) of the emulsion forming aggregates. Although the spatial distribution has been altered, the original dispersion can be restored by shaking or stirring. Sedimentation is not necessarily accompanied by droplet coagulation, although it facilitates the process.

Droplet collision may result in coagulation, which may lead to coalescence to form larger droplets. First, the smaller droplets are absorbed by the larger ones, and eventually the

dispersed phase may become a continuous phase, separated from the dispersion medium by a single interface. Two droplets can only coalesce if the intervening layer of liquid is pierced when they approach each other. Therefore, coalescence is opposed in two ways by the emulsifier film surrounding the droplets. First, the like charges of the electrical double layer prevent them from approaching each other. Second, the build-up of an elastic surface film causes the emulsion droplets to bounce off each other when they collide.

The following factors favour emulsion stability (Shaw, 1992):

1. Low interfacial tension: The adsorption of surfactant at oil-water interfaces causes a lowering of interfacial energy, enhancing the stability of the large interfacial areas associated with emulsions.
2. A mechanically strong and elastic interfacial film: surfactants also form a protective film around the droplets.
3. Electrical double layer repulsions: interparticle repulsion due to similarly charged electric double layers is an important stabilizing mechanism in o/w emulsions.
4. Relatively small volume of dispersed phase.
5. Narrow droplet size distribution
6. High viscosity: A high viscosity retards the rates of creaming or coalescence.

The type of emulsion formed for two given phases will depend mainly on the stability of the emulsifier layer surrounding the droplets. If a water-stable emulsifier envelope is formed around the oil droplets in a system containing water, oil, and emulsifier, an o/w emulsion is produced. On the other hand, the formation of an oil-stable envelope around the water droplets produces a w/o emulsion.

The volume fraction (ϕ) (Eq. 2.1) may also affect the emulsion type.

$$\phi = \frac{V_i}{V_t} \qquad \text{[Eq.2.1]}$$

where:

V_i is the volume of the internal phase
V_t is the total volume

The higher the volume fraction for one of the liquids is, the more likely this liquid is to become the continuous medium. However, the liquid with the lower volume fraction will not always be the dispersed phase if a proper emulsifier is carefully selected, although it will be more difficult to form a stable emulsion with a high volume fraction of dispersed phase.

2.6.3. Emulsifiers

If an emulsion is prepared by homogenising two pure liquids, phase separation will usually be rapid, especially if the concentration of dispersed phase is high. To prepare reasonably stable emulsions an emulsifier must be present.

Emulsifiers (Tadros, 2009) are molecules that accumulate at the interface between the dispersed phase and the continuous phase. Their hydrophilic groups project into the water and their hydrophobic groups project into the organic phase. This produces an interface film, which becomes more extensive and adsorbs additional emulsifier molecules if the droplets are broken up mechanically, until all the droplets are enveloped by a surface film.

A classification of emulsifiers can be done regarding to their surface activity (Tadros, 2009):

1. Inorganic emulsifiers: They are fine powders with low surface activity. Bentonite, fuller's earth, china clay and activated carbon are in this group. Depending on the hydrophobicity of the surface they are good to prepare o/w or w/o emulsions.
2. Natural emulsifiers: They are natural products with little or no surface activity. Most of them are added to the emulsions as protective colloids or to increase the viscosity, and consequently the stability of the emulsion (mainly when solids are present in the system). They can also be added to give the emulsion certain mechanical, optical or electrical properties, including proteins, polysaccharides, waxes or rubbers like celluloses, carboxylic acids, ligninsulfonates, gelatine or starch.
3. Surface active emulsifiers: All their molecules consist of a section that is hydrophobic (lipophilic) and sparingly soluble in the aqueous phase and another section that is hydrophilic (lipophobic) and highly soluble in the aqueous phase. The hydrophobic section of the molecule is always a nonpolar aliphatic and/or aromatic hydrocarbon group, whereas the hydrophilic section contains either an ionic group or an accumulation of OH groups (saccharose derivatives) or of poly(alkylene oxide) groups, *e.g.*, $(-CH_2-CH_2-O-)_n$. Depending on the nature of the hydrophilic section they are classified as:
    a. Non-ionic surfactants, where they do not have polar ionic groups. Fatty acid esters, fatty amines and fatty acid amines, polyglycol esters and poly(propylene glycol) esters are part of this group.
    b. Anionic surfactants, where the main entity that is left behind after dissociation is negatively charged. Carboxylates, sulfonates, sulphates and organic compounds of phosphoric acids are part of this group.

c. Cationic surfactants, where the main entity that is left behind after dissociation is positively charged. Amines and ammonium salts are part of this group.

d. Ampholitic or zwitterionic surfactants, when an ionic emulsifier forms zwitterions, that is a molecule that has a total neutral charge but different charges in different atoms. These emulsifiers may act either as acids or as bases, depending on the pH. They are derived from proteins, betaines or aminoacids.

In general (Shaw, 1992) the phase in which the emulsifier agent is the more soluble tends to be the dispersion medium (Bancroft rule). That means that oil-soluble emulsifiers form w/o emulsions and water soluble emulsifiers form o/w ones. An empirical scale of emulsifier hydrophobicity has been developed, *i.e.* the hydrophile-lipophile balance (HLB). In this scale a dimensionless number between 0 and 20 is given to each emulsifier depending on their hydrophilic character, and the least hydrophilic surfactants have the lowest HLB values. There are several formulae for calculating HLB numbers from composition data and they can also be determined experimentally. For mixed emulsifier systems, approximate algebraic additivity holds. Numbers between 0 and 9 characterize oil-soluble hydrophobic products, whereas numbers between 11 and 20 are used for water-soluble oleophobic compounds. The hydrophilic–hydrophobic equilibrium is situated at the centre (10) of this scale. Substances with an HLB value of 10 are distributed between the two phases so that the hydrophilic group projects completely into the water while the hydrophobic hydrocarbon group is adsorbed in the non-aqueous phase.

2.6.4. Droplet size and size distribution

Several studies have been done on predicting droplet size and size distribution in stirred tanks from the physical properties of the system used. It is required to highlight the ones of Calabrese (1986) and Pacek *et al.* (1998).

Sauter diameter (d_{32}) links the area of the dispersed phase to its volume which makes it useful for calculating mass transfer and chemical reaction rates. By definition (Pacek *et al.*, 1998) d_{32} is related to the maximum and minimum droplet diameter, but due to the complexity of dispersion processes in stirred vessels under turbulent conditions it is not possible to have a theoretical description of the process.

However it is possible to estimate the maximum and minimum drop diameters in dilute dispersions following the Hinze and Kolmogoroff models (Pacek *et al.*, 1988). The maximum stable drop size has been estimated from the balance between the turbulent forces tending to disrupt a drop and cohesive surface forces holding it together (Eq.2.2), while the minimum stable drop size has been estimated from the condition that the turbulent energy input into a pair of droplets is insufficient when they contact to prevent adhesion and finally coalescence (Eq.2.3)

$$\frac{d_{\max}}{L} = K_1 \cdot We^{-0.6} = K_2 \cdot N^{-1.2} \qquad \text{[Eq.2.2]}$$

$$\frac{d_{\min}}{L} = K_3 \cdot We^{-\frac{3}{8}} = K_4 \cdot N^{-0.75} \qquad \text{[Eq.2.3]}$$

where:
d_{max} and d_{min} are the maximum and minimum stable diameters
L is the dimension (diameter) of the stirrer
K_1, K_2, K_3 and K_4 are constants with appropriate dimensions

We is the Weber number, the ratio between the inertial force and the surface tension force acting on a fluid element: $We = \frac{\rho_c \cdot N^2 \cdot L^3}{\sigma}$

ρ_c is the density of the continuous phase
N is the stirring rate
σ is the interfacial tension

It has been proposed that for systems where drop size is controlled by break-up, $d_{32} \propto d_{max}$, *e.g.* $d_{32} \propto N^{-1.2}$, whereas for systems where drop size is controlled by coalescence, $d_{32} \propto d_{min}$, *e.g.* $d_{32} \propto N^{-0.75}$. On the other hand, since 1967 (Pacek *et al.*, 1998) it has been assumed that d_{32} depends only on d_{max} and it is proportional to it, following Eq.2.3. for diluted systems. For more concentrated systems Eq.2.5. was suggested.

$$\frac{d_{32}}{L} = K_5 \cdot We^{-0.6} \qquad \text{[Eq.2.4]}$$

$$\frac{d_{32}}{L} = K_6 \left(1 + K_7 \cdot \phi\right) \cdot We^{-0.6} \qquad \text{[Eq.2.5]}$$

where:
ϕ is the volume fraction (defined in Eq. 2.2.)
K_5, K_6 and K_7 are constants with appropriate dimensions

The assumption that d_{32} depends only on d_{max} in a linear way implies that the correlation for d_{32} in which the exponent on *We* is equal to -0.6 has a sound theoretical basis. Without that assumption a more general equation (Eq.2.6) would have to be used:

$$\frac{d_{32}}{L} = K_8 \left(1 + K_9 \cdot \phi\right) \cdot We^{-\alpha} \qquad \text{[Eq.2.6]}$$

where:
K_8 and K_9 are constants with appropriate dimensions

Calabrese (1986) also suggested another extended equation for non-diluted systems [Eq.2.7].

$$\frac{d_{32}}{L} = K_{10}\left(1 + K_{11}\cdot\phi\right)\cdot We^{-0.6}\cdot\left[1 + K_{12}\left(1 - K_{13}\cdot\phi\right)\cdot Vi\left(\frac{d_{32}}{L}\right)^{\frac{1}{3}}\right]^{\frac{3}{5}} \qquad \text{[Eq.2.7]}$$

where:

Vi is the tank viscosity group: $V = i\left(\frac{\rho_c}{\rho_d}\right)^{0.5}\frac{\mu_d\cdot N\cdot L}{\sigma}$

ρ_d is the density of the dispersed phase

μ_d is the viscosity of the dispersed phase

K_{10}, K_{11}, K_{12} and K_{13} are constants with appropriate dimensions

The normalised cumulative volume distribution as a function of normalised drop diameter (d/d_{32}) can be compared with analytical distributions proposed in the literature (Pacek *et al.*, 1998): the normalised cumulative normal volume distribution (Eq.2.8) and the normalised cumulative log-normal volume distribution (Eq.2.9).

$$Fv\left(\frac{d}{d_{32}}\right) = \frac{1}{2}\left[1 + erf\left(\frac{\frac{d}{d_{32}} - \mu}{\sigma\sqrt{2}}\right)\right] \qquad \text{[Eq.2.8]}$$

$$Fv\left(\frac{d}{d_{32}}\right) = \frac{1}{2}\left[1 + erf\left(\frac{\ln\left(\frac{d}{d_{32}}\right) - \mu}{\sigma\cdot\sqrt{2}}\right)\right] \qquad \text{[Eq.2.9]}$$

The normalised cumulative volume distribution (*Fv*) is related to the volume probability density function (*Pv*), for a normal distribution (Eq.2.10) and for a log-normal distribution (Eq.2.11).

$$Pv\left(\frac{d}{d_{32}}\right) = \frac{1}{\sigma\sqrt{2\pi}} \cdot e^{-\frac{\left(\frac{d}{d_{32}}-\mu\right)^2}{2\sigma^2}} \qquad \text{[Eq.2.10]}$$

$$Pv\left(\frac{d}{d_{32}}\right) = \frac{1}{\frac{d}{d_{32}} \cdot \sigma\sqrt{2\pi}} \cdot e^{-\frac{\left(ln\left(\frac{d}{d_{32}}\right)-\mu\right)^2}{2\sigma^2}} \qquad \text{[Eq.2.11]}$$

where μ and σ are the average and the standard deviation of the distribution respectively.

2.7. Microscopy

Microscopy is the technical field of using microscopes to see objects too small to be seen by the naked human eye. During and after the encapsulation process it is necessary to use microscopes of different types to check the quality of the microcapsules produced and to characterize their properties. The size and shape of the microcapsules and the structure and thickness of the polymeric capsule wall created during the process are very important parameters to study and are often related to the final properties (mechanical strength and permeability) of the microcapsules. Optical and electron microscopy (SEM and TEM) can be used to characterise the surface and structural properties of microcapsules.

2.7.1. Optical microscopy

Most common designs of an optical microscope involve passing visible light through (or reflecting from) a sample and several lenses to form a magnified view of the sample. The typical magnification is usually up to 1,500x with a theoretical resolution limit of 0.2μm due to the wavelength of the light (Amelinckx *et al.*, 2008). Systems with shorter wavelength

sources have also been developed to obtain better resolutions (Kriete *et al.*, 2008; O'Farrell, 2006). Usually a digital camera is connected to the microscope and it is possible to take pictures and record videos with the help of a computer.

Optical microscopes provide information about particle size, shape and state of aggregation and they are fast and easy to use.

2.7.2. Transmission electron microscope (TEM)

In TEM a beam of electrons is transmitted through an ultra thin sample, the electrons interact with the sample and an image is formed. Because the wavelength of the electrons is much shorter than the wavelength of light, TEM resolution is very easily below 1nm and with the more advanced aberration corrector designs below 50pm (Kisielowski *et al.*, 2009).

The main problem of using TEM is that the samples must be prepared to obtain a layer thin enough to be electron transparent. These preparations are usually very time consuming and may change the structure of the sample. In addition this technique requires vacuum in the chamber where the sample is deposited, to prevent collisions of the electron with air molecules, which causes losses in resolution. This makes TEM not suitable for samples with volatile components.

2.7.3. Scanning electron microscope (SEM)

SEM looks at the surface of bulk objects by scanning it with an electron beam, and the electrons interact with the atoms on the surface producing signals that are collected by a detector. The signals produced include secondary electrons, back-scattered electrons, characteristic x-rays or light. Almost all the SEMs have a detector for secondary electrons

(those emitted by the superficial atoms when samples are excited with the high energy electrons), and other detectors are rarer.

With the information obtained from the secondary electrons the SEM can produce very high resolution images of the sample surface, with a resolution between 1 and 5nm, and due to the narrow electron beam used a three dimensional image can be formed.

The surface of the sample needs to be electrically conductive, and a coating with a fine layer of gold or other metal is needed. Like the TEM, the SEM works under vacuum to prevent interferences, although environmental SEM (ESEM) has been developed to work at low pressure gas environment and high relative humidity.

2.8. Conclusions

Perfume is not an active ingredient in detergents, but the freshness conferred to the clothes after wash is one of the most important factors for the customers to buy a detergent. The deposition of free perfume on the clothes is very low and new technologies need to be developed to increase it (Aussant *et al.*, 2005); the use of perfume microcapsules is the most promising one.

Perfume microcapsules are designed to release the perfume at the right moment and at the right time. The wall of these microcapsules should be stable in the matrix of detergent, which is a very aggressive medium due to the presence of surfactants. Furthermore, the matrix may interact with the microcapsules allowing the release of the perfume during storage (Ness and McNamee, 2006). The capsules must survive the washing process without breaking (*e.g.* in

washing machine and tumble dryer). They should deposit onto fabric surfaces easily and they must break while people are wearing the clothes, due to rubbing with the skin, so they must have very specific mechanical properties (Caswell *et al.*, 2006).

There are many techniques available to prepare microcapsules but only two of them have been used to obtain microcapsules with the properties described before: *In situ* polymerisation and interfacial polymerisation (Su *et al.*, 2006). From them *in situ* polymerisation has been the one selected by industry as it provides capsules with lower permeability and more stable during storage (Ness, 2004). However *in situ* polymerisation microcapsules have formaldehyde (a carcinogenic chemical) in their composition and in the last years the legislation about free formaldehyde concentration in final products is getting more and more strict (Sumiga *et al.*, 2011). It is needed to develop new formaldehyde free perfume microcapsules and interfacial polymerisation has been selected as the technique able to provide them.

The main advantages of using interfacial polymerisation are the possibility of forming low permeable microcapsules without using formaldehyde (Su *et al.*, 2006) and the possibility of encapsulating water soluble actives as well, which is also of interest for our sponsor.

Perfume microcapsules made by interfacial polymerisation have been studied by our sponsor and the permeability and stability results during storage were not satisfactory. It is required to study further the effect of the different process conditions on the final properties of the microcapsules and produce a shell with the required permeability and mechanical properties.

2.9. Objectives

The main objective of this work is to study the effect of the different parameters of the interfacial polymerisation process on the final properties of the microcapsules produced and to select these parameters to produce microcapsules of industrial use.

The main parameters that have been investigated are:

- Stirring rate.
- Temperature of reaction.
- Type and concentration of organic monomers used.
- Type and concentration of aqueous monomers used.
- Addition time of aqueous monomers to the reactor.

Those parameters had an influence on the properties of the microcapsules formed:

- Microcapsule size and size distribution.
- Thickness of the wall.
- Permeability of the wall.
- Mechanical properties of the wall.

The best process conditions have been selected to produce perfume (organic soluble active) microcapsules that may be of industrial use in detergent formulations. It has also been studied the possibility of encapsulating glycerol (aqueous soluble active) for their use in cosmetic formulations (lipsticks).

CHAPTER 3:

MATERIALS

AND METHODS

An experiment is a question which science poses to Nature,

and a measurement is the recording of Nature's answer.

Max Planck

3.1. Introduction

In this chapter are described the chemicals, equipment, analytical methods and procedures used in this PhD project.

The different chemicals, showing the structure of all the monomers used, and the interfacial polymerisation technique developed during this project to prepare microcapsules are illustrated. The analytical methods used to follow the reaction leading to formation of microcapsules and posterior characterisation are described in detail. These analytical methods include the measurement of the reactivity of the monomers with the perfume via gas-chromatography, the characterisation of the reaction kinetics by monitoring the pH, the measurement of the release of the encapsulated perfume from the microcapsules via UV-spectrophotometry, the mechanical characterisation of single microcapsules using a micromanipulation technique, the morphological and structural characterisation with several types of microscopes (optical, SEM, TEM), measurement of the particle size distribution and the analysis of the microcapsules wall chemistry via spectrometry.

3.2. Chemicals

Perfume (X-Ray 2 GNF, see Table 3.1) was a gift from Procter & Gamble (Brussels, Belgium and Newcastle, UK). Sebacoyl chloride (SC), terephthaloyl chloride (TC), trimesoyl chloride (Trim), ethylenediamine (EDA), hexamethylenediamine (HMDA), diethylenetriamine (DETA), triethylenetetramine (TETA) and polyvinyl alcohol 87-89% hydrolysed, 13,000 MW (PVA) were supplied by Sigma-Aldrich (Dorset, UK) and were used as received without further purification. In Table 3.2, the chemical structure of all the monomers used is shown.

Ingredient	**Content (%wt.)**
Allyl Caproate	1-2.5
Allyl Cyclohexane Propionate	2.5-5
Beta Naphthyl Methyl Ether Extra 99	5-10
Coumarin	1-2.5
Decyl Aldehyde	5-10
Frutene	10-20
Hexyl Cinnamic Aldehyde	5-10
Hexyl Salicylate	5-10
Ligustral	5-10
Lyral	1-2.5
Methyl Nonyl Acetaldehyde	1-2.5
Nectaryl	1-2.5
Nonyl Aldehyde	2.5-5
Octyl Aldehyde	2.5-5
P. T. Bucinal	10-20
Undecalactone	1-2.5

Table 3.1 Composition of the encapsulated perfume, X-Ray 2 GNF

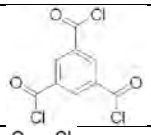
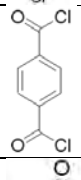
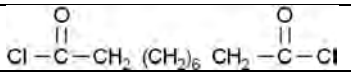
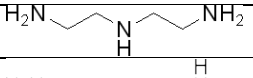
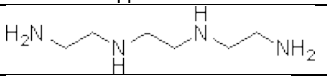
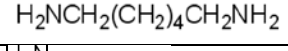
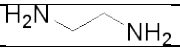
Organic monomers:	
Trimesoyl chloride (Trim)	
Terephthaloyl chloride (TC)	
Sebacoyl chloride (SC)	
Aqueous monomers:	
Diethylene triamine (DETA)	
Triethylene tetramine (TETA)	
Hexamethylene diamine (HMDA)	
Ethylene diamine (EDA)	

Table 3.2. Chemical structure of the monomers used

3.3. Interfacial polymerisation

The basic steps of the interfacial polymerisation process used to produce perfume microcapsules are illustrated in Figure 3.1.

Firstly 2g of polyvinyl alcohol (PVA) were dissolved in 200g of distilled water while heating (40°C) under magnetic stirring. Once the PVA was dissolved the solution was cooled in an ice bath and placed in a 500mL jacketed reactor (Lenz Laborglas, Wertheim, Germany) connected to a cooling circulator (Fisher Scientific, Loughborough, UK) where the temperature of the solution was finally adjusted.

At the same time the perfume (40g) was cooled in ice and when cold the acid chloride(s) (usually 45meq of COCl functions, 3.98g Trim, 4.57g TC or mixtures of them) were dissolved in it under magnetic stirring in an ice bath to prevent reaction.

A solution of the amines (usually meq of NH_2 were 5 times the COCl meq, in this case 225meq: 7.74g DETA, 13.10g HMDA, 6.76g EDA, 8.23g TETA or mixtures of them) in 10ml distilled water was also prepared and cooled in ice. When different amines were added at different times, a solution of the amine in 10ml water was prepared for each amine.

To prepare the emulsion a homogenizer (Silverson L4RT, Silverson, Chesham, UK) equipped with an Emulsor Screen was used. It was placed in the reactor and when the temperature of the water phase was adjusted (usually to -1°C), the perfume with the organic monomer(s) was added to it dropwise.

After 10min at 1,500rpm the homogenizer was stopped and the reactor was placed under mechanical stirring (500rpm) using a Rushton turbine (Figure 3.2), 4 baffles were also placed in the reactor to improve mixing. The first amine solution was added to it and the reaction between the two monomers started. If more amines were used their solutions were added sometime after (see Table 4.1 for full details of all formulations). During the whole reaction the temperature of the reactor was controlled (usually 0°C). The reaction was carried out for at least 6 hours and the resulting microcapsule suspension was stored at room temperature in a glass bottle.

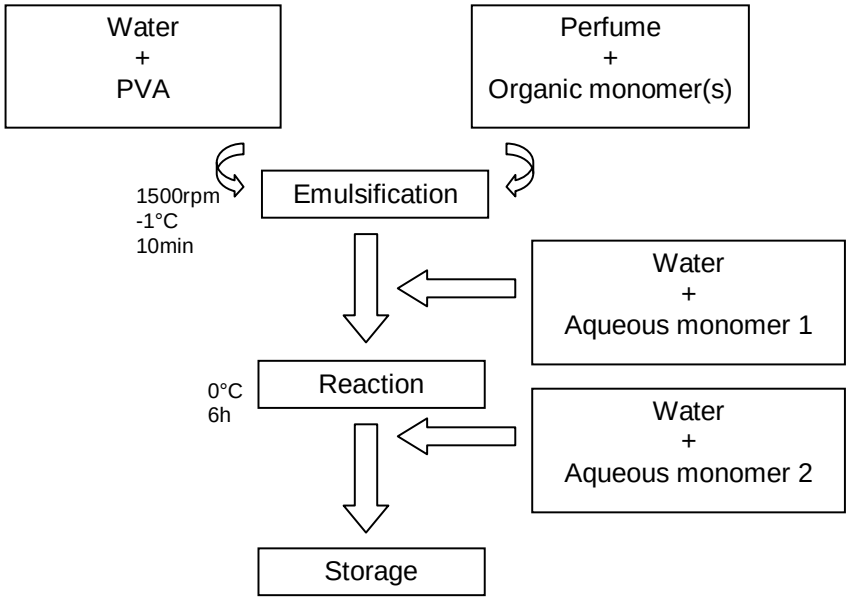


Figure 3.1. Illustration of preparation steps of the interfacial polymerisation method.

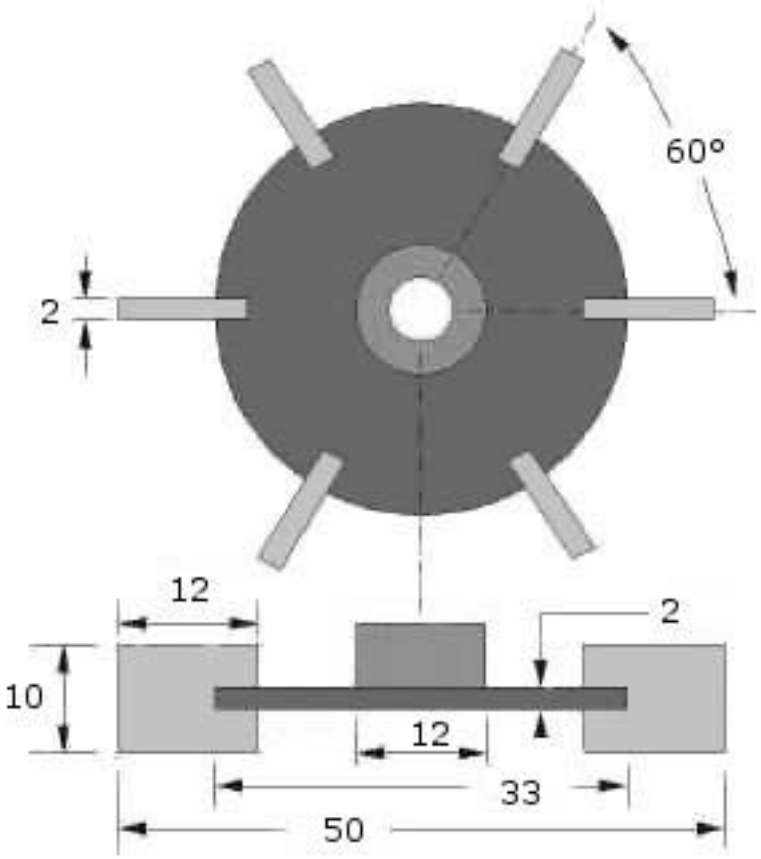


Figure 3.2 Rushton turbine geometry. Dimensions in mm.
(Adapted from http://www.dantecdynamics.com/Default.aspx?ID=507)

3.4. Reactivity of the monomers with the perfume

To study the reactivity of the monomers with the perfume at room temperature, 2g of trimesoyl chloride were added to a beaker with 50ml perfume. The monomer was dissolved in it under magnetic stirring and n a sample of 10μl was taken and dispersed in 1ml pure hexane inside a vial. The same procedure was followed with 2g of diethylenetriamine. Those two vials and one more with 10μl pure perfume in 1ml hexane were placed in a gas chromatograph (GC) system (Agilent 6850 Network GC System with autosampler, Agilent Technologies, Wokingham, UK).

GC is able to separate the different components of a mixture and identify their concentration. It allows a gas stream ("mobile phase", usually helium or nitrogen) with the sample of interest (it needs to be vaporized in advance if it is a liquid at normal conditions) to pass though a column. The gas containing the sample pushes it through the column and forces it to go out. The column has a layer of a "stationery phase" embedded or fixed on the walls (usually a liquid or polymer) which interacts (via adsorption) with each of the components of the sample in a different way, making them to separate. Columns are usually very thin and long to obtain good separations. When each of the components arrives at the end of the column (at different times) a detector measures the time at which each of them elutes and the amount of chemical present, plotting the result in the form of peaks at different times. The area contained under each peak gives a measurement of its concentration, although it needs to be calibrated. The configuration of the GC (flow rate, temperature, temperature increase profile, type of column) has an effect on the strength of the interactions between the stationary phase and the chemicals of interest. In complex mixtures it is needed to adjust all the parameters to obtain a good separation of all the components.

The GC set-up was as follows: An injection volume of 0.2µl of sample was automatically taken from the vial and placed in the injection chamber, where it was vaporized at 200°C. A flow of an inert gas (helium) met the vapour produced (split ratio 75/1, which means that only 25% of the injected sample was taken in the helium stream, the rest was discharged) and moved it in the column (Agilent 100-2000 DB5: 30.0m x 250µm x 0.10µm nominal). The stationary phase is (5%-Phenyl)-methylpolysiloxane) placed in an oven. The temperature of the oven was programmed: Initially at 60°C and held it for 5min, after that the temperature was increased at a heating rate of 5°C/min until 180°C was reached and then the heating rate

was increased to 20°C/min until 250°C was reached to ensure that all the components of the perfume left the column. At the exit of the column a FID detector (at 300°C) read the signal that was recorded in a computer.

3.5. Reaction kinetics

As described in section 2.4.2, it is possible to monitor the reaction kinetics of polyesters and polyamides formation. During the reaction a polyester or polyamide bond is formed and HCl is released to the media. This HCl reduces the pH of the media.

The pH of the reaction media was measured with a pH meter (Mettler Toledo MP230, Leicester, UK) which had been previously calibrated. The pH was monitored during the reaction time (6h).The resolution of pH reading was 0.01 units.

3.6. Leakage experiment

The leakage experiment was designed to measure the permeability of the microcapsule wall, aiming to compare the data of perfume release versus time for different microcapsule formulations.

The experiment started with dispersing 10g of PMCs slurry in 60ml distilled water in a 250ml beaker or glass bottle under magnetic stirring (150rpm). At time 0 min, 50ml of pure hexane was added carefully to the system and two phases were formed, a water phase with the slurry at the bottom and a hexane phase on the top. As the hexane is highly volatile and evaporates fast, the bottle was closed with its cap and the beaker with parafilm. The perfume that leaked out from the microcapsules to the water phase was extracted by the hexane and the perfume

concentration in the hexane was monitored over time by taking samples of perfume at times 1, 30, 60, 90, 120, 240, 360 and 480min. 50μl of hexane from the beaker (or bottle) were taken each time and diluted in 1.4ml of pure hexane in a quartz cuvette, and the absorbance was measured with a UV spectrophotometer (Cecil CE 2021, Cecil Instruments, Cambridge, UK) at a wavelength of 270nm.

A calibration curve with known amounts of perfume in hexane was previously obtained. A solution of 10μl perfume in 40ml hexane (0.025%v/v) was prepared. 5ml of this solution were mixed with another 5ml of pure hexane resulting in a 0.0125%v/v perfume solution. Repeating the process twice, 2 new solutions of 0.00625%v/v and 0.003125%v/v perfume concentrations were also prepared. Finally another 5ml of the last solution were mixed with 15ml pure hexane resulting in a 0.00078125%v/v perfume solution. Measuring a sample of every known perfume concentration with the UV spectrophotometer at 270nm, the calibration curve obtained is shown in Figure 3.3. The slope of this curve was used in all the calculations involving measurement of perfume concentrations with the UV spectrophotometer.

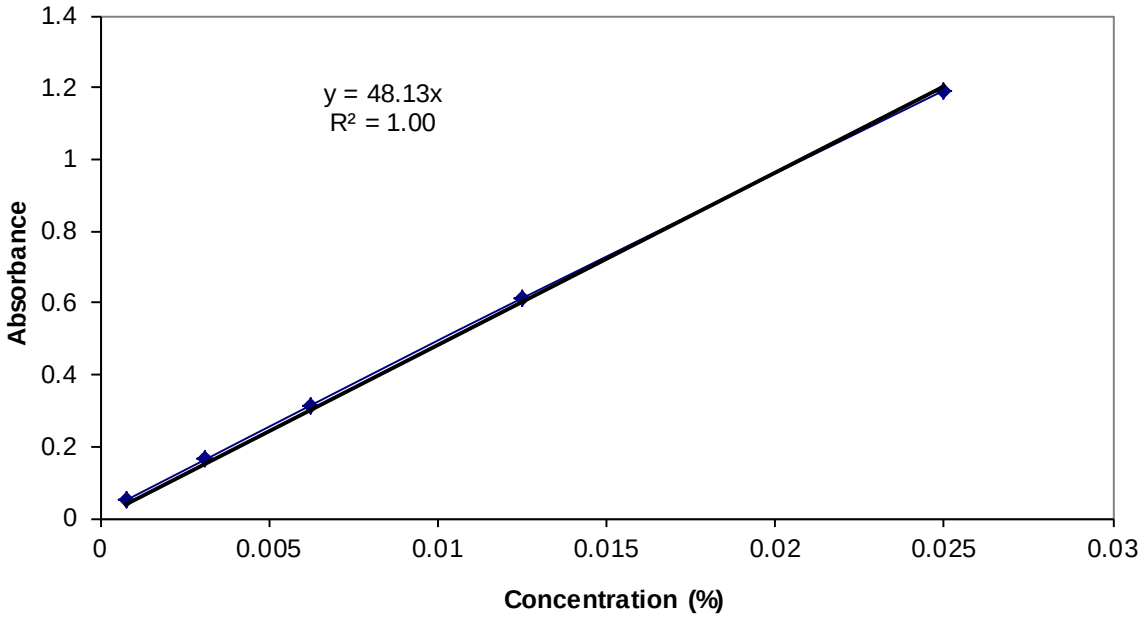


Figure 3.3. Calibration curve of the X-Ray 2 GNF perfume in the UV spectrophotometer at 270nm.

3.7. Mechanical characterisation of single microcapsules

A micromanipulation technique has been used to measure the mechanical properties of single microcapsules. This technique is based on the compression of single microcapsules between two parallel surfaces. A scheme of the micromanipulation rig is shown in Figure 3.4.

Results of compression are presented showing error bars. These error bars have been calculated by doing a "t-student" statistical analysis of the results, having considered how many microcapsules have been compressed for each sample. The value presented corresponds to a 95% confidence interval (t=0.025).

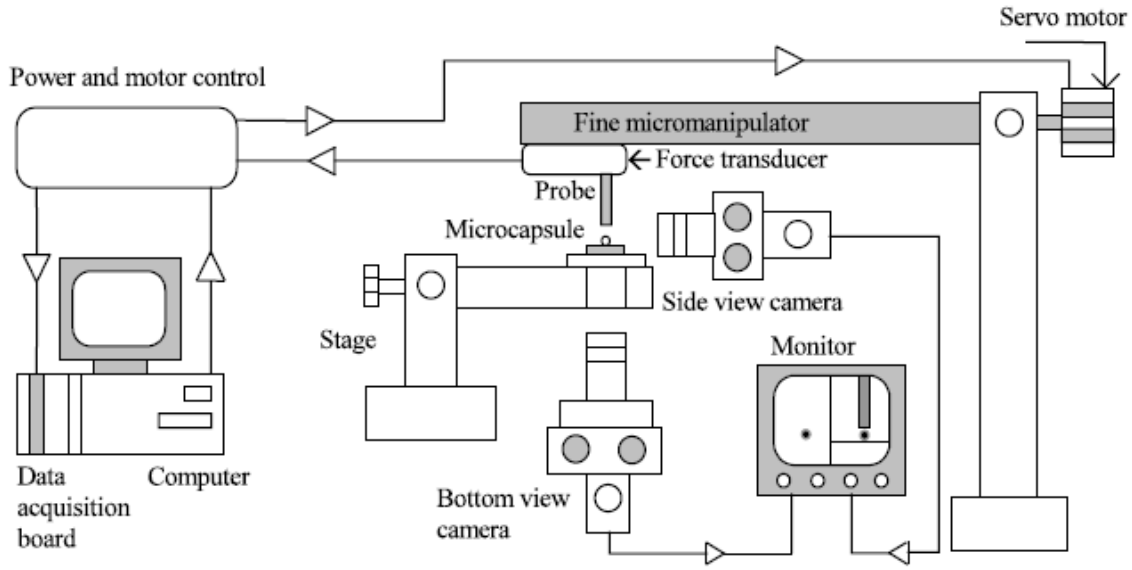


Figure 3.4. Schematic diagram of the manipulation rig. (Adapted from Sun and Zhang, 2002).

A glass probe (80μm diameter) was glued to a force transducer (Aurora Scientific Inc., Canada) which was firmly screwed to the fine micromanipulator. The microcapsules were placed on a glass slide which was firmly secured with a screw under the probe. The positions of both cameras were adjusted to focus first on the probe and later on single microcapsules. By moving the glass slide, different microcapsules were placed below the probe which permitted to choose an isolated microcapsule for compression. Once a microcapsule was chosen its diameter was measured on the TV screen with a ruler (the dimensions of the screen were previously calibrated) and the compression took place measuring simultaneously the force imposed on the single microcapsules and the time. The data were collected by a computer program and exported to an Excel® file where they were analysed.

Three different types of compression experiments have been carried out:

1. Compression of single microcapsules up to burst at 2μm/s: Between 30 and 50 microcapsules were compressed for each experimental condition to have

representative data. The force, pseudo stress and deformation at rupture were calculated.

2. Compression to different deformations at three speeds (2, 5 and 8μm/s) and then holding each microcapsule under the probe. The visco-elastic behaviour of the microcapsules if any was determined.
3. Compression to different deformations and release, at three speeds (2, 5 and 8μm/s). The plastic behaviour of the microcapsules was determined.

3.8. Morphological and structural characterisation

3.8.1. Optical microscopy

Digital images of microcapsules under different magnifications were taken and processed using an image analysis software package (Leica Qwin Standard) in a desktop computer connected to an optical upright microscope (Leica DMRBE, Leica Microsystems, Milton Keynes, UK).

As described in section 2.7.1 the resolution of optical microscopy is 0.2μm.

3.8.2. SEM microscopy

A Philips XL-30 FEG ESEM with Oxford Inca EDS (Philips Electronics UK Ltd, Guildford, UK) was used to look at the microcapsules and take micrographs. A sample of microcapsules was placed in the chamber and after the vacuum was generated they were covered with a thin layer of gold to increase the sample electric conductivity, focused and photographed.

The principle of the technique was explained in more detail in section 2.7.3, as indicated there the resolution of the SEM is between 1 to 5nm.

3.8.3. TEM microscopy

3.8.3.1. Sample preparation

The sample preparation was made by the personal of the Centre for Electron Microscopy (CEM) in the University of Birmingham. The procedures they used involve two steps: firstly dehydration of the sample using ethanol and secondly embedding of the sample in a resin so that it can be cut into thin layers for microscopy.

Previously to the dehydration step 0.5g of slurry and 1ml 2.5% glutaraldehyde fixative solution (used to strengthen the walls of microcapsules) were put in an eppendorf tube, which was centrifuged at 2,000rpm for 2min. The excess fixative solution was removed from the sample and the dehydration step begun: up to 10ml 50%v/v ethanol was placed into snap top vials where the sample was transferred using a pipette. The ethanol was left for 30min in contact with the sample. After that the sample was centrifuged (2,000rpm, 1min) and the ethanol top layer removed from the sample. The process was repeated using 70%v/v ethanol, 90%v/v ethanol and finally absolute ethanol. Once the sample was dehydrated, the absolute ethanol was removed, the sample was embedded in 50% ethanol/LR White Resin and left in a rotator (4rpm) for one day. The resulting resin was again embedded in pure LR White Resin for another two days (in a rotator at 4rpm). After 2 days the mixture was put into a mould in an oven at 60°C overnight and a polymerisation reaction occurred. Ultrathin sections were obtained by sectioning and trimming the polymerised resin with a ultramicrotome.

3.8.3.2. Sample analysis

A Jeol 1200EX TEM (Jeol Ltd., Welwyn Garden City, UK) was used to look at the microsections and take micrographs. The obtained micrographs were scanned and the wall thickness and microcapsule size were measured using photographic software (ImageJ and GIMP2).

The principle of the technique was explained in section 2.7.2, as indicated there the resolution of TEM is usually below 1nm, achieving 50pm.

3.9. Particle size distribution measurement

The particle size distribution measurements were done using a Mastersizer 2000 instrument (Malvern Instruments Ltd, Malvern UK) and its software package. The instrument measured the volume fraction of particles in a number of size intervals. A refractive index of 1.5 for the polyamide (Brandrup *et al.*, 1999) microcapsules was used in the calculations. The data obtained from the instrument was analysed by the software, the results exported to an Excel® file where the probability density function (PDF) was calculated.

According to Malvern Instruments webpage, the technique of laser diffraction is based on the principle that particles passing through a laser beam will scatter light at an angle that is directly related to their size. As the particle size decreases, the scattering angle increases logarithmically. This angle and the intensity of the scattered light are measured by detectors placed conveniently

The particle size distributions are calculated by comparing the sample's scattering pattern with the Mie Theory using a mathematical inversion process. Mie Theory provides a rigorous solution for the calculation of particle size distributions from light scattering data and is based on Maxwell's electromagnetic field equations. It predicts scattering intensities for all particles, small or large, transparent or opaque within the following assumptions:

- The particles being measured are spherical
- The suspension is dilute, such at the scattered light is measured before it is re-scattered by other particles.
- The optical properties of the particles and the medium surrounding them are known.
- The particles are homogeneous

It is assumed that the particles being measured are spherical. The particle size of irregular particles is therefore expressed in terms of a spherical equivalent diameter. In the case of laser diffraction, the diameter of the sphere that would produce an equivalent light scattering pattern to the measured particle is reported.

3.10. Analysis of microcapsule wall chemistry

Infra Red spectra provide valuable information on the bonding state of polyamides. A Fourier Transform Infrared (FTIR) spectrometer (Thermo Nicolet 380, Thermo Scientific, Welwyn Garden City, UK) with an accessory for single reflection diamond ATR analysis (Smart Orbit) has been used to obtain the spectra of several polyamide walls.

The microcapsules were broken with glass beads under magnetic stirring in hexane to extract all the perfume from the polymer walls. Once the microcapsules were broken, the solids were

filtered and washed twice with hexane and left to dry in air on the filter at room temperature and stored. The spectrum from the obtained powders was measured by placing them directly on the diamond crystal, compressing them with the pressure tower of the ATR accessory and recording the spectrum against air.

For the pure liquid amines, a drop of pure amine was also placed directly on the diamond crystal of the ATR accessory and the spectrum was measured against air.

In infrared spectroscopy, IR radiation is passed through a sample. Some of the infrared radiation is absorbed by the sample and some of it passes through (is transmitted) and it is measured by a detector. The resulting infrared spectrum represents a fingerprint of the sample with absorption peaks which correspond to the frequencies of vibration between the bonds of the atoms forming the material. Because each different material is a unique combination of atoms, no two compounds produce the exact same infrared spectrum.

FTIR measures all of the infrared frequencies simultaneously. For doing that it is used a simple optical device called an interferometer. The interferometer produces a unique type of signal which has all of the infrared frequencies "encoded" into it. This signal can be measured very quickly. The measured interferogram signal needs to be “decoded” in order to provide a frequency spectrum. This can be accomplished via a well-known mathematical technique: the Fourier transformation. This transformation is performed by the computer automatically presenting the desired spectral information for analysis.

Because a relative scale for the absorption intensity is required, a background spectrum must also be measured. This is normally a measurement with no sample in the beam. This can be compared to the measurement with the sample in the beam to determine the "percent transmittance" This technique results in a spectrum which has all of the instrumental characteristics removed. Thus, all spectral features which are present are strictly due to the sample.

CHAPTER 4:

PRODUCTION OF PERFUME MICROCAPSULES

An expert is a man who has made all the mistakes

which can be made in a very narrow field.

Niels Bohr

Summary

Perfume microcapsules with polyamide walls have been produced. Polyester walls were studied in an early stage but they were not satisfactory. Different batches of microcapsules made with different monomer formulations and reaction conditions have been produced. Microcapsules have been characterised and the results are presented in this and the next chapter.

In this chapter, the different experimental conditions used to produce the microcapsules are described. The results about the size and size distribution of the capsules formed and their correlation with theoretical models are presented. The reaction kinetics and the influence on it of the different monomer formulations and reaction conditions have been also investigated. Finally results about microscopy image analysis (morphology of the capsules and thickness of the wall) are shown.

4.1. Introduction

Microcapsules developed for carbonless copy paper were soon also used for encapsulating perfumes and “Scratch and Sniff” stickers became very popular in the late 1970s and early 1980s (Nelson, 1991; Lawton and Forbes, 1980). That popular technology consisted of coating usually small paper with perfume microcapsules, which delivered their perfume when they were broken by scratching with the fingers.

When perfume microcapsules started being used in liquid matrixes the problem of their stability and perfume leakage over time arose. It was found especially difficult to obtain microcapsules with low perfume release in detergents where the surfactants present in them

interacted with the microcapsules making holes on the walls and increasing the perfume release during storage (Ness and McNamee, 2006).

The use of microcapsules in laundry compositions (detergents and fabric enhancers) should also have very specific mechanical properties. They should survive various processing and handling steps where mechanical forces are generated, such as pumping, mixing washing and drying, but they should break by the mechanical forces generated during normal use of the clothes (Caswell *et al.*, 2006).

Perfume microcapsules should be very resistant to leakage during storage and have specific mechanical properties. Melamine-formaldehyde microcapsules were found to accomplish with this properties better than other materials (Ness, 2004), but they use formaldehyde in their formulation.

Formaldehyde is suspected to be a carcinogenic compound (with limited evidence of carcinogenic effect) and its concentration in final products is highly controlled by law (Sumiga *et al.*, 2011). The free formaldehyde present in the final slurry after making the microcapsules can be scavenged using some chemicals, but it has been found that the formaldehyde forming part of the capsule walls is in equilibrium with the free formaldehyde in the slurry, therefore it is not possible to completely eliminate the free formaldehyde from the slurry, and there will always be a residual concentration.

Therefore there is a need to change the technology used to encapsulate perfume for laundry products and interfacial polymerisation may be used.

Interfacial polymerisation is a technique developed more than 50 years ago. Over the years it has been used to encapsulate oil and water soluble substances, with the main application in encapsulation of agrochemicals with sustained release. Recently new applications have been found, like encapsulation of phase change materials, which needed completely different microcapsule properties and interfacial polymerisation has demonstrated its versatility being one of the techniques used.

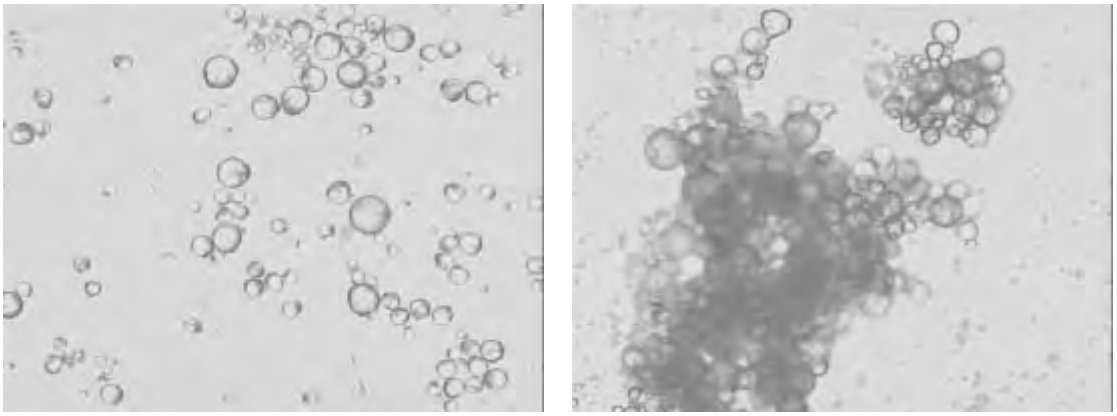
4.2. Preliminary work (Polyester walls)

There was no previous knowledge about how to make perfume containing microcapsules using interfacial polymerisation and a process had to be developed from the beginning. An extensive bibliographic search was done and once the experimental conditions were chosen it was decided to start encapsulating soya oil instead of perfume in the first steps of development to reduce costs. It was also decided to use polyester as an encapsulating polymer to form the walls of the capsules. The first objective was to choose the best experimental conditions to produce stable microcapsules.

Polyester is formed in the reaction of an acid chloride as organic monomer (sebacoyl, trimesoyl and/or terephthaloyl chloride) with a polyol as aqueous monomer (trimethylol propane and/or polyethylene glycol). This reaction takes place on the interface, where the two monomers meet. The COCl group of the polyacid chloride reacts with the OH group of the polyol and they form an ester bound (-CO-C-) and a molecule of HCl is released to the media. Polyols are very soluble in water but polyacid chlorides do not dissolve in soya oil easily. In order to dissolve terephthaloyl chloride it was needed to heat at about 35-40°C, but trimesoyl chloride was mainly insoluble in it so that all the experiments have been done with

terephthaloyl chloride. An alkali (sodium carbonate) was added to neutralize the hydrochloric acid formed during the reaction.

After trying different combinations of these monomers some soya oil-polyester microcapsules were produced at temperatures ranging from room temperature 25°C to 65°C. Using a 0.5% of PVA as surfactant and a stirring rate of 3,000rpm capsules of about 35microns were produced. The microcapsules were evaluated using a microscope and image analyzer. It was found that the presence of sodium carbonate in the media made the capsules form huge aggregates, as it is shown in Figure 4.1, and the addition of calcium chloride (to prevent the formation of such aggregates) did not generate a significant effect on the aggregates but made the capsules weaker. Consequently, the bigger capsules were broken and only the smaller ones remained.

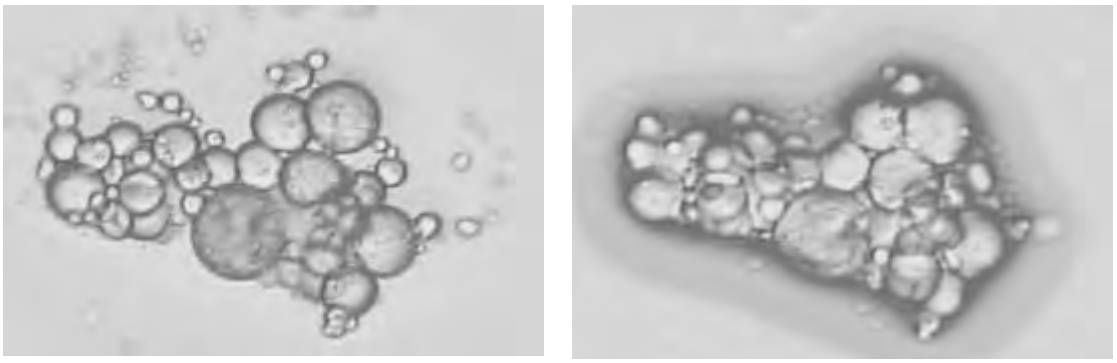


a b

Figure 4.1. Soya oil-polyester microcapsules. Without sodium carbonate (a) and with it (b).

The main result of the research with polyester walls was that the capsules formed were very weak. All the microcapsules made with different formulations collapsed when the capsules were dried. The rupture was directly observed under the microscope when the slides

containing the microcapsules dried because of the water evaporation at room temperature while photographs were being taken. This effect is shown in Figure 4.2.



a b

Figure 4.2. Microcapsules, wet (a) and dry (b).

In the light of the results obtained with polyester it was decided to change the wall material and to make microcapsules with polyamides.

Soya oil was successfully encapsulated at room temperature using polyamides. The capsules formed did not collapse when they were dried, therefore polyamide has been selected as the polymer to encapsulate the perfume in the following work.

4.3. Experimental conditions

Starting with the experimental configuration used to produce soya oil microcapsules with polyester walls many experiments have been done to adjust the experimental conditions to produce perfume microcapsules with polyamide walls.

A big difference between the two systems is the core material to be encapsulated. Perfume has different physicochemical properties from soya oil, which leads to use a lower stirring rate to

produce the same capsule sizes. To produce capsules of about 30 microns at room temperature, a stirring rate (using a Silverson homogenizer device to prepare the emulsion) of 3,000rpm was used to encapsulate soya oil and only 1,500rpm was required to encapsulate the perfume. In addition, as the core material is different and the organic monomer has to be dissolved in it, the solubility of the monomer in the organic phase is different. In soya oil trimesoyl chloride was almost insoluble and terephthaloyl chloride had to be heated to dissolve, however in perfume oil terephthaloyl chloride was soluble at room and low temperatures and the solubility of trimesoyl chloride was high enough to produce capsules with it.

The interaction of the monomers with both oils was also different. There was no significant change in the soya oil when the monomers were dissolved in it, however if perfume was heated to dissolve the terephthaloyl faster or if trimesoyl or sebacoyl chlorides were added to the perfume at room temperature, a colour change in the perfume was observed. The monomer reacted with some of the components in the perfume and it changed colour from pale yellow to dark brown or even pink. To minimize the loss of perfume and monomer and the formation of new unwanted chemical species by reaction, the perfume was cooled before adding the monomers and those were dissolved using an ice bath to maintain a low temperature during the dissolving step.

The aqueous monomers used in both systems are also different. Instead of polyols (for polyester) polyamines are used to produce polyamide. The main experimental difference between them is that using polyamines it is not needed to add an alkali to the system to neutralize the hydrochloric acid formed during the reaction because the own polyamines are

alkalis and neutralize it. It is possible to use an external alkali (usually sodium carbonate) but other authors (Soto Portas *et al.*, 2003) have found that the capsules produced are better when an excess of polyamines instead of another alkali is added to the system.

It was early found that only some combinations of organic and aqueous monomers produced microcapsules at room temperature. When sebacoyl chloride was used as organic monomer and diethylenetriamine as aqueous monomer, aggregates of very small microcapsules were produced and most of the perfume was not encapsulated. When hexamethylenediamine or ethylenediamine was used as aqueous monomer and trimesoyl chloride as organic monomer, a polymer was formed and no capsules were found. Furthermore part of the perfume was embedded in the polymer and the rest was found to be free. Only when terephthaloyl or trimesoyl chloride was used as organic monomer and diethylenetriamine as aqueous one, free microcapsules were produced at room temperature. Later on it was found that the other amines produced good quality capsules when the reaction was carried out at low temperatures (around 0°C).

The **temperature of reaction** was also found to have a very big influence on the properties of the capsules. All experiments made above 20°C produced weak capsules that (like the polyester ones of the previous section) collapsed when the capsules were dried. Capsules made below 18-20°C were stable in dry conditions and it was possible to use the micromanipulation rig to measure their mechanical properties. It seems that the reaction kinetics has a big influence on the wall structure of the capsules. The lowest temperature it was possible to work at is 0°C (there was water in the system) and **four experiments** at

different temperatures (0, 6, 12 and 18°C) have been done to study the effect of the temperature on the properties of the final capsules.

The effects of the **organic monomer type and concentration** have also been studied. **Six experiments** have been done: Trimesoyl chloride (Trim) alone, half the concentration of trimesoyl chloride, terephthaloyl chloride (TC) alone and combinations of trimesoyl and terephthaloyl chloride but fixing the amount of trimesoyl chloride and adding terephthaloyl chloride to the system (10, 18 and 25% COCL functions of terephthaloyl chloride). In all the experiments diethylenetriamine was used as aqueous monomer.

The **type of aqueous monomer** on the final properties of the capsules was also investigated. **Four experiments** using the four aqueous monomers alone, with trimesoyl chloride as organic monomer, have been done: triethylenetetramine (TETA), diethylenetriamine (DETA), hexamethylenediamine (HMDA) and ethylenediamine (EDA).

Apart from the temperature of reaction, the availability of aqueous monomer in the system was also varied to control the reaction kinetics. Some experiments were done by adding the aqueous monomer dropwise and very slowly (18ml in 14min) to the system but the capsules obtained were big and the size distributions were wide, presenting 2 peaks in some cases (see Figure 4.3).

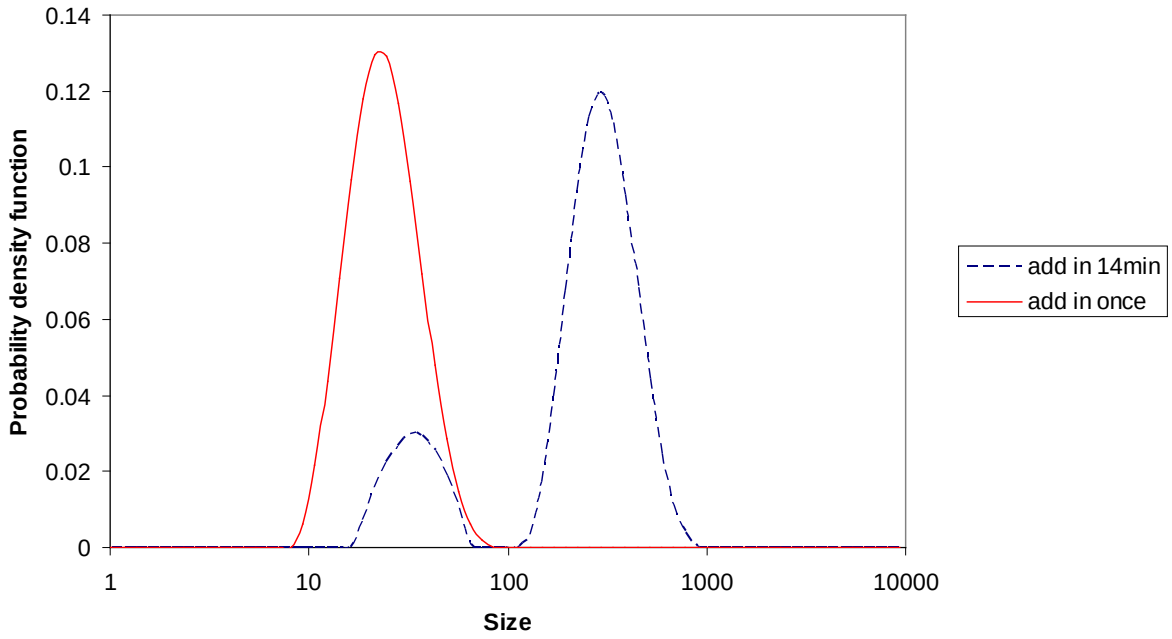


Figure 4.3. Size distribution of microcapsules prepared by adding the aqueous monomer dropwise in 14min and adding it in one time

It seems that the reaction speed was very slow and the droplets formed in the emulsion step coalesced before the first membrane was formed around them. The addition rate was increased until a good capsule size distribution was obtained (18ml in 1min) but it results in too fast addition of the monomer to control the kinetics so other approach was used. It is supposed that a critical amount of aqueous monomer is needed at time 0 to create the first membrane around the droplets to prevent them from coalescence. Therefore a small amount of aqueous monomer was added at time 0 and more monomer was added at different times after that. Previous work suggested use of 5 to 30 mol of aqueous functional groups for one organic one (Mathiowitz and Cohen, 1989a; Soto-Portas *et al.*, 2003), as the concentration of amine in the reaction site (organic side of the membrane formed) is lower than in the bulk and hydrochloric acid is produced during the reaction and needs to be neutralized by an excess of amine. The lowest limit (5X) has been selected to maintain a slow reaction rate and it has been found that the reaction finished in a reasonable time. Some experiments have been done

by adding all the aqueous monomer at time 0, and others by adding 1X at time 0 and at least 4X between time 10 and 60min. Moreover, several experiments have been done by adding different monomers at different times. The rule followed is that the monomer with slower reaction kinetic is added first so, as it is possible to see in section 4.5, the addition order has been DETA-HMDA-EDA.

Batch Name	Monomers added (meq) and addition time (min)*						T reaction(°C)
	Trim	TC	DETA	TETA	HMDA	EDA	
0°C (or DETA)	45	-	225	-	-	-	0
6°C	45	-	225	-	-	-	6
12°C	45	-	225	-	-	-	12
18°C	45	-	225	-	-	-	18
½ Trim	22.5	-	112.5	-	-	-	0
TC	-	45	225	-	-	-	0
5TC	45	5	250	-	-	-	0
10TC	45	10	275	-	-	-	0
15TC	45	15	300	-	-	-	0
TETA	45	-	-	225	-	-	0
HMDA	45	-	-	-	225	-	0
EDA	45	-	-	-	-	225	0
DETA1+4	45	-	45+180(15')	-	-	-	0
DETA+EDA	45	-	45	-	-	180(10')	0
HMDA15'	45	-	45+20(15')	-	160(15')	-	0
HMDA1h	45	-	45	-	180(60')	-	0
HMDA+EDA	45	-	45	-	70(10')	210(20')	0
TC+HMDA	45	10	55	-	200(15')	-	0
All50	45	5	50	-	250(15')	50(60')	0
All250	45	5	250	-	250(15')	50(60')	0
20%paraffin	45	10	275	-	-	-	0

Table 4.1. Experiment formulations. *addition time is 0min when not stated.

Finally one experiment was done to study the effect of the viscosity of the organic phase on the leakage of the perfume. In theory, the more viscous is the core material, the lower is the leakage since it is more difficult for the perfume to diffuse from the inside of the capsule to the wall. To increase the viscosity, paraffin oil was added to the perfume. Adding a 20% of paraffin oil the viscosity of the organic phase at 0ºC was almost doubled, from 5.4cP to 9.9cP (measured with a AR1000, TA Instruments (Elstree, UK) using a cone/plate geometry and a

constant shear rate of $100s^{-1}$). Table 4.1 shows the composition of all the experiments that have been characterized.

4.4. Capsule size and size distribution

4.4.1 Sauter mean diameter

As described in section 2.6.4, Sauter mean diameter can be calculated using a semiempirical equation (Calabrese *et al.*, 1986; Pacek *et al.*, 1998). This equation [Eq.2.4, 2.5, and 2.6] depends on the *We* number, which is a function of the density of the continuous phase and the interfacial tension.

The presence of the organic monomer dissolved in the perfume made it difficult to measure the interfacial tension (σ) of the system because the monomer reacts with the perfume at room temperature and it forms a layer at the interface in contact with the water phase at low temperature. Only the interfacial tension of one of the systems used has been measured (using a K100 tensiometer from Kruss (Hamburg, Germany) with a Wilhelmy plate): perfume with only trimesoyl chloride dissolved in it as organic phase and water with 1% PVA as aqueous one, at 2°C. The value of interfacial tension obtained was: σ=4±0.04mN/m. There are 12 experiments done with this system (Table 4.2) in which the size and size distribution of the microcapsules were measured. A Mastersizer 2000 was used to measure both of them.

Experiment	Stirring rate (rpm)	d_{32} (μm)	span
Exp.1	1000	35.61	1.187
Exp.2	1200	31.778	1.028
Exp.3	1200	30.877	1.01
Exp.4	1200	30.596	0.95
Exp.5	1200	29.807	1.036
Exp.6	1300	26.565	1.071
Exp.7	1300	25.041	1.147
Exp.8	1400	26.612	0.977
Exp.9	1400	23.162	1.047
Exp.10	1400	24.663	0.928
Exp.11	1400	23.454	1.029
Exp.12	1500	22.594	1.275

Table 4.2. Experiments used in this section to correlate d_{32} data and particle size distribution.

The 12 experiments showed in Table 4.2 were not done at the same temperature, an interval between -2 and 15°C was used. As a first approach it was assumed that the density and interfacial tension of the system did not differ too much for all the experiments done in this short interval of temperatures and a constant value for them has been used in the calculation of the Weber number: σ=4mN/m and ρ_c=1000kg/m^3. Having this in consideration, using [Eq. 2.6] and doing some mathematical modifications (taking logarithms in both sides), Eq. 4.1 was obtained, from which the exponent α (Figure 4.4) was calculated as the slope of representing log(d_{32}/L) vs. log(*We*).

$$\log\left(\frac{d_{32}}{L}\right) = -\alpha \cdot \log(We) + \log(K_{10}) \qquad \text{[Eq.4.1]}$$

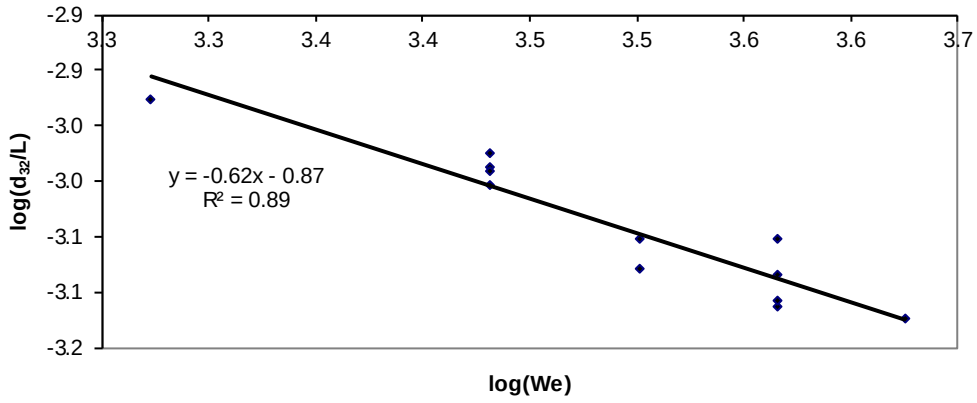


Figure 4.4. log(d_{32}/L) vs. log(*We*), calculation of α.

The value of the exponent of the Weber number obtained is -0.62, which is almost the same than the theoretical one (-0.6). This small difference in the exponent of the Weber number does not really improve the fitting (Figure 4.5), therefore the theoretical value has been selected to use in the equation. The data fit the theoretical model quite well, which means that the variation of the density and interfacial tension of the system in the interval of temperatures used is not significant, therefore the values measured at 2°C have been used in all the calculations.

Once α was determined and having considered that a constant volume fraction (ϕ) has been used in all the experiments, this value was included in the constant and [Eq.2.4] has been used to correlate the normalised Sauter diameter with the Weber number (Figure 4.5).

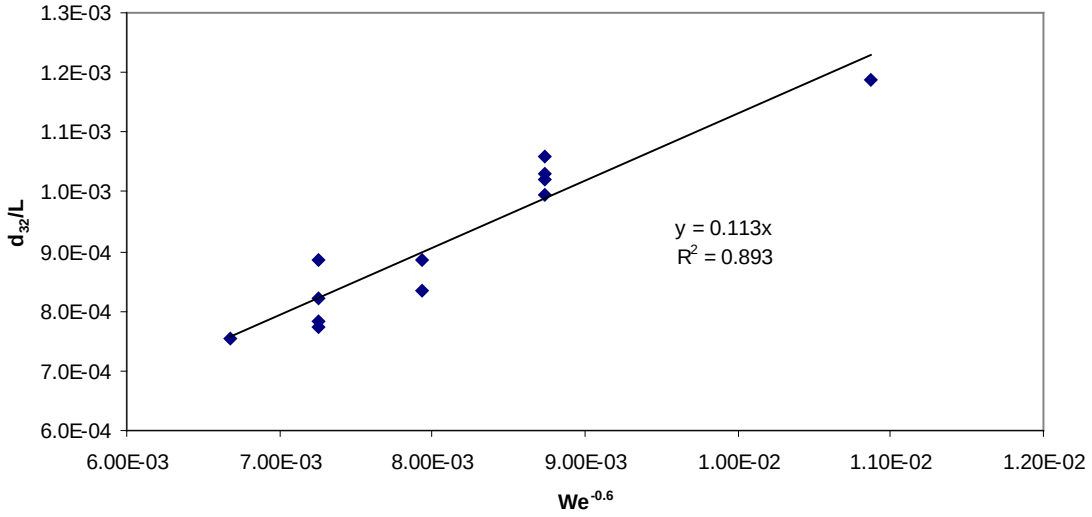


Figure 4.5. Normalised Sauter diameter vs. *We* number at the power of -0.6.

The final equation to calculate the Sauter mean diameter is [Eq.4.2]

$$\frac{d_{32}}{L} = 0.113 \cdot We^{-0.6} \qquad \text{[Eq.4.2]}$$

Or for comparative purposes, having considered the value of ϕ (0.167 in all the experiments), it is possible to express the results in[Eq.4.3]

$$\frac{d_{32}}{L} = 0.075(1 + 3\phi) \cdot We^{-0.6} \qquad \text{[Eq.4.3]}$$

Calabrese *et al*. (1986) also suggested an extended equation for non diluted systems [Eq.2.7]. In this case the Sauter diameter is correlated by:

$$\frac{d_{3}}{L} = 0.1 \cdot We^{-0.6} \cdot \left[1 + 0.0 \cdot We^{0.6}\left(\frac{d_{3}}{L}\right)^{\frac{1}{3}}\right]^{\frac{3}{5}} \qquad \text{[Eq.4.4]}$$

where:

Vi is the tank viscosity group: $V = i\left(\frac{\rho_c}{\rho_d}\right)^{0.5} \frac{\mu_d \cdot N \cdot L}{\sigma}$

ρ_d is the density of the dispersed phase
μ_d is the viscosity of the dispersed phase

[Eq.4.4] is an implicit equation and consequently more difficult to solve (the utility Solver from Excel was used to do it) than [Eq.4.2] and it does not improve the fitting of the experimental data to the theoretical model, therefore [Eq.4.2] is selected. In Figure 4.6 it is possible to see their fittings. [Eq.4.4] gives always a slightly lower value than [Eq.4.2]. The root mean square deviation ($\sigma_{rms)}$ of both equations has also been calculated (using [Eq.4.5]).

$$\sigma_{r\ m} = \left[\frac{1}{s} \frac{1}{n_D} \cdot \sum_{i=1}^{i=n_D} \left(\frac{d_{3\ E2} - d_{3\ P2}}{d_{3\ E2}} \right)^2 \right]^{\frac{1}{2}} \cdot 1 \qquad \text{[Eq.4.5]}$$

where:
d_{32E} is the experimental data for the Sauter diameter
d_{32P} is the predicted data for the Sauter diameter using [Eq4.2] or [Eq.4.4]
n_D is the number of experiments

With both equations the value obtained for the root mean square deviation is the same:

σ_{rms} = 4.7 %.

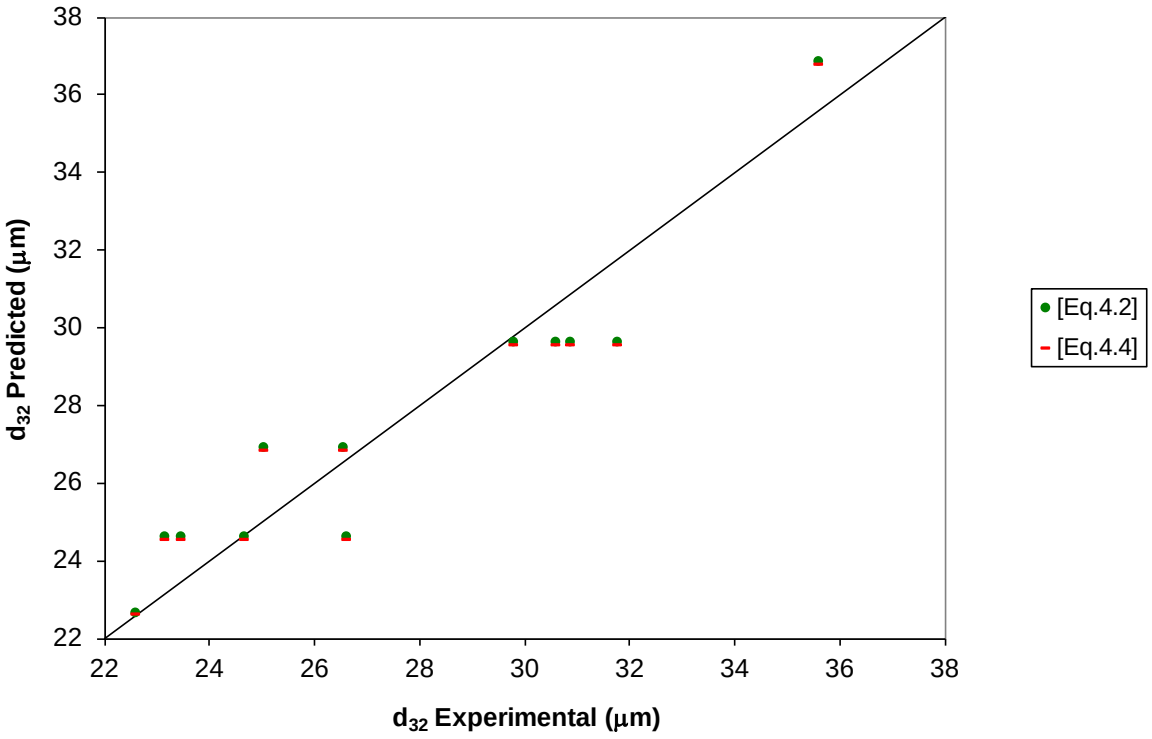


Figure 4.6. Predicted Sauter diameter using Eq.4.2 and Eq.4.4 vs. experimental values.

The value of the constant in [Eq.2.5] obtained by Calabrese *et al.* (1986) was 0.054, in contrast with the value obtained in this work in the equivalent equation ([Eq.4.3]), which is 0.075. This difference in the constant may be due to the different geometry used in the experiments. Calabrese *et al.* (1986) obtained the equation by working with a baffled cylindrical tank of standard geometry equipped with a six-blade Rushton turbine and this work has been done using a non-baffled cylindrical tank of standard geometry equipped with a Silverson turbine with a stator on the workhead.

4.4.2 Particle size distribution

It has been observed that the probability density function obtained using the system described in our experiments is a log-normal distribution (while Calabrese *et al.* (1986) obtained Normal distributions in their system).

[Eq.2.9] and [Eq.2.11] have been used to correlate the data obtained in the experiments and [Eq.4.6] and [Eq.4.7] have been generated after determining the parameters with Excel.

$$Fv\left(\frac{d}{d_{32}}\right)=\frac{1}{2}\left[1+erf\left(\frac{\ln\left(\frac{d}{d_{32}}\right)-1.11}{0.42\sqrt{2}}\right)\right] \qquad \text{[Eq.4.6]}$$

where:
Fv is the cumulative volume frequency
$\frac{d}{d_{32}}$ is the normalised diameter

Differentiation of Eq. 4.6 with respect to d/d_{32} gives

$$Pv\left(\frac{d}{d_{32}}\right)=\frac{1}{\frac{d}{d_{32}}0.42\sqrt{2\pi}}\cdot\exp\left[-2.8\left(\ln\left(\frac{d}{d_{32}}\right)-1.11\right)^2\right] \qquad \text{[Eq.4.7]}$$

where:
Pv is the probability density function for volume frequency.

In Figure 4.7 it is possible to see the experimental data fitted by Eq.4.6

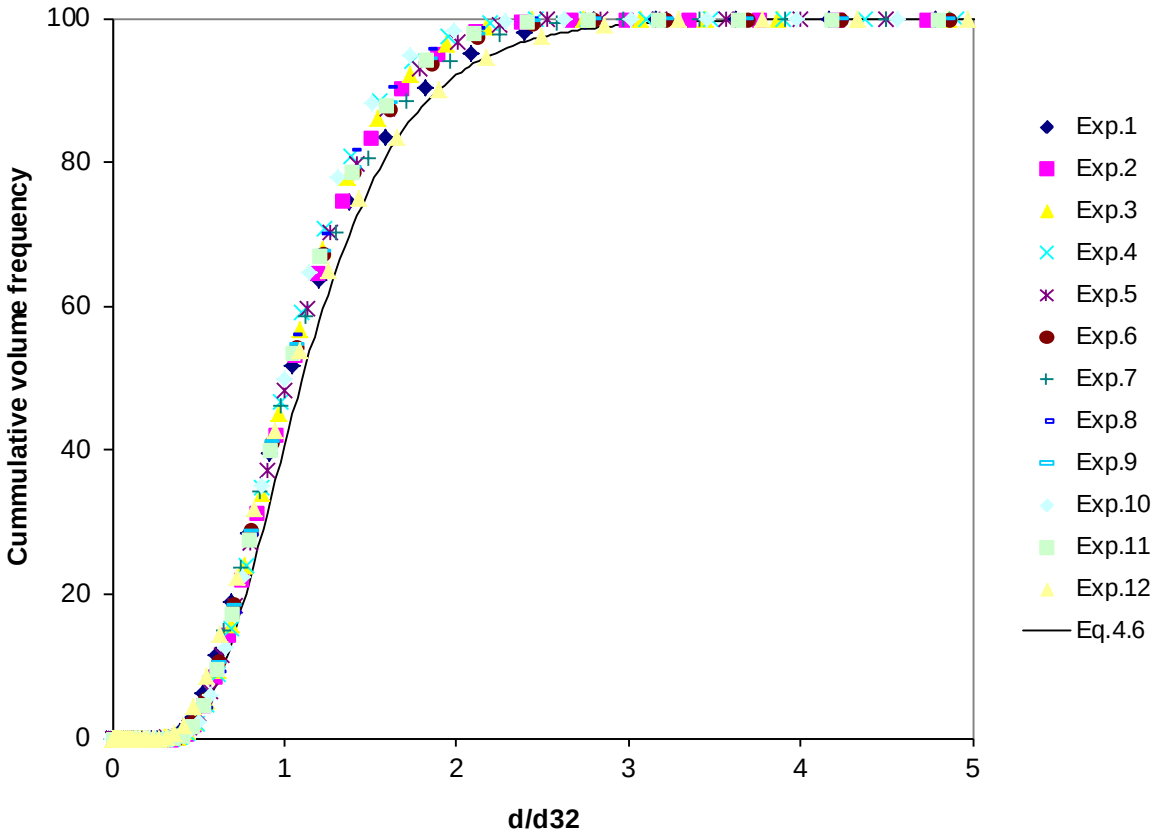


Figure 4.7. Cumulative volume frequency of the 12 experiments and Eq.4.6.

The results obtained from the Mastersizer based on the probability density function were compared with the correlation of Eq.4.7 (Figure 4.8). As can be seen, both are in good agreement.

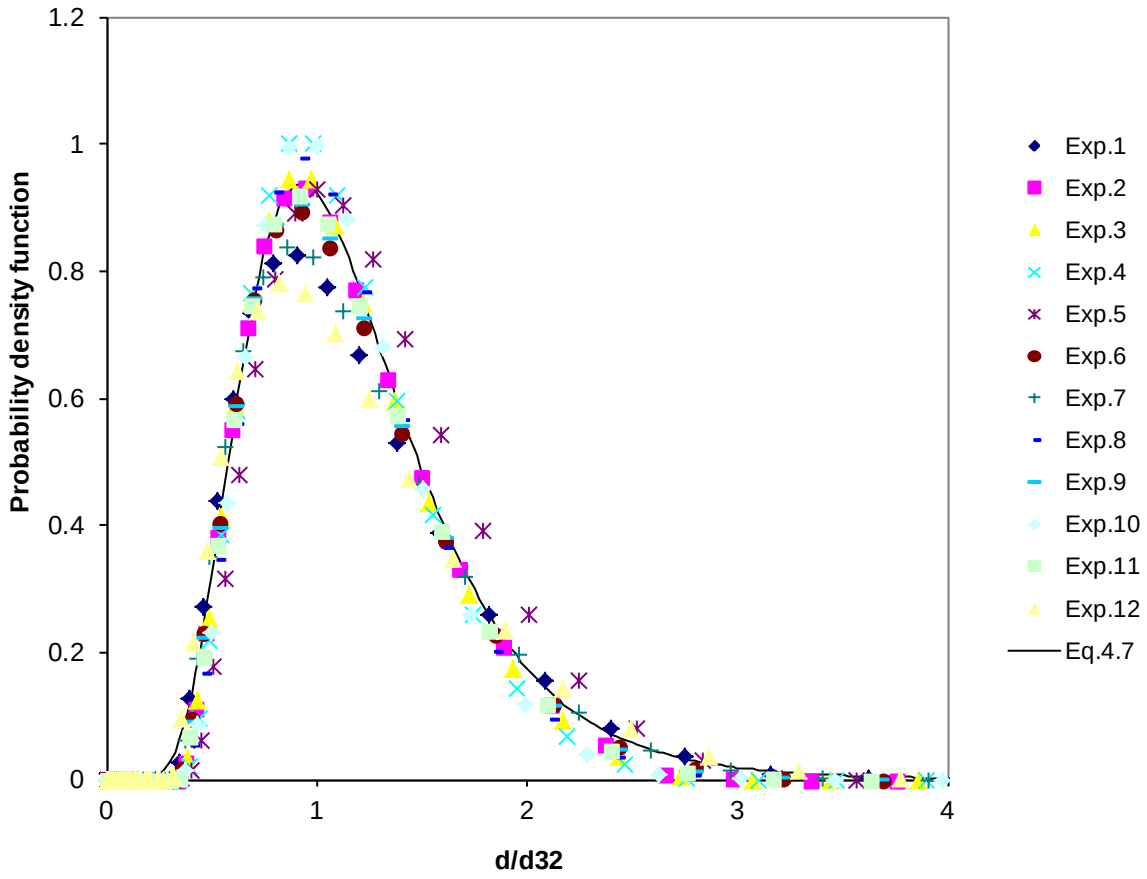


Figure 4.8. Probability density function of the 12 experiments and Eq.4.7

When a Silverson turbine was not used and the emulsion was prepared using a Rushton one in a baffled reactor, a log-bimodal distribution for the particle size was obtained instead of a log-normal distribution (see Figure 4.9).

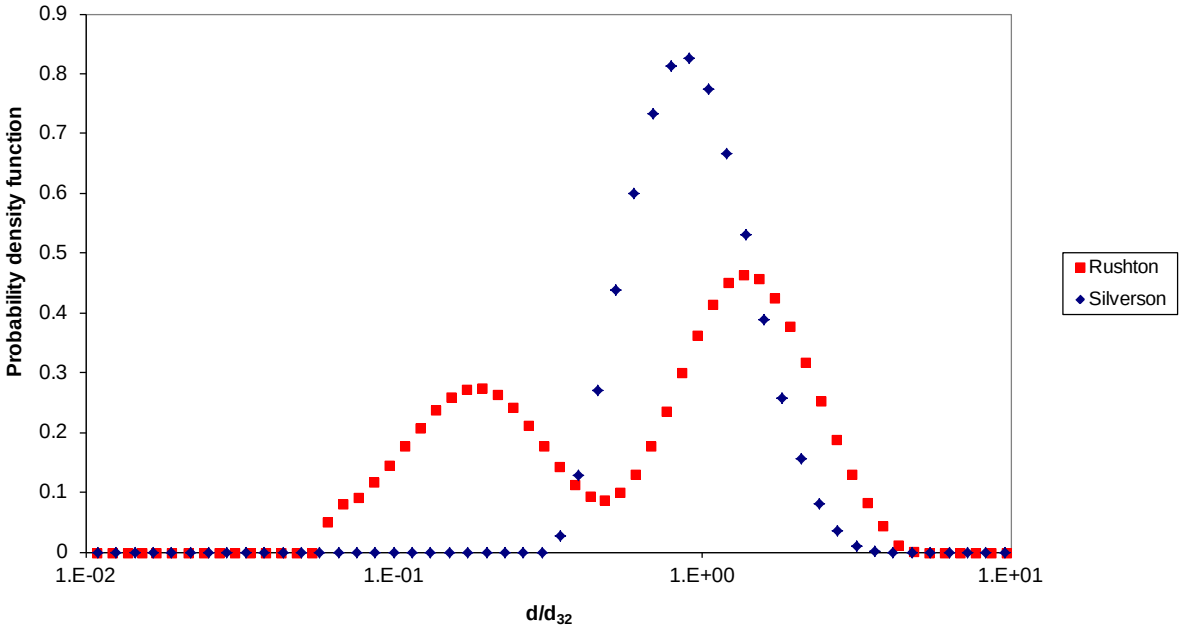


Figure 4.9. Probability density function of perfume microcapsules prepared with Silverson and Rushton turbines at 1000rpm.

4.5. Reaction kinetics

It is possible to identify 3 different rate-limiting stages during the process of capsule formation: Firstly the aqueous monomer diffuses from the continuous phase to the microcapsule's surface, secondly this monomer diffuses thought the shell already formed around the microcapsule and finally it meets the organic monomer and reacts with it forming polymer which accumulates growing the wall. The chemical reaction between the monomers is usually instantaneous (Arshady, 1989) and using enough aqueous monomer and turbulence in the system the diffusion of the monomer from the aqueous phase to the microcapsule's surface is usually much faster than the diffusion of the monomer thought the already formed membrane (once this membrane is formed, not in the first moments of reaction), which is the limitating step on the reaction.

The reaction of an acid chloride with an amine forms polyamide and delivers a molecule of hydrochloric acid to the media. Because of the consumption of the amines and the formation of hydrochloric acid, the pH of the system is reduced while the reaction is taking place. It is possible to monitor the reaction kinetics following the variation of pH with time. However it was not possible to take the first pH data (first pH data in all the graphs is the maximum pH measured by the pH meter, usually around 5 min after the amines were added) because the pH meter needs some time to stabilize a measurement and the pH (and also the temperature) is changing due to the reaction that is taking place, faster at the beginning of the reaction. The effects of the reaction temperature, the surfactant concentration and the aqueous monomer used were studied.

4.5.1. Effect of temperature

Four experiments have been done at different temperatures (0, 6, 12 and 18°C) using the same monomer concentrations. In all the experiments, trimesoyl chloride has been used as organic monomer and diethylenetriamine as aqueous one. A bath circulator connected to the reactor has been used to maintain a constant temperature. The pH was monitored during 6h and it was left overnight, the last pH data was taken next day, more than 22 hours after the experiment began. The experimental results are shown in Figure 4.10.

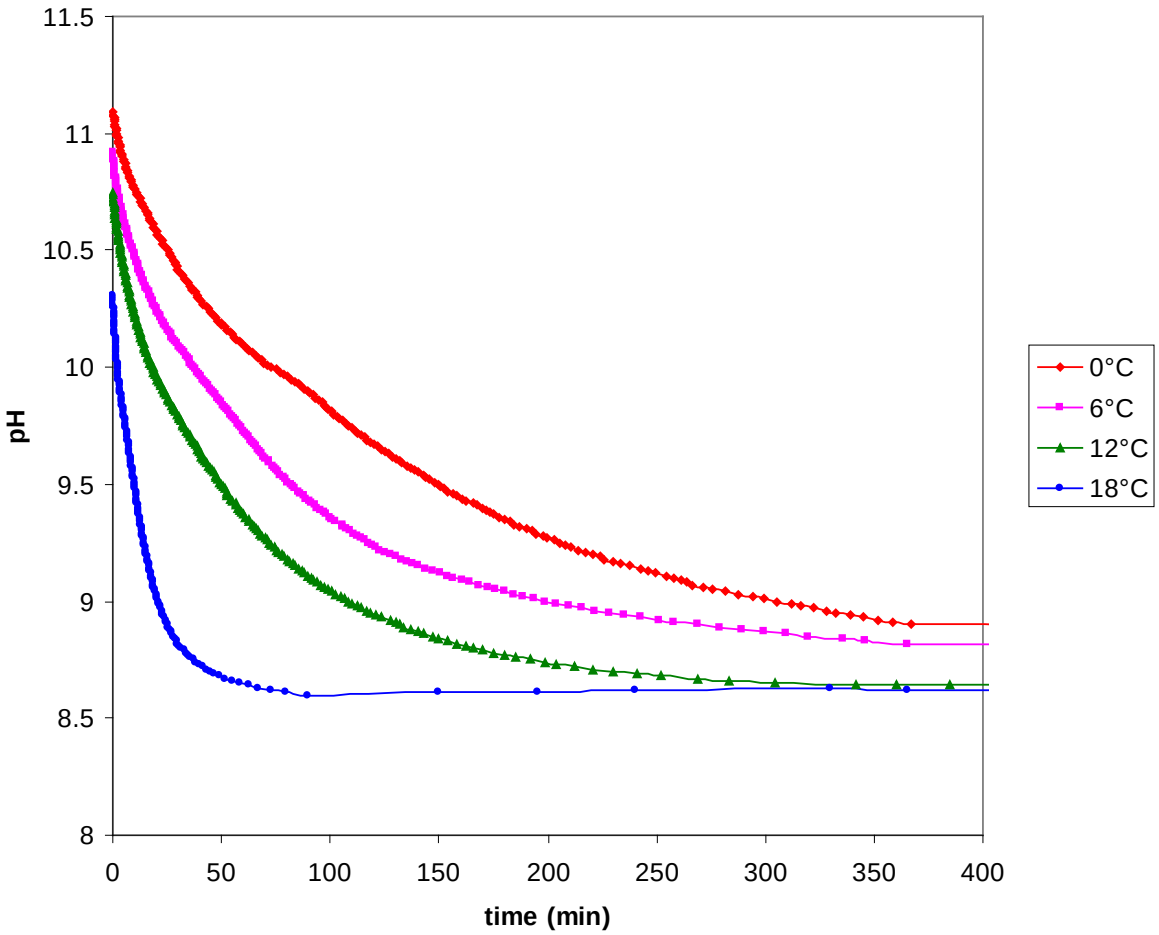


Figure 4.10. pH vs. time during the reaction at different temperatures.

It is possible to see that, as expected, the reaction is faster when the temperature is higher. At 18°C the reaction finishes before 100 min while at lower temperatures the reaction finishes much later. However as the initial and final pH is not the same in all the experiments (pH depends on the temperature), the advance of the reaction (% of pH change at a time over the total change of pH) with time is shown in Figure 4.11 to make comparison of the results easier.

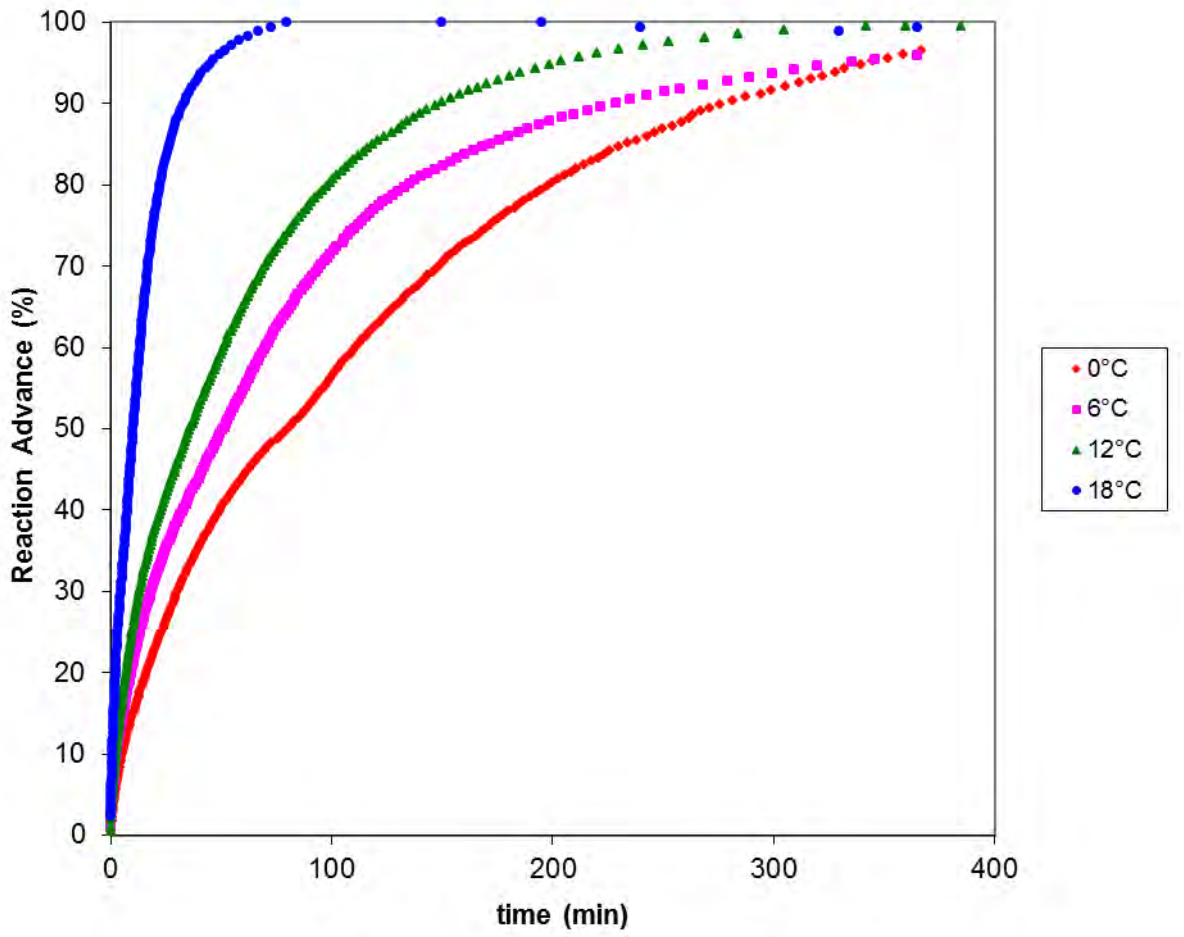


Figure 4.11. Reaction advance vs. time at different temperatures.

Figure 4.11 shows clearly that the higher is the reaction temperature, the faster is the reaction and the earlier it finishes. From the data, it is possible to see that the time needed for a 50% reaction is: 80.5, 51, 36 and 10.4min respectively; and for a 95% reaction: 345, 335, 203 and 46min at 0, 6, 12 and 18°C.

In the graphs presented the pH at the end of the reaction is different depending on the temperature of reaction, but this is merely an effect of the temperature at which the pH was measured. The pH for the four experiments was measured some days after the reaction at room temperature (22°C) and all of them had similar pH values: from 8.67 to 8.69.

4.5.2. Effect of particle size

While studying the particle size and size distribution of the microcapsules, some experiments were done with a higher concentration of surfactant in the system (5% instead of 1%). A higher surfactant concentration in the system produced smaller microcapsules (see Figure 4.12). The reaction kinetics of these experiments were also monitored and the results (Figures 4.13 and 4.14) show that the reaction rate for capsules made with higher surfactant concentration (smaller capsules) is slower than for capsules made with the lower surfactant concentration (bigger capsules).

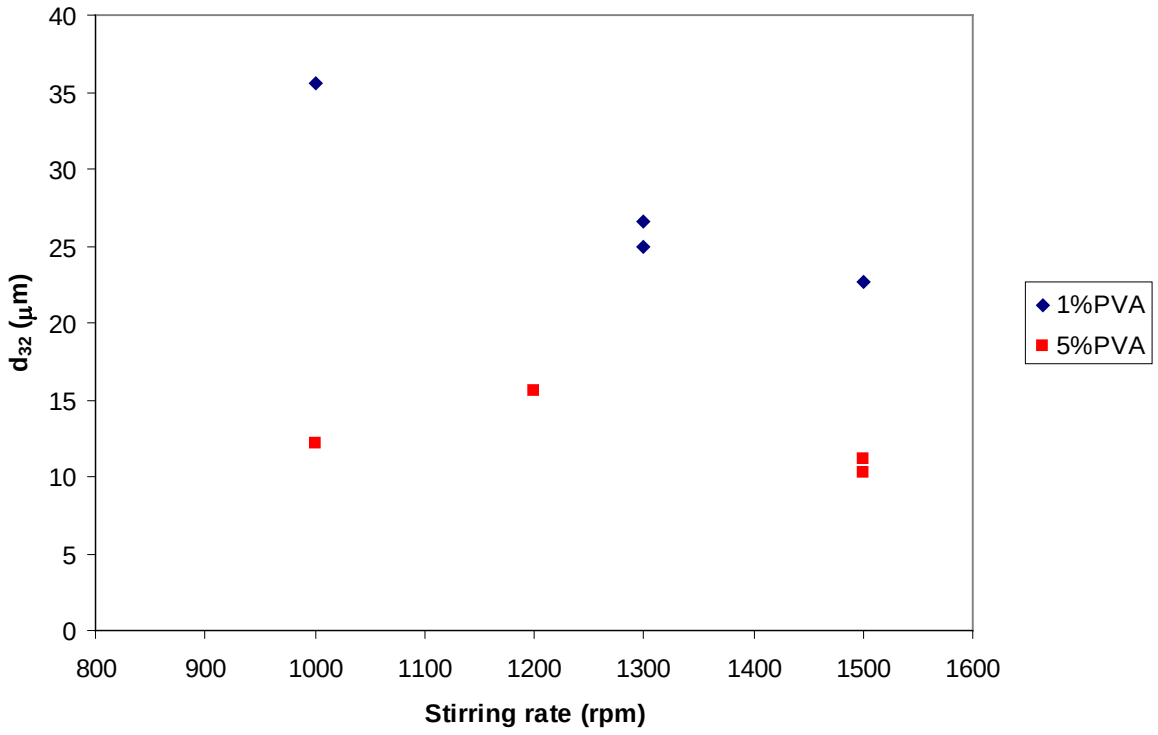


Figure 4.12. Influence of the surfactant concentration on the capsule size

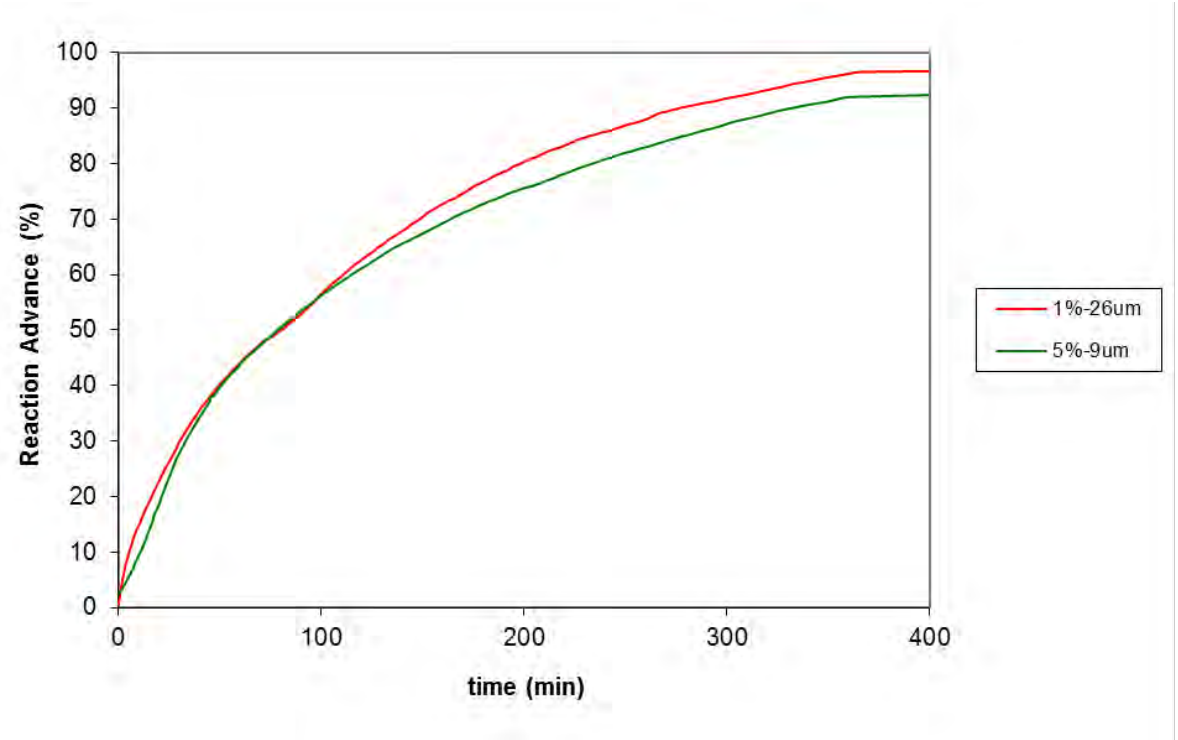


Figure 4.13. Reaction advance vs. time for different surfactant concentrations. Comparison for 1% PVA (0ºC) and 5% PVA (2°C)

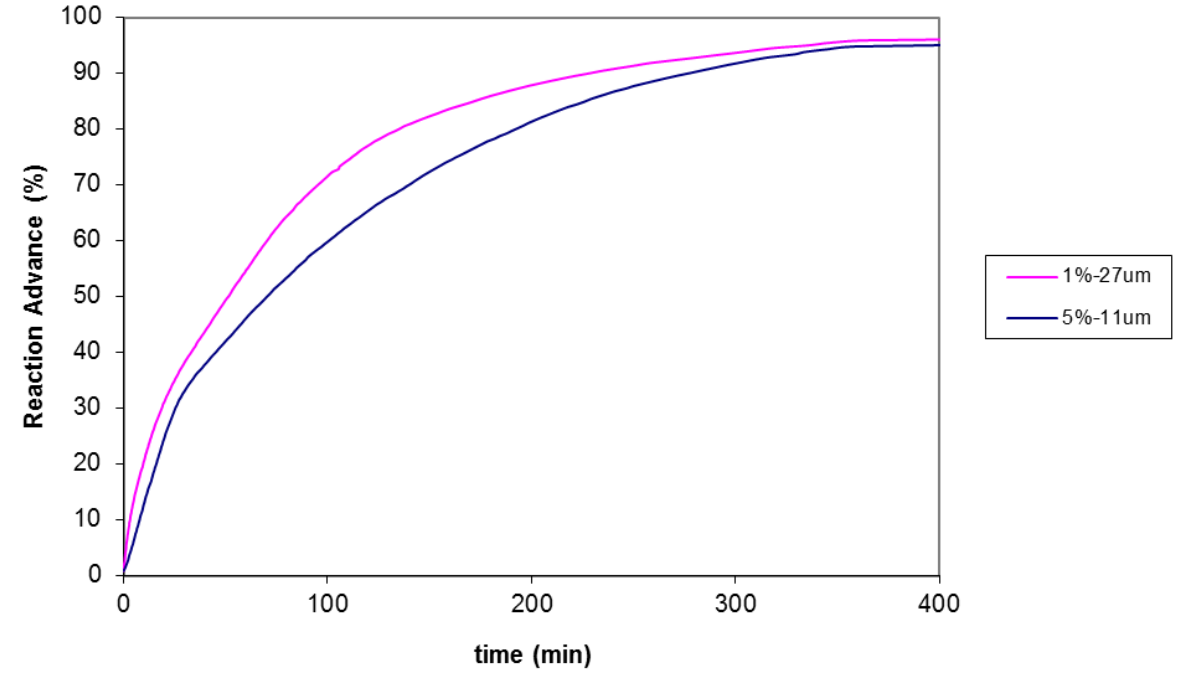


Figure 4.14. Reaction advance vs. time for different surfactant concentrations. Comparison for 1% PVA (6°C) and 5% PVA (7°C)

Figures 4.13 and 4.14 show the reaction rate is slightly faster for 1% PVA than 5% for similar experimental temperatures. It was expected that smaller capsules produced using 5% PVA might have faster reaction rate due to their larger surface/volume ratio which provides a better contact between the monomers. But it seems that a higher concentration of surfactant on the interface has created more difficulties for the aqueous monomer to diffuse from the continuous phase to the microcapsule's surface, which slowed down the reaction.

4.5.3. Effect of aqueous monomer

Three different monomers have been studied: diethylenetriamine, hexamethylenediamine and ethylenediamine. Equal amounts of NH_2 functions of each of them have been used in each experiment. The same organic monomer (trimesoyl chloride) concentration has also been used. The results are shown in Figure 4.15 (pH change) and Figure 4.16 (% Reaction advance).

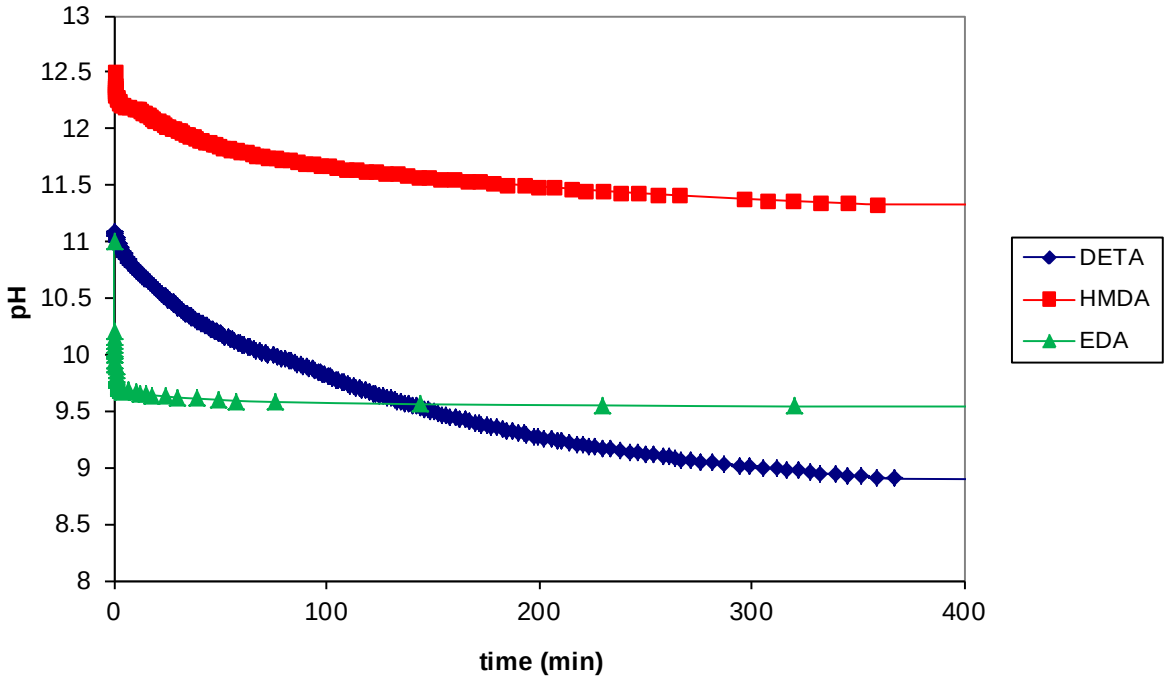


Figure 4.15. pH vs. time for different aqueous monomers.

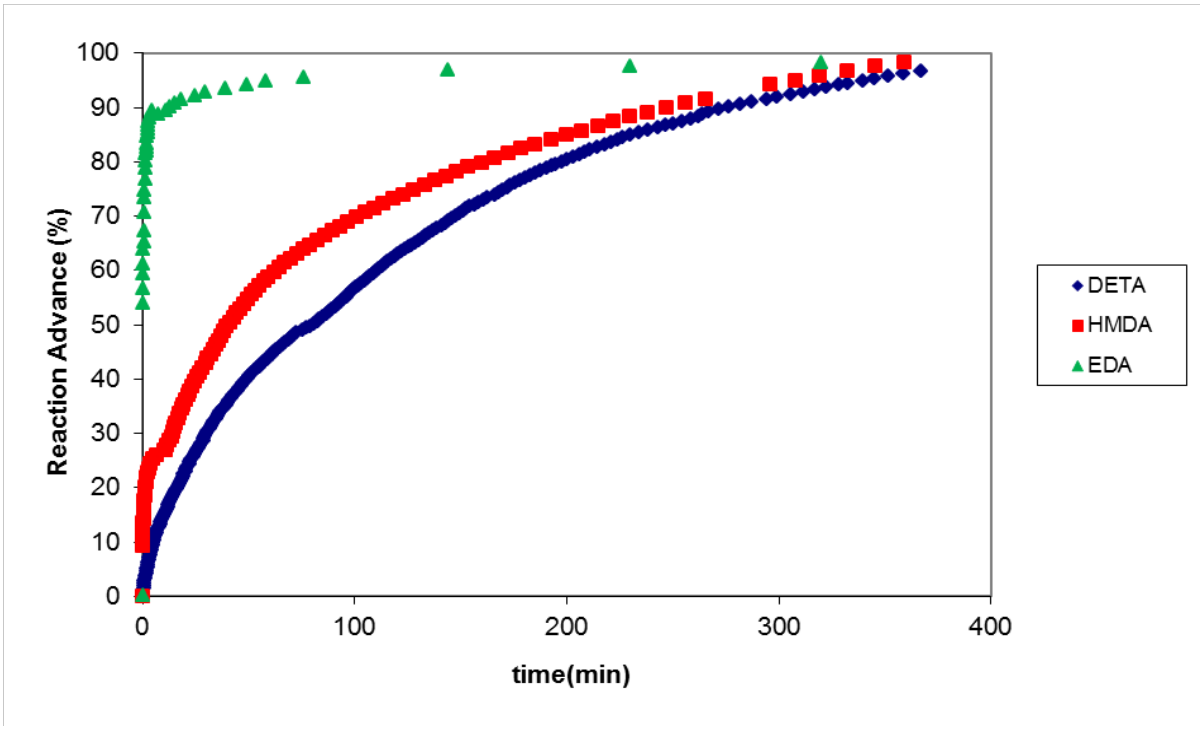


Figure 4.16. Reaction vs. time for different aqueous monomers.

Figure 4.16 shows that the polymerisation reaction when HMDA is used is faster than when DETA is used and it is almost instantaneous (90% of the reaction takes place in less than 5min) when EDA is used.

A qualitative way to measure the initial reaction kinetic is measuring the temperature increase when the aqueous monomer is added to the reactor. The polymerisation reaction is highly exothermic and the temperature of the reactor increases during the reaction. A jacked reactor connected to a bath circulator was used to maintain a constant temperature during the reaction but when the aqueous monomer was added to it the temperature increased faster than the cooling capacity of the circulator. Working at 0°C when DETA was added the temperature of the reactor increased about 2°C, when HMDA was added it increased about 3.5°C and when EDA was added in the reactor the temperature increased about 5.5°C.

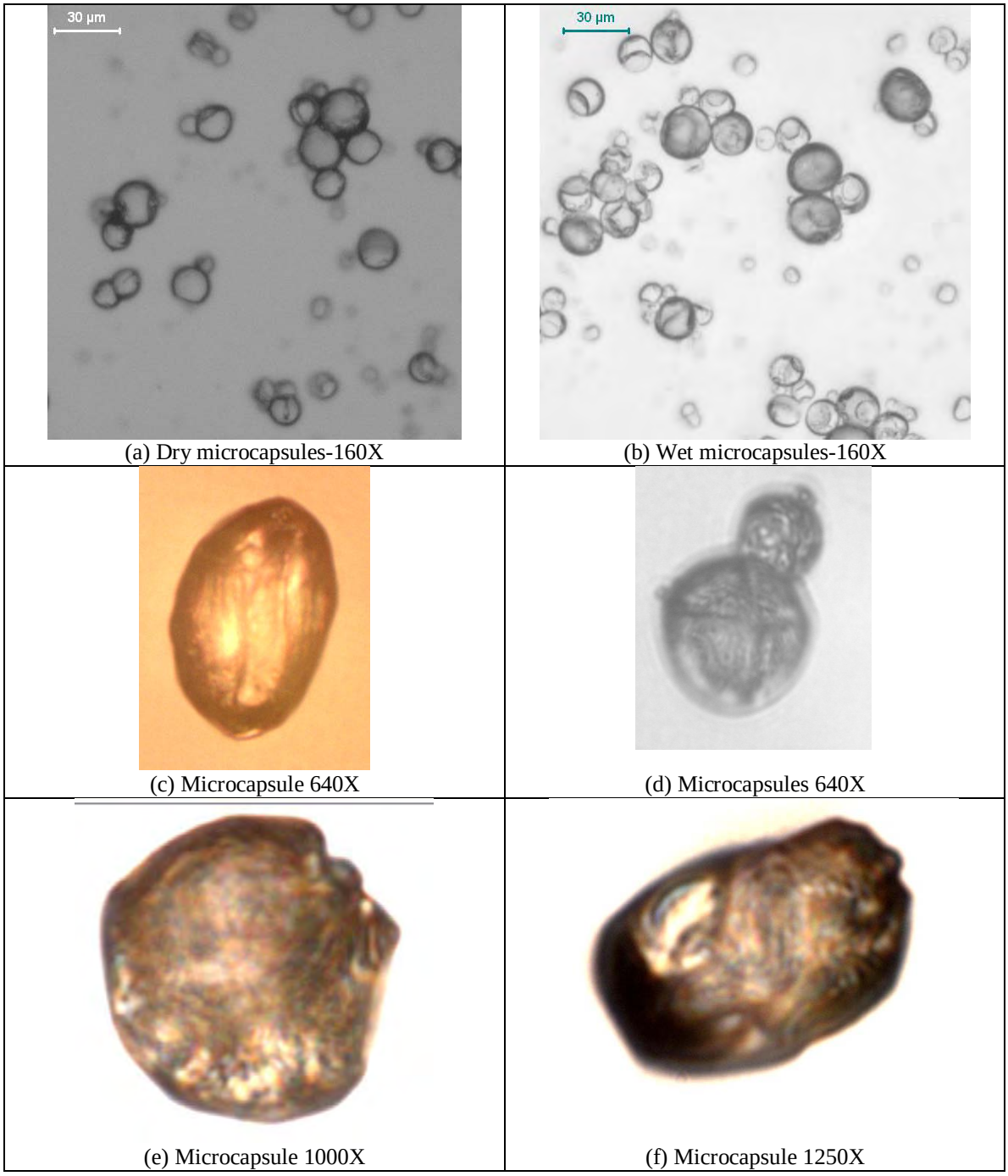
Another qualitative method to see the advance of the reaction is to see the change in colour of the reacting media. The perfume emulsion in water had a yellowish colour while the final

capsules suspension had a milky white colour. The time needed to observe a white colour in the reactor was much shorter when EDA was added to the system (almost instantaneous) than when any of the other monomers was used. It was also shorter when HMDA was used than when DETA was added to the reactor.

4.6. Wall properties

Perfume microcapsules made with different monomers have been developed. In the previous sections the importance of the selected monomers in the production process and experimental design has been shown. However the properties of the capsules may be very dependent on the monomers used in their production. In this section the relationship between the monomers used and the nature and thickness of the wall formed is presented.

Perfume-polyamide microcapsules with average diameter of approximately 25 microns have been produced. As remarked before, the reaction kinetics was very fast and the capsules had the shape of the drop which was being encapsulated. According to the images taken, the capsules were not completely spherical and they presented a surface not as smooth as capsules produced by *in situ* polymerisation (Hwang *et al.*, 2006; Su *et al.*, 2006b; Hong and Park, 1999). In Figure 4.17 it is possible to see some photographs of polyamide-perfume microcapsules taken with different magnifications.



(a) Dry microcapsules-160X

(b) Wet microcapsules-160X

(c) Microcapsule 640X

(d) Microcapsules 640X

(e) Microcapsule 1000X

(f) Microcapsule 1250X

Figure 4.17. Optical microscope photographs of perfume-polyamide microcapsules taken with different magnifications (160-1250X).

Photographs (a) and (b) show that the capsules are stable in wet and dry conditions, contrary to previous capsules prepared with polyester (Figure.4.2). Capsules seem to be round although some of them present irregular shape. It is possible to see scrapes on the capsule

walls in photos (c) and (d). Some microcapsules are not completely spherical, they present a shape more like a coffee bean (photos (c) and (f)) and the surface is not smooth and regular, which presents some scrapes, protrusions and valleys (photos (c), (d), (e) and (f)).

SEM photomicrographs have been also taken (Figure 4.18). To take them, a sample of microcapsules was placed in the chamber of SEM and vacuum was generated. The capsules shrank in those conditions and the walls formed structures with clear edges (micrographs (a) and (d)). It is possible to see also that the surface walls seem to be porous (micrographs (b), (c) and (d)).

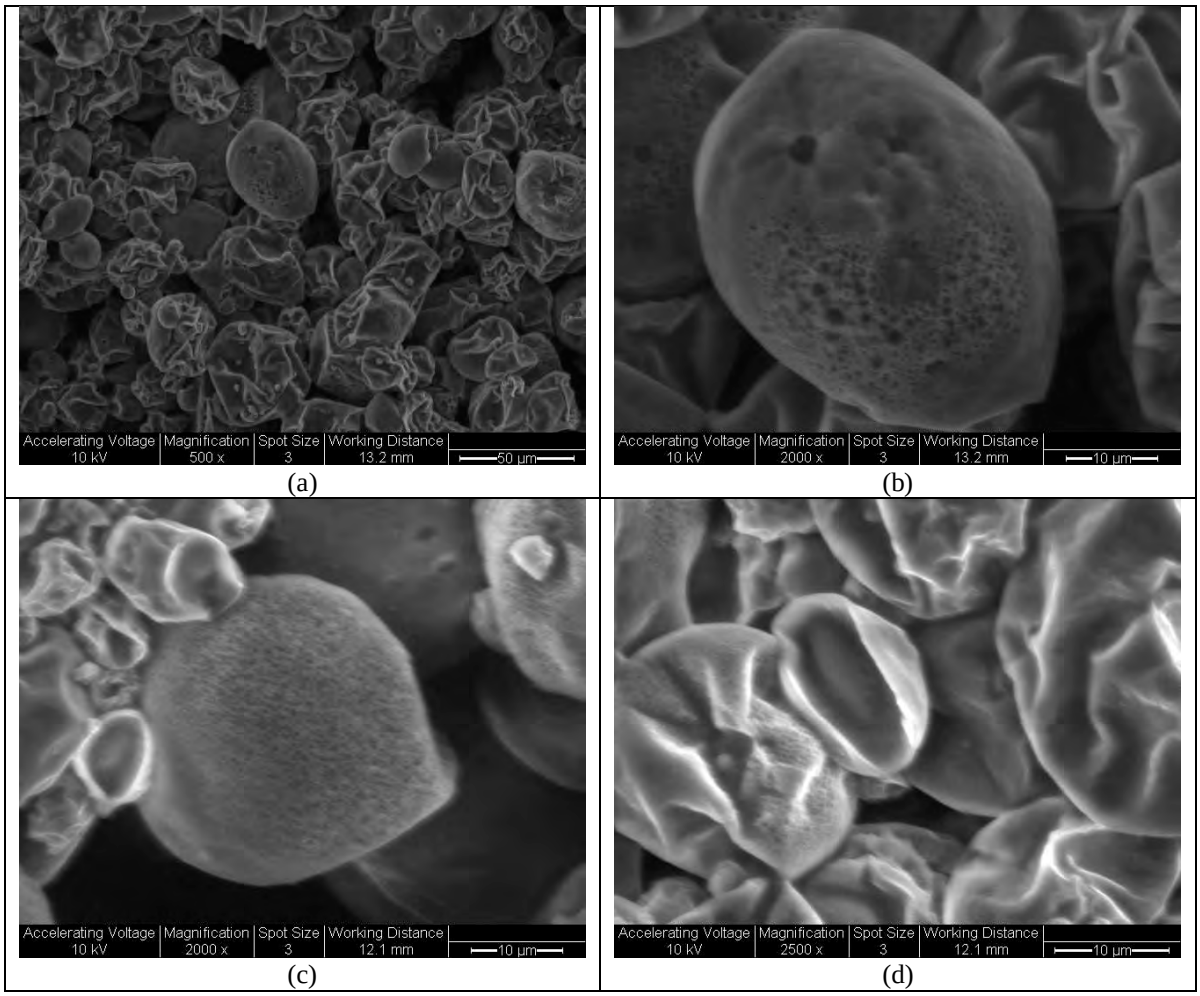


Figure 4.18. SEM micrographs of perfume-polyamide microcapsules. Scale bar is 50µm in micrograph (a) and 10µm in (b), (c) and (d).

4.6.1. Chemistry

An FTIR instrument has been used to study the chemical structure of the polymer formed. Samples of different batches (DETA, EDA, HMDA, TC and All50 - details in Table 4.1) were prepared: capsules were broken using glass beads under magnetic stirring and perfume was extracted with hexane, after that the polymer obtained was dried in an oven at 40°C for 30min. The solid polymer was placed on the FTIR and measured directly. The spectra of pure EDA and DETA (liquid) is shown in Figures 4.19 and 4.20. Results of the polyamides are presented in Figure 4.21, 4.22, 4.23, 4.24 and 4.25.

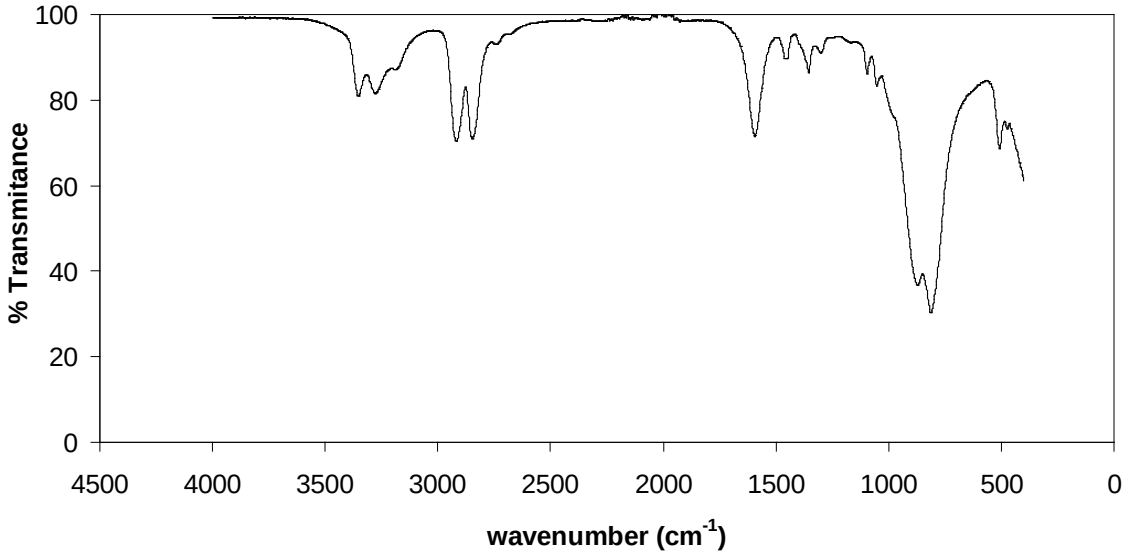


Figure 4.19. FTIR spectra of pure EDA monomer

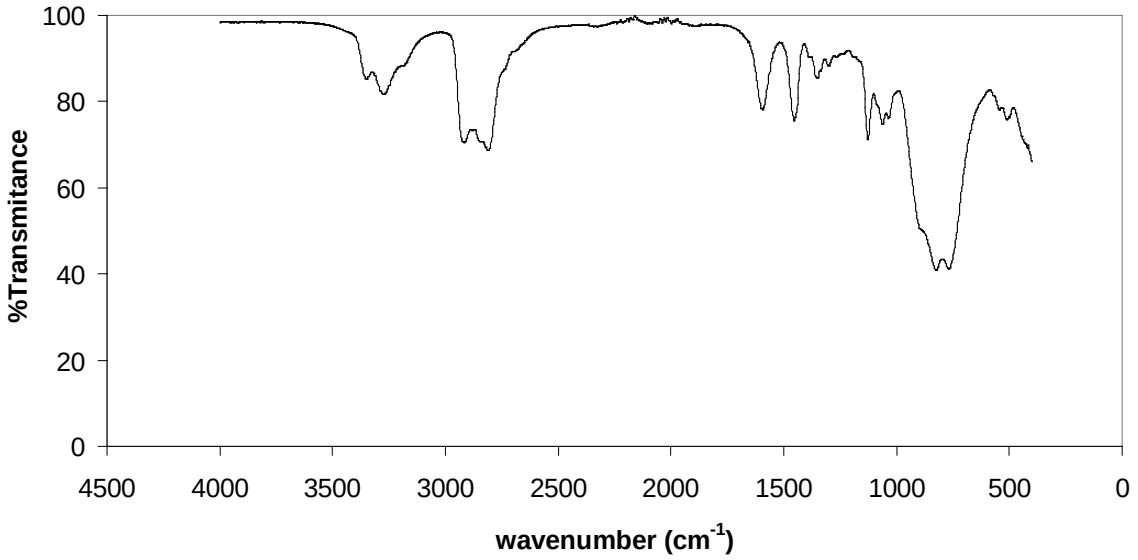


Figure 4.20. FTIR spectra of pure DETA monomer

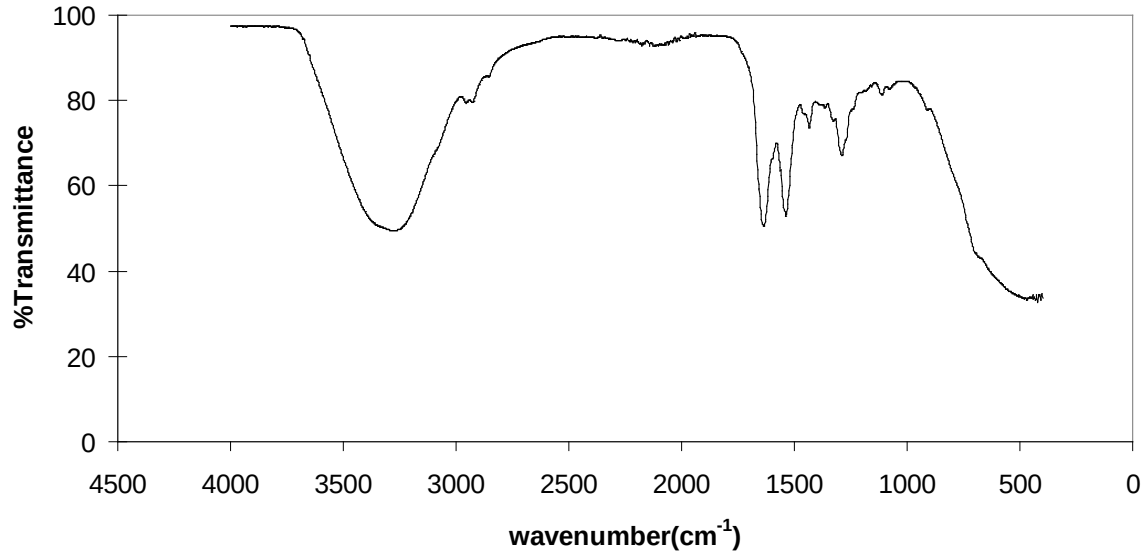


Figure 4.21. FTIR spectra of polymer formulation EDA

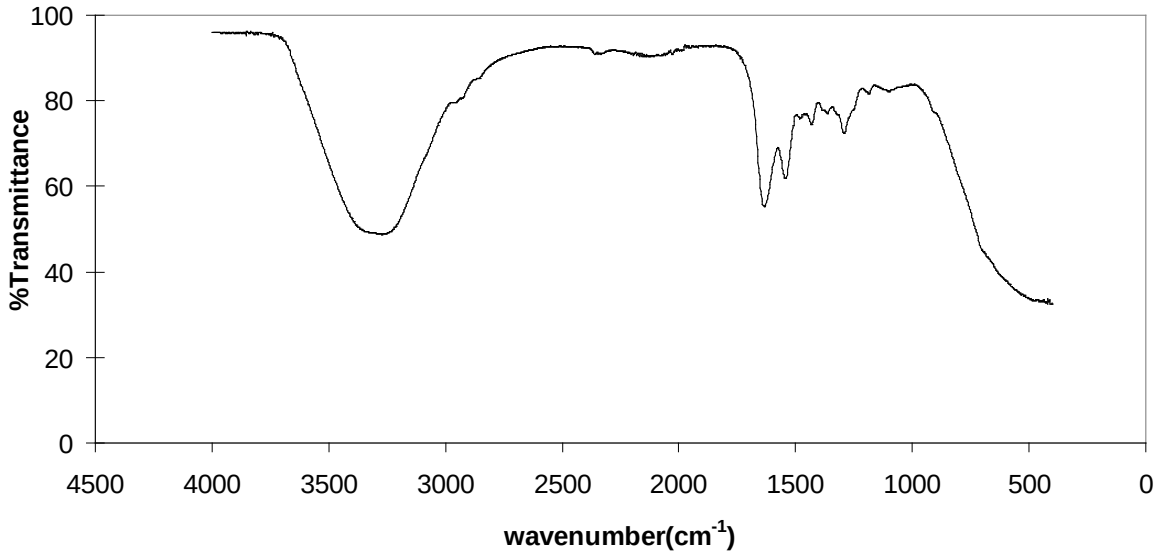


Figure 4.22. FTIR spectra of polymer formulation DETA

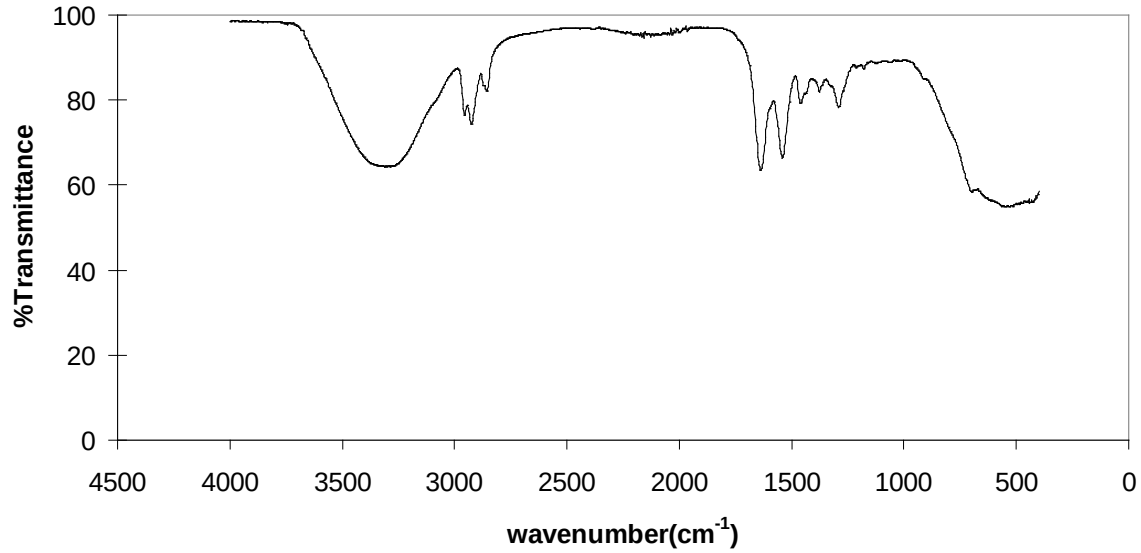


Figure 4.23. FTIR spectra of polymer formulation HMDA

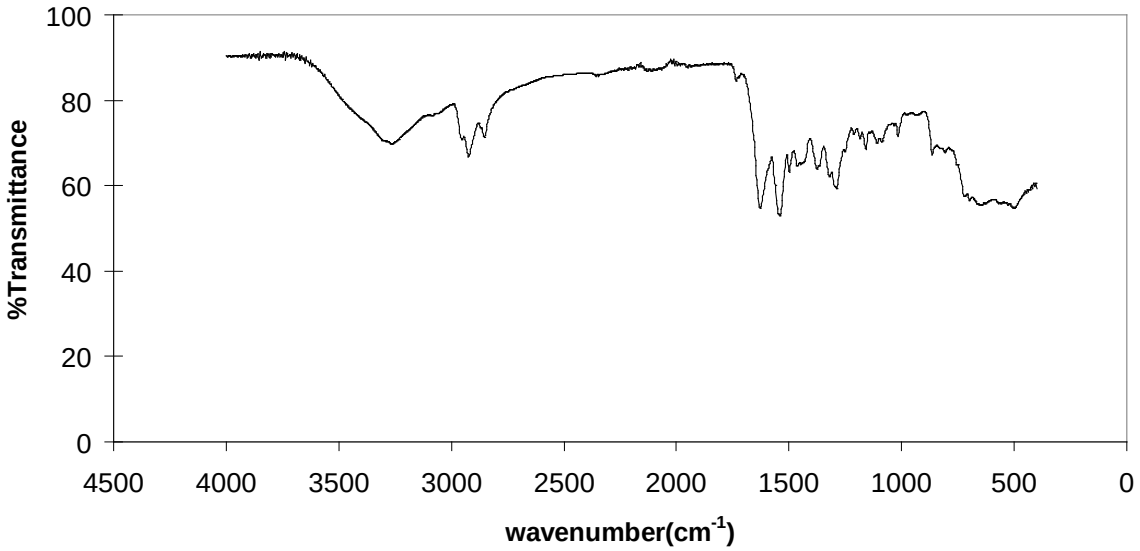


Figure 4.24. FTIR spectra of polymer formulation TC

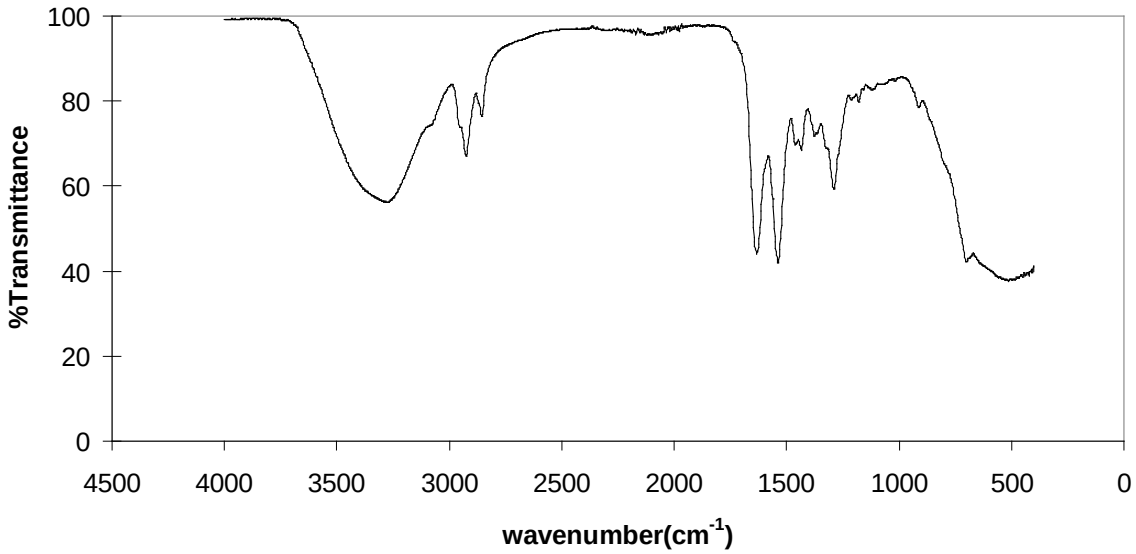


Figure 4.25. FTIR spectra of polymer formulation All50

To interpret this data, first it is needed to describe what the peculiarities of the amines and amides spectra are (Mathiowitz and Cohen, 1989a; Forrest *at al.*, 2007; Haynes, 2011): The NH_2 stretching vibration of both primary amines and amides gives an intense doublet between 3500 and 3170 cm^{-1}. The N-H stretching vibration of secondary amines and amides only gives a single strong band in this interval. Tertiary amines and amides do not give N-H absorption

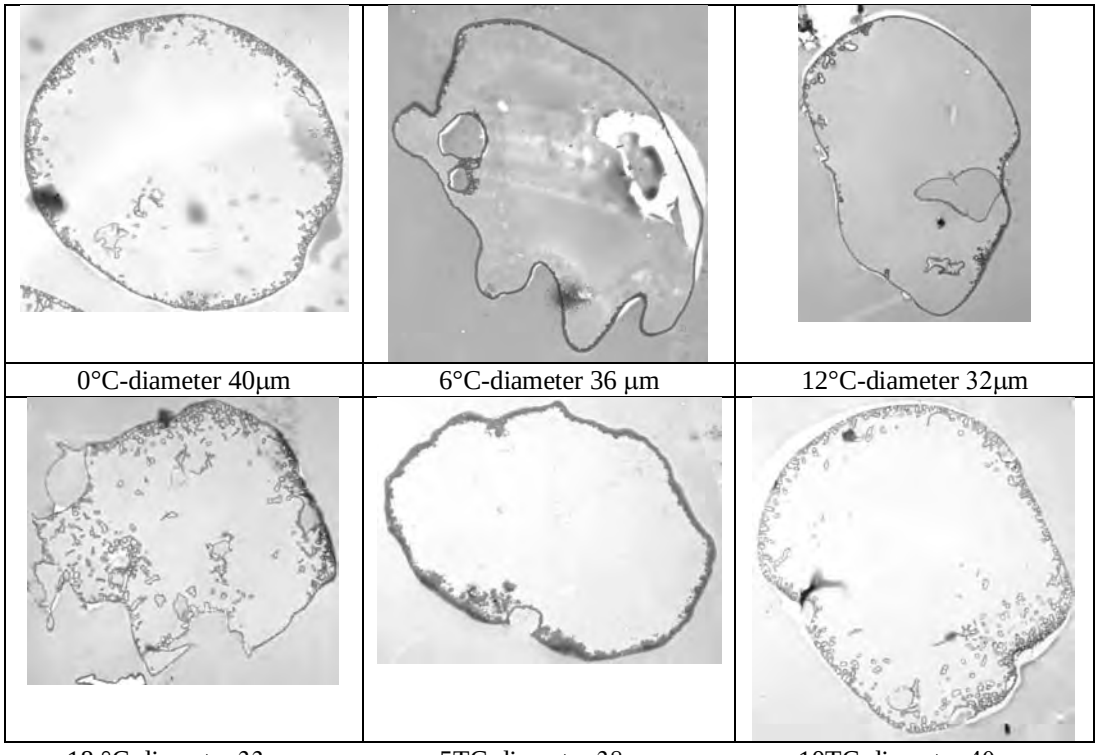
band. Amide spectra is characterised by two strong bands, called Amide I and Amide II. Amide I band is due to C=O stretching and it appears at 1665 $\pm$ 30 cm^{-1}. Amide II band is due to N-H bending trans to the carbonyl oxygen and appears at 1620 ± 30 in primary amides and at 1530 ± 30 in secondary amides. In addition it is possible to identify other peaks in the figures: C-H stretching bands appear between 2800 and 3000 cm^{-1} and a weak C-N stretching band appears at 1430cm^{-1}.

From the shapes and intensities of the observed bands it is possible to extract several pieces of information. First, it is possible to note that after the capsules have been formed the doublet associated with the NH_2 bond of the amine monomer (Figure 4.18 and 4.19) disappeared and it was replaced with a single band (Figures 4.20, 4.21, 4.22, 4.23 and 4.24). This means that little or no unreacted NH_2 was present in the final polymer. However, due to the breadth of the N-H absorption band it is not possible to conclude that none is present in the final polymer.

Mathiowitz and Cohen (1989a) observed weak bands at 1770 cm^{-1} in polymers prepared with trimesoyl chloride, and those weak bands were associated with the presence of unreacted acid chloride groups. There was no presence of these bands in the microcapsules prepared with trimesoyl chloride in this work, but a very weak band appeared in the capsules prepared with terephthaloyl chloride, the bifunctional aromatic organic monomer (Figure 4.23). The presence of acid chlorides in the microcapsules and membranes produced by Mathiowitz and Cohen were justified with the increased crosslinking of the walls produced by the trifunctional monomer, but in this work they appeared when the bifunctional monomer was used.

4.6.2. Thickness

Samples of most experiments have been prepared to be observed using Transmission Electron Microscopy (TEM - sample's preparation is shown in Chapter 3). Some micrographs have been taken from each sample and the thickness of the microcapsules has been measured using image analysis software. The micrographs show that the microcapsules were affected by the sample preparation: some of them were deformed, others were broken and some samples were completely destroyed during the preparation step. Nevertheless, some images of microcapsule's sections can still be seen in Figure 4.26.



0°C-diameter 40μm	6°C-diameter 36 μm	12°C-diameter 32μm
18 °C-diameter 33μm	5TC-diameter 38μm	10TC-diameter 40μm

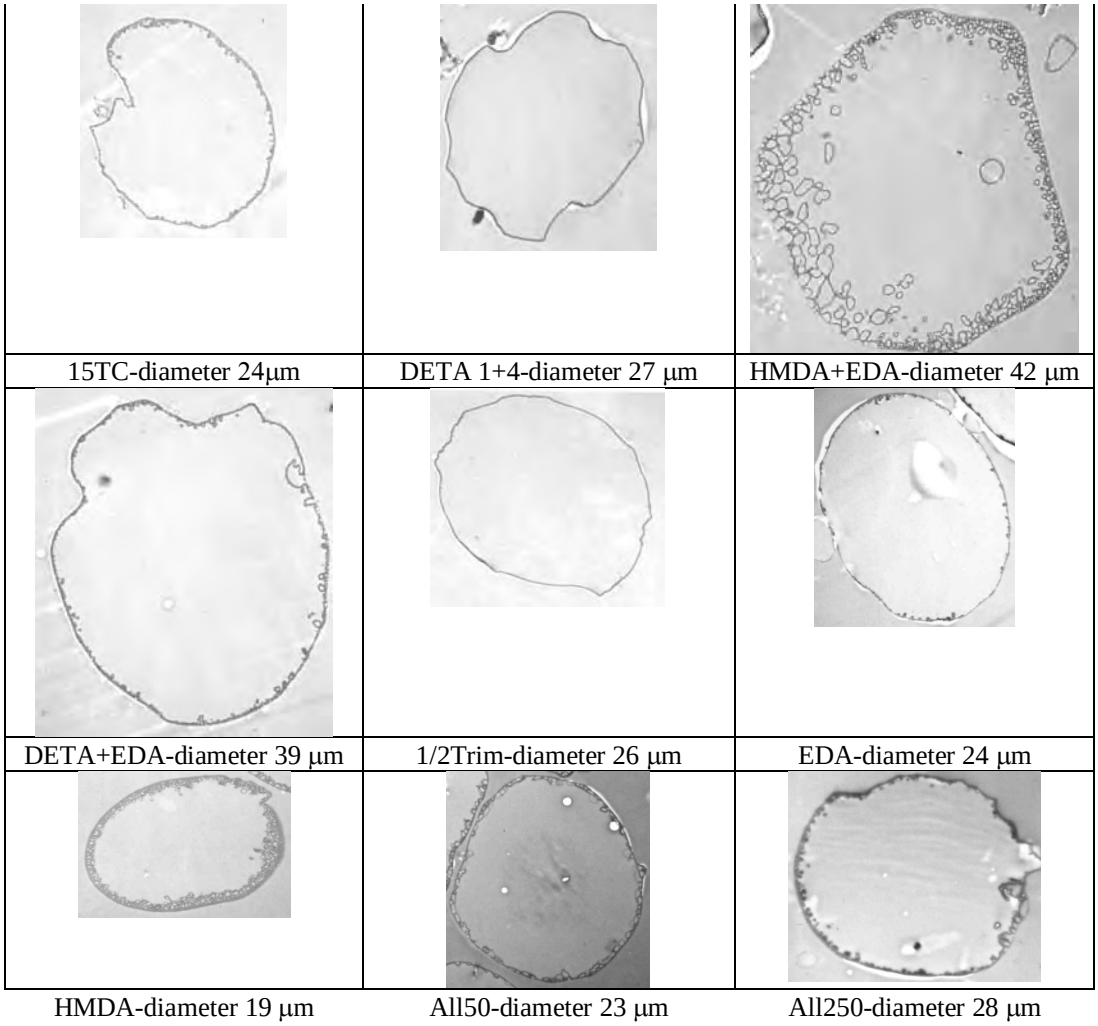


Figure 4.26 Microcapsule's sections of different formulations.

During the preparation of the samples for TEM, the liquid inside the capsules was removed (see sample preparation in Chapter 3) and the solid part (the wall) fixed with a resin before being sliced with a microtome. That means that the more or less round and dark line in the micrographs is the wall of the capsules. From the micrographs it is possible to see that the reaction took place inside the capsule (the wall outside is much more smoother), the membrane grew from the inside part (organic phase) similar to previous works (Mathiowitz and Cohen, 1989a). It is possible to observe a different shape and structure of the wall depending on the formulation and temperature of the reaction.

- Capsules made at low temperature (0°C) maintained the shape of the capsule much better than capsules made at higher temperatures (18°C), the latter of which seemed to be much more sensitive to the preparation. That might be related to the higher mechanical resistance of the capsules made at the lower temperature, which will be verified by micromanipulation measurements of their mechanical strength later.
- When terephthaloyl chloride (TC) was added to the reaction media a thicker wall was obtained. When 10% of TC was added (experiment 5TC) a homogeneous compact thick wall was obtained but when 18% TC was added (experiment 10TC) many small polymer bubbles were formed inside the capsule.
- When less organic monomer was used (experiment 1/2Trim), a thinner wall was obtained. Not all the capsules survived the preparation process and the ones who did it presented breakages in the wall.
- The biggest difference in the structure of the capsule walls was observed when different aqueous monomers were used:
    - EDA formed a very thin membrane wall and capsules made with it did not survive very well the preparation step. Many capsules were found to show ruptures in the membrane.
    - HMDA formed a thick membrane wall with many big bubbles of polymer growing from it. The thickness of the membrane was much larger but there was organic phase entrapped in it. Some aggregation between microcapsules was observed.
    - DETA formed a membrane wall with small bubbles of polymer growing from it. The thickness of the membrane was much smaller than the ones made with HMDA.

- Capsules formed with TETA did not survive the preparation process.

- When a combination of all the monomers was used, the capsules produced presented a structure between the one using DETA and the one using HMDA alone as described before, with larger bubbles of polymer when the HMDA/DETA ratio is higher, but smaller than when using HMDA alone and only one or two layers of them.

The micrographs also showed that the wall thickness was not homogeneous, therefore it has been measured at 5-10 points per microcapsule, and the minimum and average thickness for each microcapsule has been calculated. In Table 4.3, the minimum and average thicknesses of the different batches studied are shown, based on a number of microcapsules from 2 to 13 measured from each batch. From the measurements done it is possible to say that for any batch the wall thickness of the microcapsules in that batch is independent of the diameter of the capsule, with the minimum thickness almost constant or very similar in most of the batches. However due to the different morphology of the walls there are big differences in the average thickness between different batches.

Batch name	Minimum thickness(nm)	Average thickness(nm)	Average diameter (μm)
½ Trim	160 ± 23	243 ± 14	28 ± 2.0
18°C	107 ± 13	249 ± 21	34 ± 2.9
EDA	171 ± 7	267 ± 17	24 ± 3.2
All50	206 ± 47	324 ± 52	26 ± 4.4
12°C	120 ± 23	336 ± 37	37 ± 2.9
DETA1+4	200 ± 0	340 ± 24	32 ± 2.8
15TC	173 ± 10	346 ± 60	26 ± 0.5
10TC	187 ± 13	431 ± 7	41 ± 0.6
6°C	187 ± 13	443 ± 6	38 ± 2.2
All250	264 ± 24	450 ± 70	19 ± 3.3
HMDA	218 ± 17	465 ± 25	21 ± 1.8
DETA+EDA	187 ± 13	502 ± 123	34 ± 6.2
0°C	147 ± 27	566 ± 101	40 ± 0.3
HMDA+EDA	173 ± 13	607 ± 71	36 ± 6.0
5TC	400 ± 0	741 ± 37	42 ± 3.6

Table 4.3. Thickness of polyamide-perfume microcapsules.

The results show the influence of the temperature of reaction and the monomer composition on the structure of the wall (the batch "0°C" has been taken as reference for the comparisons):

- For the four experiments done using the same monomer composition at different temperatures (0, 6, 12 and 18ºC), the average thickness of these 4 samples increased as temperature decreased.
- The use of the bifunctional organic monomer in addition to the trifunctional one increased the thickness of the wall when it was used in small amounts (5TC), and reduced the average thickness (but increased the minimum one) when it was used in larger amounts (10TC and 15TC). The average thickness of the microcapsules was reduced when the amount of bifunctional monomer used was increased from 10 to 18 and 25%.
- The use of EDA as aqueous monomer reduced (in comparison with DETA) the average wall thickness. The thickness of microcapsules made with HMDA was not easy to measure due to the morphology of the wall: there were many layers of polymer "bubbles" next to the membrane but only the first membrane has been measured. Microcapsules made with HMDA had a larger minimum thickness and a slightly smaller average one.

4.7. Conclusions

Perfume has been successfully encapsulated using polyamide as a wall material. The use of polyester was also studied but the microcapsules obtained were very weak and broke when they were dried.

Microcapsules using different monomer formulations and reaction conditions have been produced. The effect of the temperature of reaction and the influence of the monomer concentration on the final properties of the microcapsules have been studied.

The **temperature of reaction** has a big influence on the final properties of the capsules. It was only possible to obtain stable capsules when the reaction temperature was below 18-20°C, when trimesoyl chloride and diethylenetriamine monomers were used, and lower temperatures were required for other aqueous monomers. Microcapsules have been produced at 4 different reaction temperatures (0, 6, 12 and 18 °C).

The **monomer type and concentration** used in the formulation of the microcapsules has been found to have a big influence on their final properties. Three different organic monomers and four different aqueous ones have been used. It has been found that when sebacoyl chloride (aliphatic) was used as organic monomer no capsules were produced, only when aromatic organic monomers were used stable microcapsules were formed. It has also been found that a minimum concentration of organic monomer in the system was needed to produce capsules. Several formulations have been prepared for their characterisation.

The **size and size distribution** of the microcapsules produced have been correlated to theoretical models and the parameters in these equations have been calculated. The Sauter mean diameter measured from the different experiments is well correlated by the Hinze and Kolmogoroff model. The size distribution data taken from the experiments approached a log-normal distribution when a Silverson homogenizer was used. When a Rushton turbine was

used the size distribution obtained approached a log-bimodal distribution. A Silverson turbine is preferred to a Rushton one.

The **reaction kinetics** was highly influenced by the temperature of reaction, the aqueous monomer used and the concentration of the surfactant in the system. As expected the kinetics was faster when the temperature was higher and when the aqueous monomer used was smaller (follows the order: $k_{EDA}>>k_{HMDA}>k_{DETA}$) as it is easier for the monomer to cross the membrane to react at the organic side of it. It has been found that the kinetics was faster when the surfactant concentration in the system was lower (also leading to bigger microcapsules). It seems that the presence of a higher concentration of surfactant at the interface made the monomer diffusion more difficult.

An FTIR instrument has been used to identify the **chemistry of the wall**. Results show that no NH_2 remained unreacted and that an amide bond has been formed during the reaction. A polyamide has been produced.

Different **microscopy techniques** have been used to study the microcapsules produced. Microcapsules looked not completely spherical when an *optical microscope* was used. When an *SEM microscope* was used, microcapsules shrank due to the vacuum produced by it, they showed edges and not a very smooth surface and it was possible to see some pores on the walls. Micrographs taken using a *TEM microscope* permitted to see the inner structure of the wall of the microcapsules produced and to measure their wall thickness. The morphology of the wall was very different depending on the aqueous monomers used in the reaction. EDA

produced a thin and smooth wall, DETA produced a thicker one and HMDA produced a thick wall with core material occluded in it.

CHAPTER 5:

CHARACTERISATION OF PERFUME MICROCAPSULES

If experience was so important,

we'd never have had anyone walk on the moon.

Doug Rader

Summary

Perfume microcapsules made using the methods described in Chapter 4 were characterized. The permeability of the polymer wall to the perfume (leakage), the loading and encapsulation efficiency and the mechanical properties, including strength the microcapsules were measured and compared. The influence of the temperature of reaction and the organic and aqueous monomers used in the formulation on the final properties were studied, and the details are presented in this chapter.

5.1. Introduction

Microcapsules were produced in this project to have a very specific objective: perfume microcapsules are designed to be added to a detergent matrix, protect the perfume from the aggressive environment and be deposited on the fibres during the washing process. After the clothes are dried the microcapsules should be able to release the perfume by breakage of the polymer wall. To accomplish it they should have appropriate mechanical properties: microcapsules have to be strong enough to survive the handling, mixing, washing and drying processes but weak enough to break during the normal usage of the clothes where they are deposited. In addition the polymer wall has to be non-permeable to the perfume, which should not allow releasing of perfume from the microcapsules to the detergent during storage.

A spectrophotometric method has been used to measure the leakage of perfume from the microcapsules when these microcapsules were dispersed in water and a layer of hexane was added. Perfume has a peak of absorption in the UV spectrum and this property has been used to monitor the perfume concentration in hexane over time. Similar approaches have been used

by other authors to study the permeability of microcapsules made with polyamide compositions (Janssen *et al*., 1993).

Due to their small size, the mechanical characterisation of the microcapsules had been a difficult task until in the last years a new micromanipulation technique (Zhang *et al*., 1991) has been developed. This technique was initially used to characterize single cells but was soon adapted to measure the rupture force and the deformation at rupture of single melamine-formaldehyde microcapsules (Zhang *et al*., 1999) and the elastic-plastic behaviour of different polymer shells (Sun and Zhang, 2001 and 2002). The use of this technique allows acquiring reliable force-deformation data during a single capsule compression. Several microcapsules (between 25 and 50) have been compressed for each formulation to have more representative data.

A study of the effect of the formulation and process conditions on the final leakage and mechanical properties of the microcapsules is shown in this Chapter. More detailed elastic-plastic characterisation of the microcapsules with desirable properties for the formulations and processing conditions investigated has also been done.

5.2. Loading and encapsulation efficiency

5.2.1. Reactivity of the monomers with the perfume

Due to the high reactivity of the monomers used, it is expected that part of them react with some components of the perfume. To measure the percentage of perfume which reacts with the monomers, a set of experiments have been done. Using a gas chromatography instrument, three different samples prepared at room temperature have been compared: a sample of pure

perfume, a sample of perfume with trimesoyl chloride dissolved in it and a sample of perfume with diethylenetriamine. To prepare the samples 4g trimesoyl chloride and 8g diethylene triamine were dissolved in 40ml perfume oil. The signals from the three samples are showed in individual chromatograms (Figures 5.1, 5.2 and 5.3). They have also been plotted in the same graph (Figure 5.4) for better comparison. This last figure shows that most of the peaks for the perfume components are present in the three samples but with a lower signal when the monomers were added indicating that the monomers have reacted with the perfume. To quantify the amount of perfume which has reacted, the total area below the curves (the summatory of the areas for all the peaks) has been calculated, giving the value of 100% to the total area below the curve of the free perfume. The total areas of the trimesoyl chloride and diethylene triamine samples are 84% and 69% respectively. This indicates that at room temperature if the monomers are put in contact with the perfume a 16% of the perfume reacts with the organic monomer (trimesoyl chloride) and a 31% of the perfume reacts with the aqueous monomer (diethylene triamine). In the light of these results working at low temperature is preferred to minimise the reaction of the monomers with the perfume. In all the experiments done the organic monomer has been dissolved in the perfume at 1°C in an ice bath and the aqueous monomer was not in contact with the perfume due to the formation of the polymer wall.

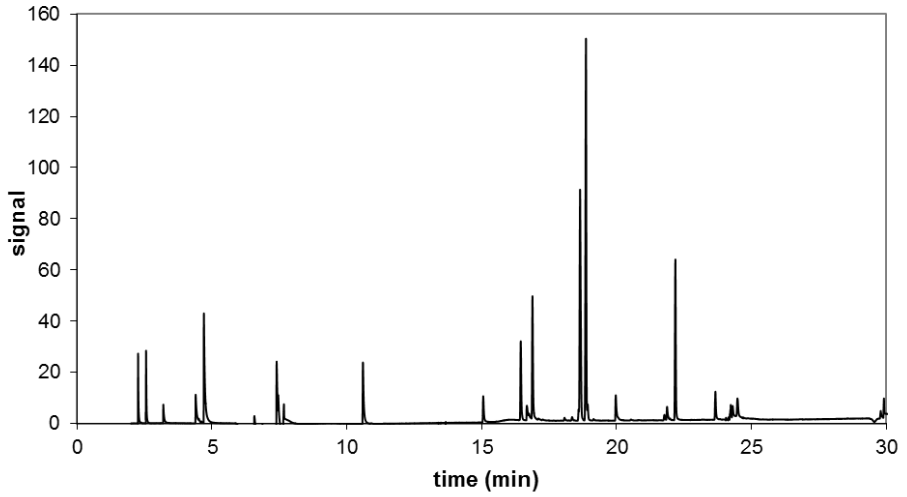


Figure 5.1. Chromatogram of pure perfume

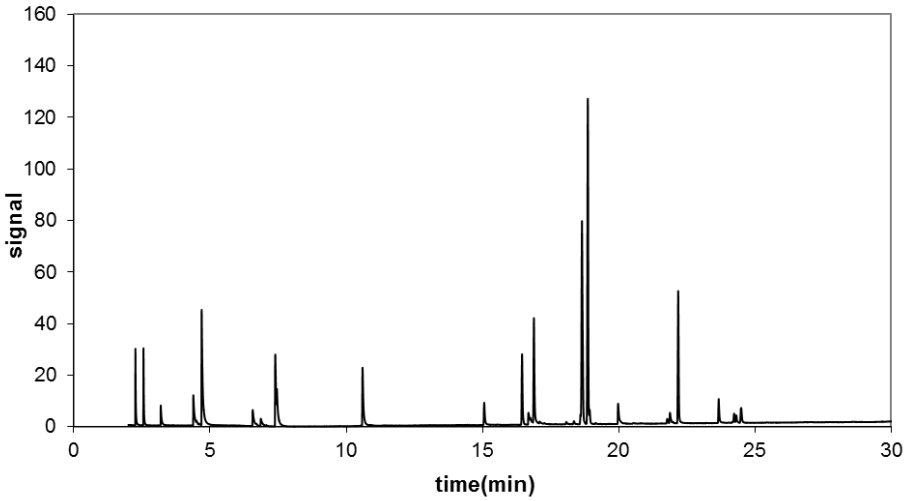


Figure 5.2. Chromatogram of perfume with trimesoyl chloride

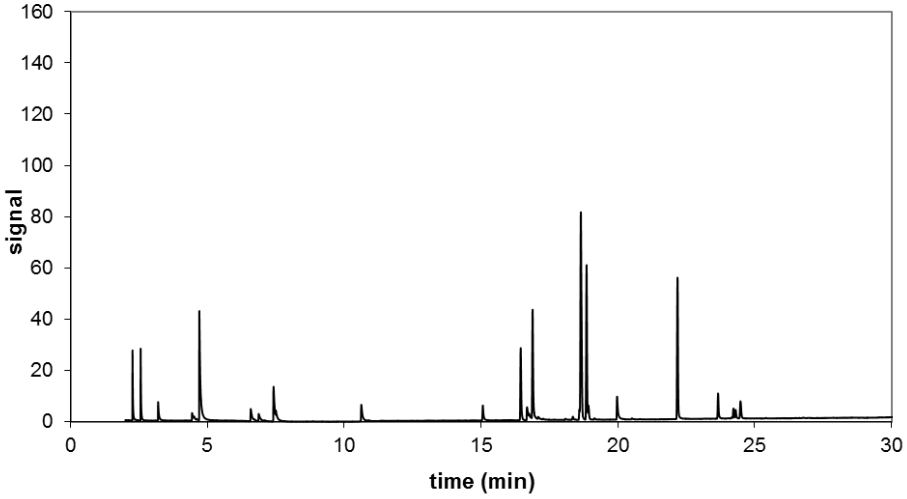


Figure 5.3. Chromatogram of perfume with diethylene triamine

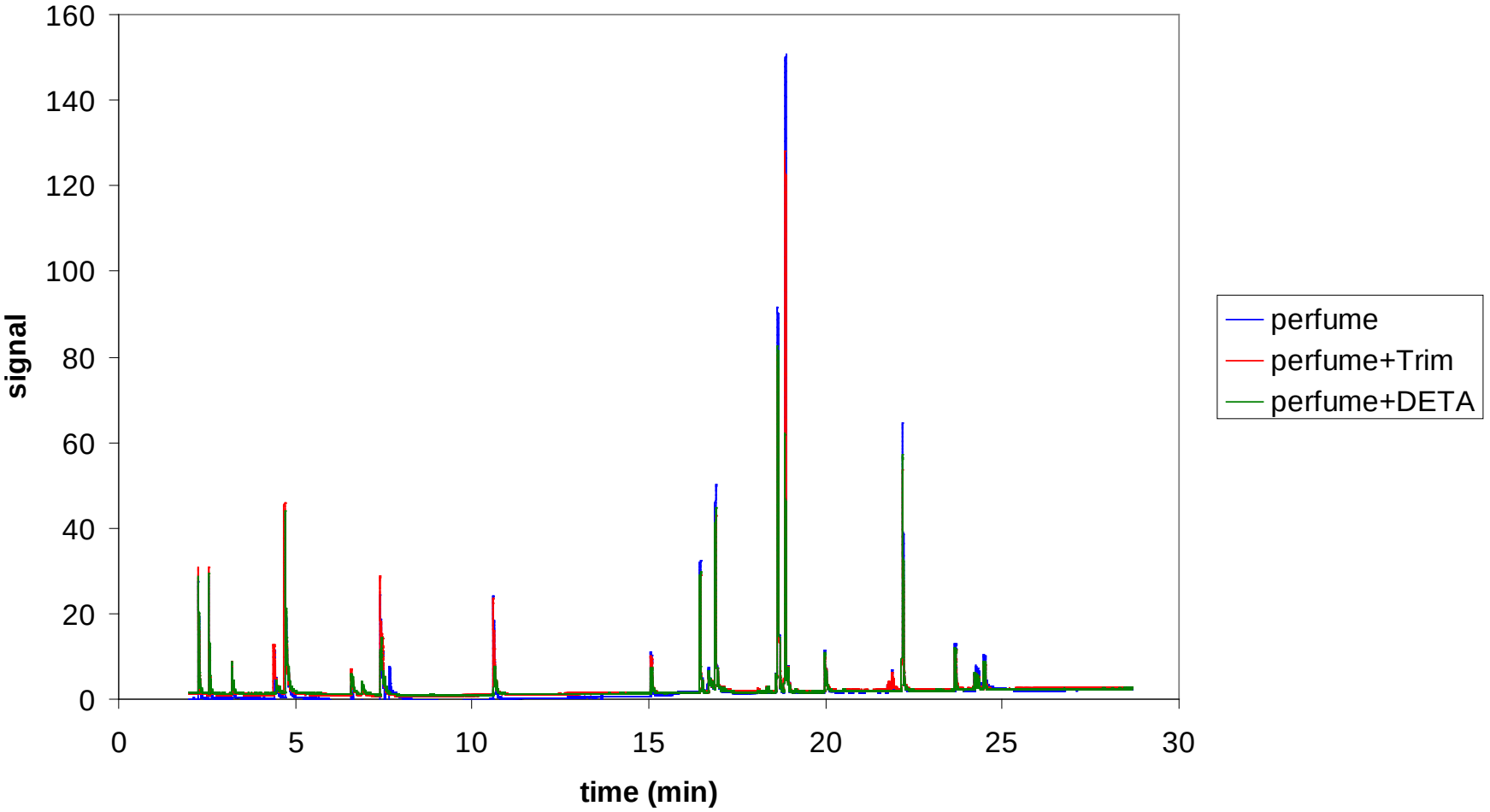


Figure 5.4. Comparison of the chromatogram for pure perfume (blue), perfume + trimesoyl chloride (red) and perfume + diethylenetriamine (green).

From the figure it is possible to see that there is only one peak that appears in the pure perfume and not in the perfume + monomers, it is at time 7.7min, all the rest of the peaks overlap perfectly showing less signal than the pure perfume, which means that part of the perfume has reacted with the monomers. It is easy to see that the blue line is always equal or higher than the red one and this one is always higher than the green one.

5.2.2. Loading of the capsules

The loading of the capsules is defined as the percentage of the capsule volume that is occupied by the perfume.

From the micrographs taken using the TEM device it is possible to measure the diameter of the capsules and the thickness of the wall and using these data it is possible to calculate the percentage of volume occupied by the perfume inside the capsule, assuming that the capsules are perfect spheres. There is no loading data for some formulations because the corresponding capsules have not survived the preparation process and no capsules have been seen under the TEM microscope. The loading results of the formulations that have survived the preparation process are shown in Table 5.1.

Formulation	d_{32}(μm)	thickness(nm)	loading(%vol.)
5TC	25.4	747 ± 28	83.4 ± 0.6
HMDA+EDA	22.3	621 ± 44	84.2 ± 1.1
0°C	22.6	553 ± 26	86.0 ± 0.6
DETA+EDA	23.2	456 ± 23	88.6 ± 0.6
HMDA	24.2	463 ± 18	89.0 ± 0.4
10TC	24.3	430 ± 18	89.8 ± 0.4
All250	25.8	434 ± 21	90.3 ± 0.5
6°C	26.6	442 ± 14	90.3 ± 0.3
12°C	25.0	333 ± 18	92.2 ± 0.4
15TC	26.4	347 ± 16	92.3 ± 0.3
DETA1+4	26.6	344 ± 12	92.5 ± 0.3
All50	24.7	301 ± 18	92.9 ± 0.4
EDA	22.8	277 ± 11	92.9 ± 0.3
18°C	35.6	241 ± 12	96.0 ± 0.2
1/2Trim	45.4	247 ± 8	96.8 ± 0.1

Table 5.1. Perfume loading (% vol.) of capsules made with different formulations.

Table 5.1 shows that the loading of the capsules is high, between 83 and 97%, as expected from capsules made using interfacial polymerisation techniques. Of course thicker walls led to lower loading. Similar results were obtained from previous works (Danicher *et al*., 2000).

5.2.3. Encapsulation efficiency of the process

The encapsulation efficiency (or yield) of the process is defined as the percentage of perfume that has been encapsulated.

The initial amount of perfume added to the system was known after the perfume was weighed before being added to the system. The perfume recovered from the microcapsules is calculated as the difference between the total perfume recovered from the slurries and the perfume that is dissolved in the slurry and not encapsulated.

The total perfume in the slurries was measured by breaking the microcapsules using glass beads under stirring with a vortex top mixer (FB15012 TopMix Evolution ZX, Fisher Scientific, UK). Hexane was added to extract all the perfume present and the perfume concentration was measured using a UV-spectrophotometer. All capsules were broken due to the collision of the glass beads and the microcapsules. Table 5.2 shows the total perfume recovery from different monomer compositions.

Experiment	Recovery(%)
HMDA	76.4
All50	78.2
DETA	99.5
EDA	97.6

Table 5.2. Total perfume recovered from different slurries formulations.

The results presented in Table 5.2 show that in formulations where HMDA was not used (DETA and EDA) the perfume recovery was almost 100%, that is, all the perfume emulsified in the first step was in the final slurry, which means that working at low temperature there was no reaction between the monomers and the perfume. On the other hand, when there was a HMDA monomer present in the formulation (HMDA and All50), there was less perfume recovery by approximately 20%. It is suspected that this amount of perfume was present inside the capsules but it was not possible to recover it. There is no reason to think that HMDA is more reactive with the perfume than EDA and DETA (in fact EDA is the most reactive monomer of the three) when the three monomers are chemically similar. It is supposed that this amount of perfume is embedded in the polymer walls. This can be explained by looking at the TEM micrographs presented in the previous chapter. When HMDA was used in the formulation a more complex wall structure was obtained and perfume

bubbles were occluded in polymer next to the wall. When the wall was ruptured the perfume in the bulk was released, but not the perfume that was in those wall polymer structures.

As it has been supposed that the perfume was present inside the capsules (it has been encapsulated) but it was not possible to fully recover it for some capsules, two different encapsulation efficiencies are calculated:

- Total encapsulation efficiency: it is the percentage of the perfume that has been (or it is suspected to be) encapsulated. It has not considered the maximum recovery of the perfume.
- Useful encapsulation efficiency: it is the percentage of perfume that is possible to be recovered from the microcapsules.

The maximum recovery experiment has not been done for all the formulations, but it is expected to be linked to the wall structure and this is related to the aqueous monomer used in the experiment. In all the formulations where HMDA was used, a recovery of 76.4% was obtained except in the All250 where the recovery was 78.2%. In the rest of formulations, the value for the DETA experiment was 99.5%, except in the EDA formulation where 97.6% was obtained.

The non-encapsulated perfume was measured during the leakage test, and it is calculated with the value obtained at time 0. The results show that the free perfume in solution was very low in all experiments, from 0 to 7%. Table 5.3 shows the percentages of the perfume recovered, non-encapsulated and the encapsulation efficiencies (Total and Useful) of all the formulations studied (formulations ordered by increasing Total EE).

Formulation	Non-encapsulated(%)	Recovered(%)	Total EE(%)	Useful EE(%)
20%paraffin	6.8 ± 4.4	99.5	93.2 ± 4.4	92.7 ± 4.4
1/2Trim	4.0	99.5	96.0	95.5
HMDA 15'	4.6	76.4	96.4	72.8
DETA+EDA	3.2	99.5	96.8	96.3
TC	3.1	99.5	96.9	96.4
kin12	3.1 ± 0.4	99.5	96.9 ± 0.4	96.4 ± 0.4
kin18	2.9	99.5	97.1	96.6
TETA	2.7	99.5	97.3	96.8
DETA 1+4	2.4	99.5	97.6	97.1
kin6	2.1	99.5	97.9	97.4
10TC	2.1	99.5	97.9	97.4
5TC	2.1	99.5	97.9	97.4
HMDA 60'	2.7	76.4	97.3	73.7
kin0	2.1	99.5	97.9	97.4
15TC	2.0	99.5	98.0	97.5
TC+HMDA	2.4	76.4	97.6	74.0
EDA	3.2	97.6	96.8	94.4
HMDA+EDA	1.7	76.4	98.3	74.7
All250	0.7 ± 0.2	78.2	99.3 ± 0.2	77.5 ± 0.2
HMDA	1.7	76.4	98.3	74.7
All50	0.3 ± 0.1	78.2	99.7 ± 0.1	77.9 ± 0.1

Table 5.3. Amounts of non-encapsulated perfume, perfume recovered and encapsulation efficiencies (EE) of all the formulations studied.

The amount of non-encapsulated perfume is low (below 7% in all cases and usually below 3%) therefore the total encapsulation efficiency of the process is high (over 93% in all cases). Other authors reported encapsulation efficiencies up to 95% (Soto-Portas *et al.*, 2003).

5.3. Leakage

All the leakage experiments done are expressed as the percentage of perfume released from the maximum recovered perfume calculated in the previous section.

5.3.1. Influence of the temperature on the leakage test

The leakage test is a measurement of the release kinetics of the perfume from inside the microcapsules. Experiments were done at room temperature, but this temperature was not constant from one day to another and it has been found that the temperature at which the

experiment was done had a huge influence on the final results obtained. An increase in the experimental temperature led to an increase in the leakage results (Figure 5.5).

This increase of the leakage with the temperature at which the experiment was done can be due to a combination of factors:

- An increase in the experimental temperature might produce a dilatation of the pores of the polyamide capsule's wall. The pores might become bigger and the leakage of perfume from inside the microcapsules is favoured.
- An increase in the experimental temperature can also increase the diffusion coefficients of perfume components, favouring the leakage kinetics.

Error bars in the following graphs represent the standard error of the repeated experiments (from 3 to 5).

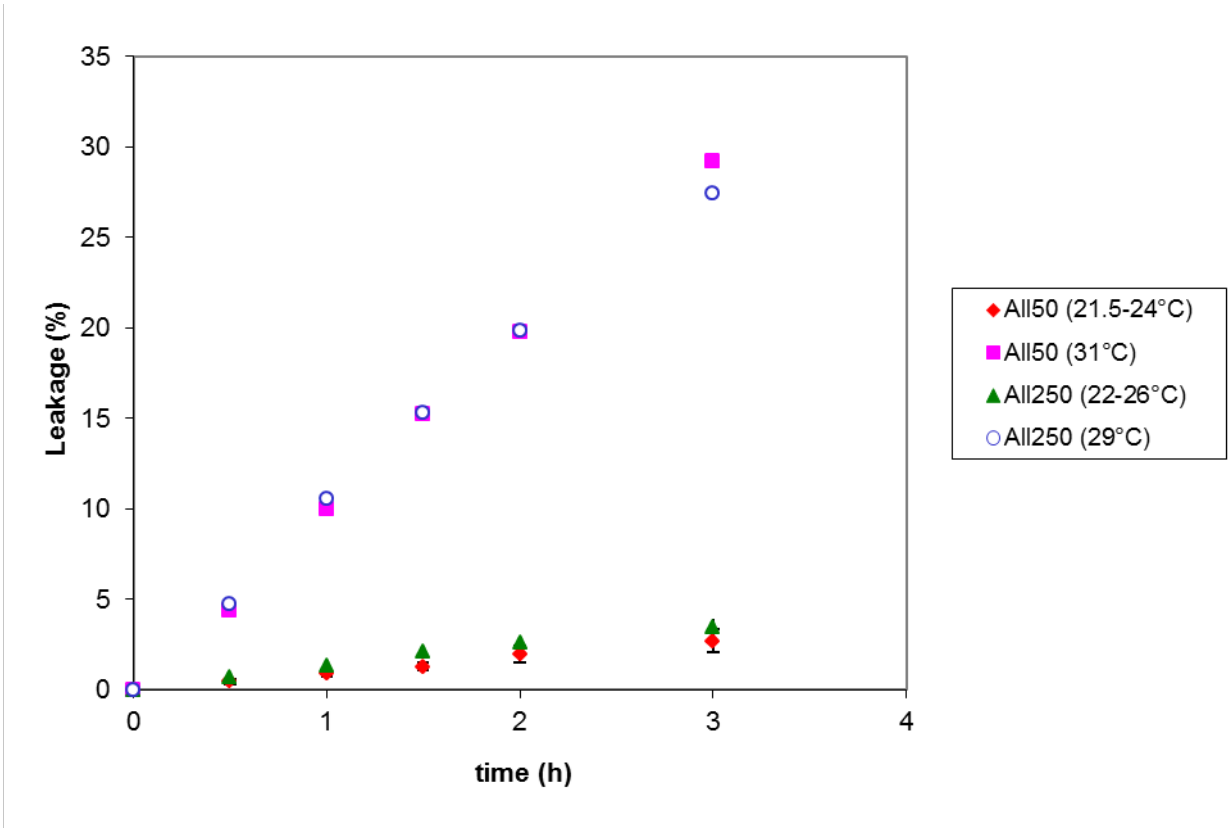


Figure 5.5. Effect of the temperature on the release kinetics for 2 samples.

The temperature at which each leakage experiment was done is noted next to the formulation name.

5.3.2. Solubility of the perfume in water

A method to measure the leakage of perfume from the microcapsules has been developed (described in Chapter 3). Briefly this method consists of dispersing a sample of capsules in water and adding a layer of hexane, the hexane will extract the perfume from the water phase under magnetic stirring and the concentration of perfume in the hexane can be monitored using a UV-spectrophotometer. The perfume is a mixture of many components, and some of them have a high affinity for water so that not all the perfume is extracted by the hexane. An experiment has been done to measure the percentage of perfume which is extracted by the hexane.

A known amount of perfume was added to 90mL water, 50mL of hexane was added and the concentration of perfume in the hexane phase was monitored under magnetic stirring. The percentage of perfume extracted from the water phase was 78±5%, which means that 22±5% of the perfume remained in the water phase.

The partition coefficient of the perfume in the system water/hexane has been calculated:

$$H=[\text{perfume}]_w/[\text{perfume}]_h=0.16\pm0.05$$

where:
$[\text{perfume}]_w$ is the concentration of perfume in the water phase
$[\text{perfume}]_h$ is the concentration of perfume in the hexane phase

5.2.3. Influence of temperature of reaction

The effect of the temperature of reaction on the leakage of perfume from the capsules has been studied. Different batches of microcapsules made at 0, 6, 12 and 18°C have been prepared and their leakage measured. These experiments have been done using only one organic monomer (trimesoyl chloride) and one aqueous monomer (diethylenetriamine). The leakage results are shown in Figure 5.6.

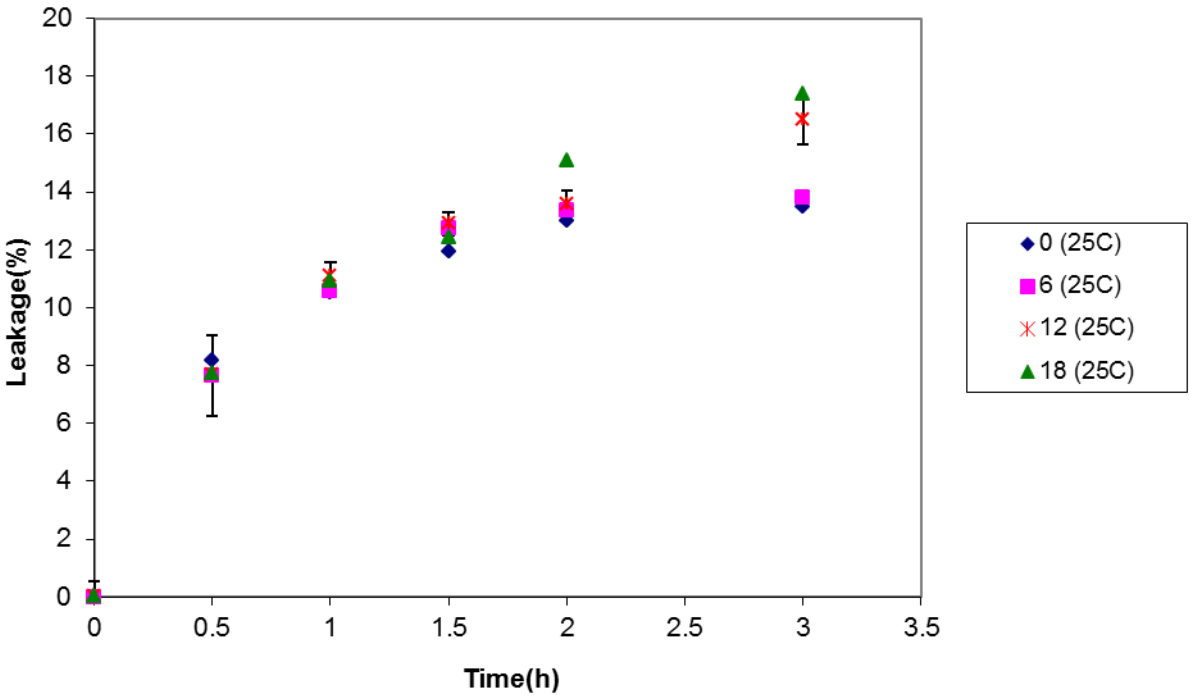


Figure 5.6. Leakage of perfume from capsules made at 0, 6, 12 and 18°C.

The leakage profiles of the samples made at different reaction temperatures are very similar. The temperature of reaction does not seem to have an effect in the perfume release from the microcapsules.

5.2.4. Influence of organic monomer type and concentration

Three different organic monomers have been used to prepare capsules: sebacoyl chloride (bifunctional aliphatic), terephthaloyl chloride (bifunctional aromatic) and trimesoyl chloride (trifunctional aromatic). In all cases only one aqueous monomer has been used, diethylenetriamine (trifunctional).

Working with sebacoyl chloride has been unsuccessful since no single capsules have been made. Although some polymer was produced, the reaction formed a continuous polymer instead of discrete microcapsules and the perfume was not encapsulated. It seems that the use of aliphatic organic monomers led to less rigid capsules and these ones were not rigid enough to form a discrete capsule (Persico, 2005).

Using terephthaloyl and trimesoyl chloride led to microcapsules with the desired size. However, these capsules had very different leakage properties. In Figure 5.7 it can been seen that there are different leakage profiles between a sample made with terephthaloyl and a sample made with trimesoyl chloride (diethylenetriamine used as aqueous monomer in both cases).

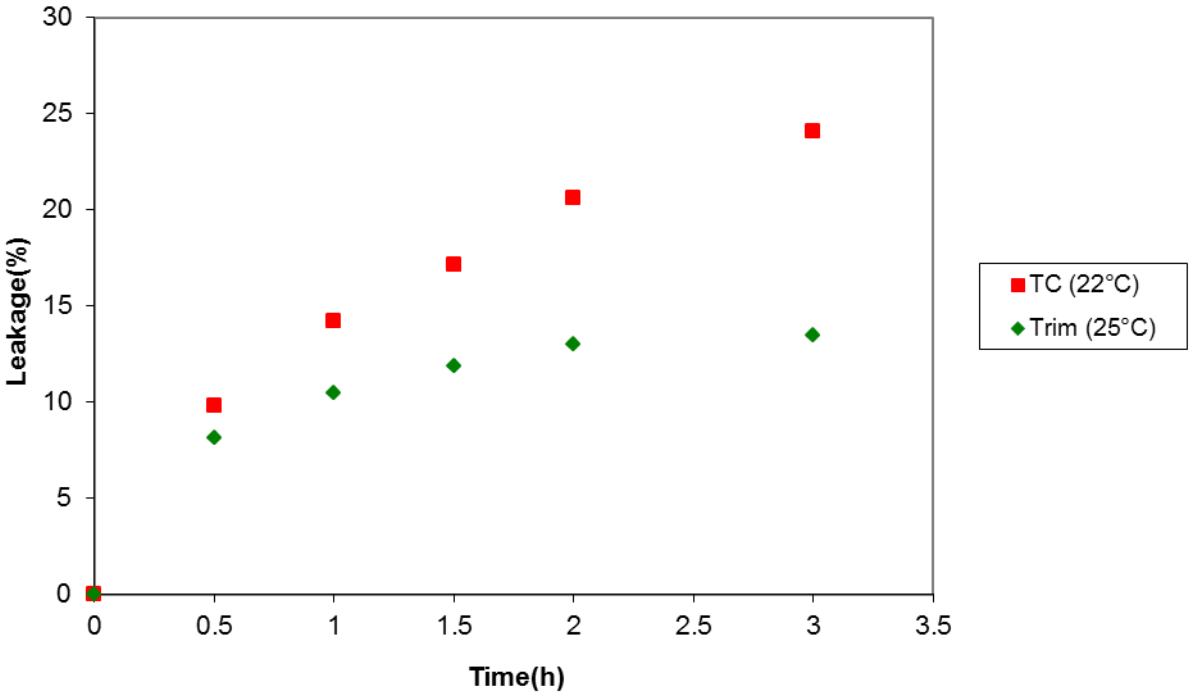


Figure 5.7. Leakage of perfume from capsules made with different organic monomers: terephthaloyl and trimesoyl chloride.

Leakage of perfume from capsules made with the trifunctional monomer (trimesoyl) is almost half of that from the bifunctional (terephthaloyl) one. The use of a trifunctional monomer might lead to a highly cross-linked polymer which provided a better resistance to leakage.

Some combinations of these two monomers have also been studied. In Figure 5.8 the leakage results of some mixture formulations are shown.

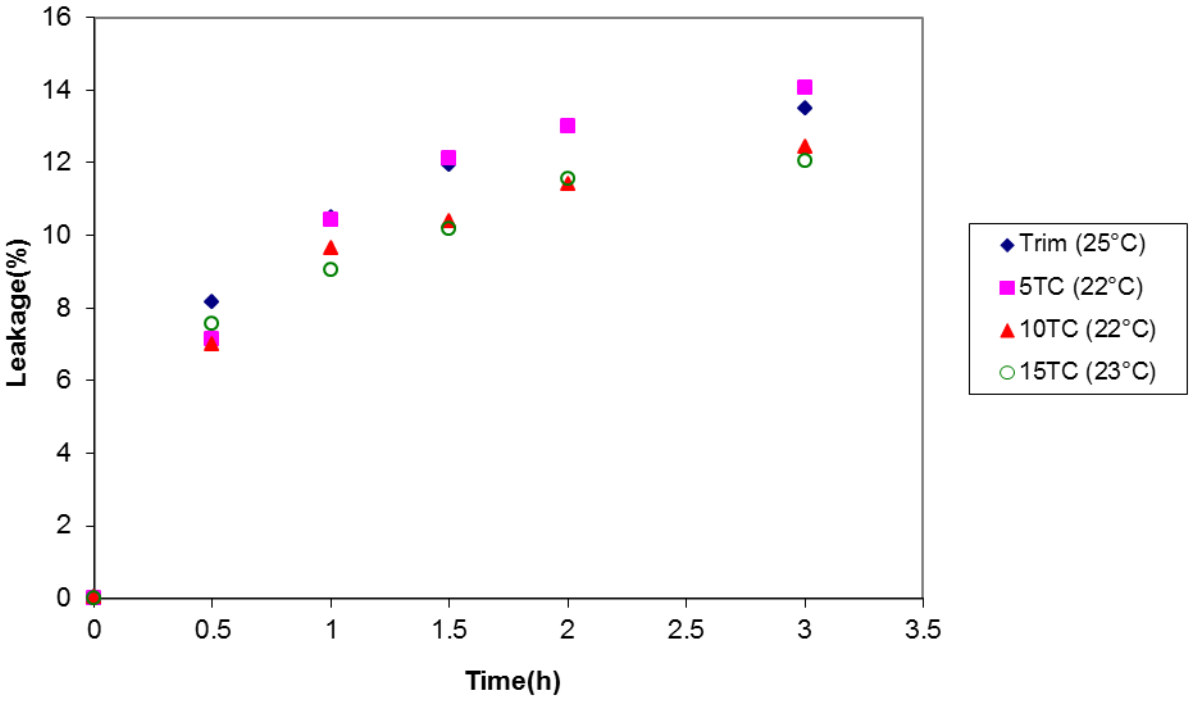


Figure 5.8. Leakage of perfume from capsules made with different organic monomer ratios.

It is possible to see that making capsules with a mixture of both monomers did not increase the leakage from the capsules. Microcapsules might still be highly cross-linked and there was more monomer available for reaction.

The amount of organic monomer available may also be very important to the final leakage properties of the capsules. An experiment using half the amount of the trifunctional monomer has been made. In this case no bifunctional monomer was added. The leakage resulting from the capsules formed is really high, reaching 54% in 3h. Another experiment using a third of the amount of the trifunctional monomer was made and no capsules were produced.

5.2.5. Influence of aqueous monomer type and addition time.

Four different aqueous monomers have been used to prepare microcapsules: ethylenediamine (bifunctional short chain), hexamethylenediamine (bifunctional long chain), diethylenetriamine (trifunctional) and triethylenetetramine (tetrafunctional).

Experiments have been done to see the influence of the monomer used on the leakage of perfume from the microcapsules. Equal amounts of active groups of each monomer have been used in different experiments with a fixed amount of organic monomer (trimesoyl chloride only). The leakage results of these experiments are shown in Figure 5.9.

The leakage from the microcapsules made with triethylenetetramine is not shown in the graph due to its exceptionally high value. The value measured for the leakage from the triethylenetetramine experiment is 59% in 3h.

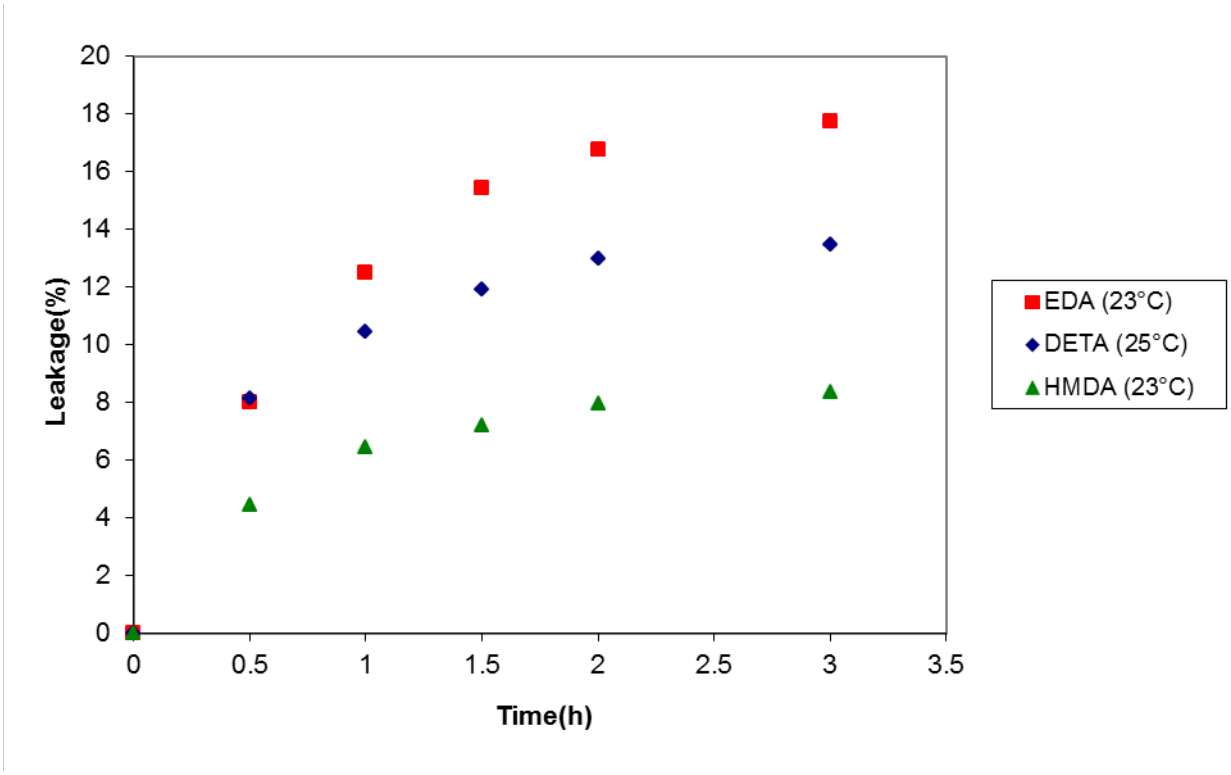


Figure 5.9. Leakage of perfume from capsules made with different aqueous monomers.

Figure 5.9 shows that using hexamethylenediamine the leakage is reduced to 60% of that from diethylenetriamine microcapsules. Capsules made with ethylenediamine present worse leakage properties. In previous works it was found that the use of diamines in addition to triamines reduced the wall permeability (Janssen *et al.*, 1992), specially the use of hexamethylenediamine (Toubeli and Kiparissides, 1998) which formed walls with smooth and dense surfaces and low porosity (Persico, 2005).

Considering that the aqueous monomer used has a big influence on the leakage properties of the final capsules, formulations using various aqueous monomers and addition times have been studied. Due to the method used to produce microcapsules, the aqueous monomers can be added at different times (Mathiowitz and Cohen, 1989a). According to the theory of capsule formation by interfacial polymerisation (Arshady, 1989), the lower the initial reaction rate is, the better properties the final polymer will have. Diethylenetriamine has been selected as the initial monomer added to the reaction because as seen in the previous chapter its reaction kinetics is the slowest of all the monomers. Different combinations of organic and aqueous monomers and addition times of aqueous monomers have been studied. Their formulations are shown in Table 4.1 in Chapter 4 and their leakage results are shown in Figure 5.10.

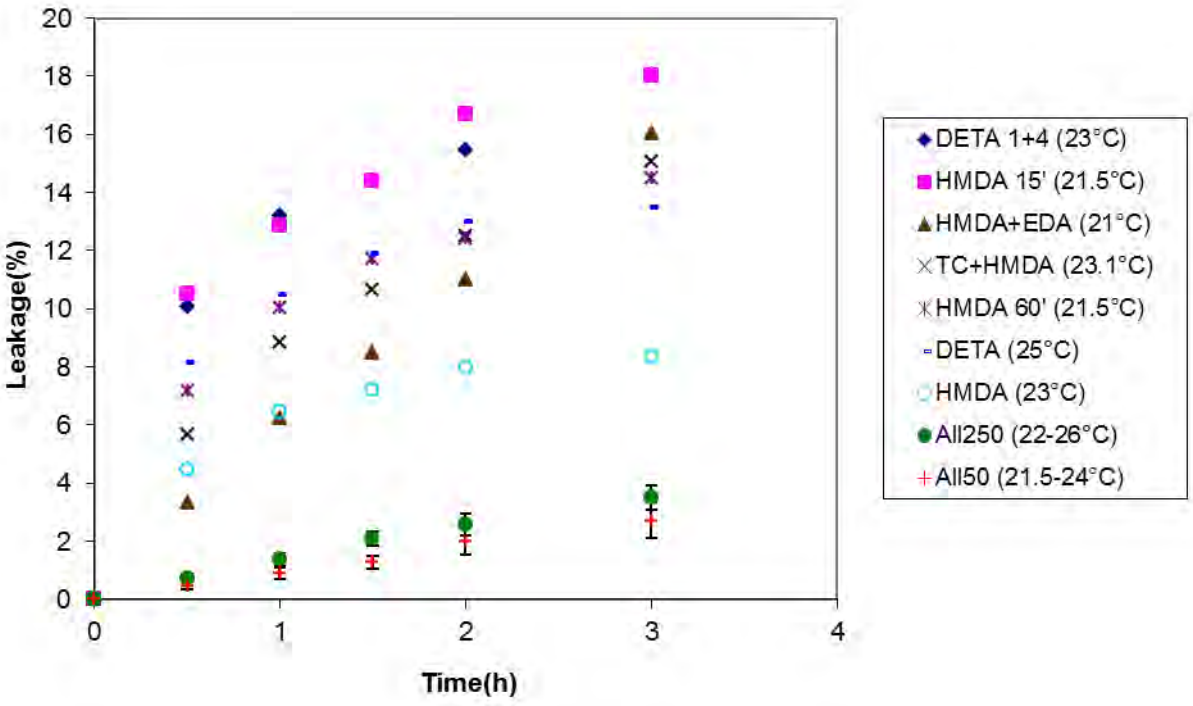


Figure 5.10. Leakage from capsules made with different monomers and addition times

It can be seen that there are only two formulations that reduced the leakage in comparison with the previous formulations DETA and HMDA. These two formulations show leakages three times lower than the hexamethylene capsules alone, around 3% in 3h. The best results were obtained with a formulation made using one part of a combination of trimesoyl chloride and 10% terephthaloyl chloride as organic monomers and one part of diethylenetriamine added at time 0, five parts of hexamethylenediamine added at time 15min and one part of ethylenediamine added at time 1h.

The leakage experiments for these two best formulations (All50 and All250) have been repeated 4 and 5 times respectively and the standard error of the average is presented in the error bars. It was not possible to control the temperature at which the experiment was done and an interval of temperatures is indicated next to the formulation name.

5.2.6. Effect of the viscosity of the encapsulated perfume

The increase of the viscosity of the encapsulated phase is supposed to have an influence on the leakage because it can provide a resistance to the movement of the perfume from inside the capsule to the shell.

An experiment encapsulating a mixture of perfume and 20% paraffin oil has been done. This addition resulted in an increase of the viscosity of the encapsulated phase from 5.5cP to 9.5cP. The leakage results show that this change in the viscosity of the encapsulated phase had no effect on the leakage properties of the capsules, as shown in Figure 5.11. Both formulations, with and without paraffin have a very similar leakage profile. The main difference between them is that the formulation with paraffin oil presents a higher concentration of non-encapsulated perfume in the slurry. The leakage indicates that the mass transfer resistance inside the capsules is not significant for the conditions investigated.

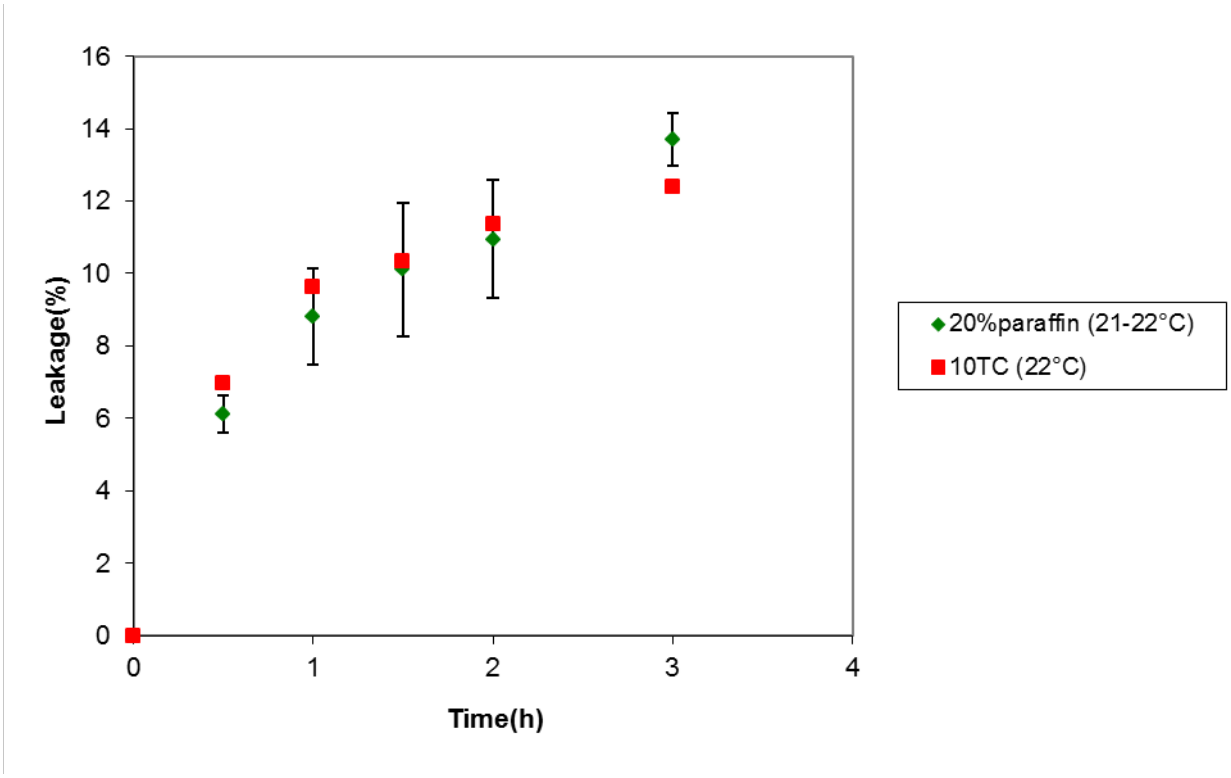


Figure 5.11. Effect of the addition of paraffin oil to the encapsulated perfume on the leakage of the microcapsules.

5.4. Mechanical properties

5.4.1. Mechanical strength

5.4.1.1. Influence of temperature of reaction

The temperature of reaction seems to be one of the most important factors on the final properties of the capsules. Experiments show that working above 20°C the capsules formed were very weak and they broke when they were dried, which released the perfume from the capsules. Capsules produced at a temperature below 18°C did not break when they were dried, and their mechanical strength could be measured using the micromanipulation rig. Figure 5.12 shows the nominal stress (force divided by the cross sectional area of a capsule) at rupture of samples prepared at different reaction temperatures.

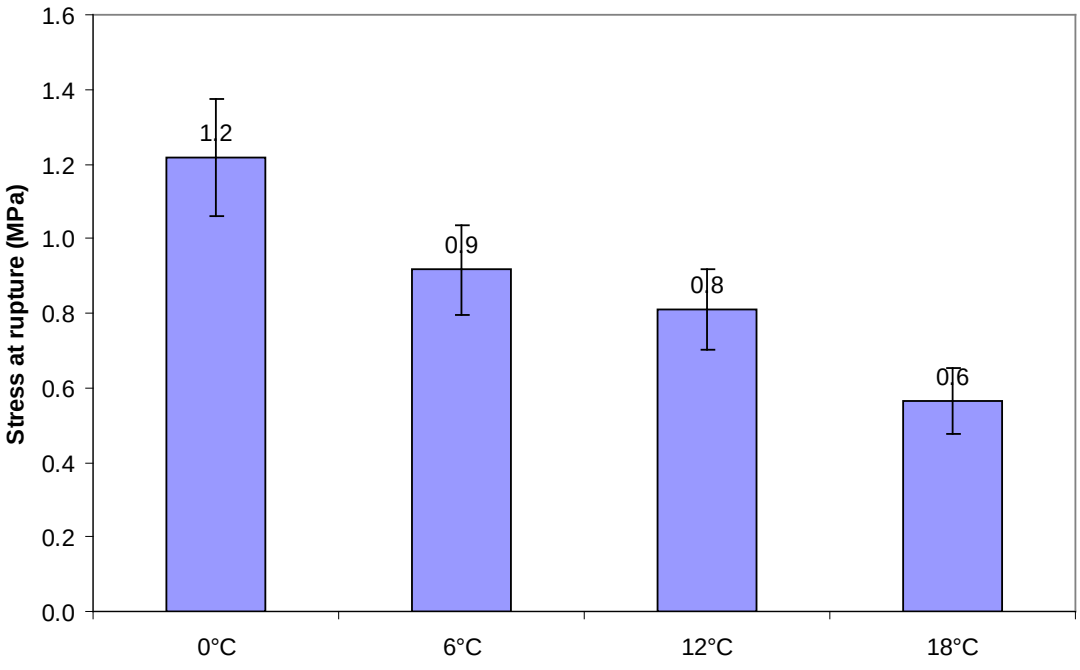


Figure 5.12. Nominal stress at rupture of capsules produced at different temperature.

The results show that capsules made at 18°C were much weaker than the ones made at 0°C. Capsules made over 18°C were too weak and brittle to be stable. As shown in previous

chapter the reaction kinetics has a high dependence on temperature and at a higher temperature the reaction rate is faster. It seems that over 18°C the kinetics is too fast and the polymer formed has no time to form a stable structure.

5.4.1.2. Influence of organic monomers

Different monomers have been used and capsules have only been produced using the aromatic ones. Capsules were not formed when sebacoyl chloride was used as organic monomer, only trimesoyl and terephthaloyl chloride led to stable capsules. This behaviour has been reported before by other authors (Persico, 2005) for some internal phases, and it seems that the encapsulated phase has a big influence on whether capsules can be formed for the same organic monomers. Authors reported that using aliphatic acid chlorides it is possible to encapsulate toluene, benzene or xylene (Mathoiwitz and Cohen, 1989a; Soto-Portas *et al.*, 2003; Persico, 2005) but it is not possible to obtain stable capsules of jojoba oil (Persico, 2005) using the same conditions as with toluene.

In addition it has been found that capsules made only with the bifunctional aromatic monomer (terephthaloyl chloride) or low concentrations of the trifunctional one were not stable after a few days due to their high leakage, as shown in the previous section, and low rigidity. Experiments showed that at least 70% of functional groups (COCl) of the trifunctional monomer (trimesoyl chloride) were needed to obtain good quality capsules. Capsules with different ratios of both monomers have been made (Figure 5.13) and the ones with a ratio 82:18 (Trim:TC) presented the highest nominal strength at rupture. This result is quite unexpected because all the previous authors found that a small amount of trifunctional

monomer in the organic phase in comparison with the bifunctional one used was adequate. This means again that the encapsulated active has a big influence on the formation of capsules. If a much higher concentration of cross-linking organic monomers is required to encapsulate perfume, there are the following impacts: the trifunctional monomer is much more reactive than the bifunctional one and more perfume reacts with the monomer, therefore the monomer should be dissolved in the perfume at low temperatures to prevent this reaction.

The trifunctional organic monomer is supposed to be the one which provides cross-linked walls (Persico, 2005), resulting in an improved structure of the capsule. But the trifunctional monomer is less soluble in the perfume than the bifunctional one. The solubility of trimesoyl chloride in the perfume has been measured by adding increasing amounts of monomer in a known amount of perfume until it was not dissolved anymore, being the monomer crystals at the bottom of the beaker. The solubility at 1ºC was of about 0.1g of monomer per gram of perfume. Some terephthaloyl chloride was added to increase the amount of functional groups, which increased the amount of polymer formed.

An experiment using half the amount of trimesoyl chloride (22.5 mmol instead of 45mmol of COCl functions) without terephthaloyl chloride added was also carried out. The nominal stress at rupture of this sample was by far higher than the ones made by the experiments with higher monomer concentrations. This may be explained according to the reaction mechanism: the slower the reaction is, the more crystalline the structure of the wall and more resistant mechanically the capsule. But there is a limit on the monomer concentration to obtain capsules, an experiment with one third (15mmol) of the monomers was done and capsules were not produced. On the other hand capsules made with half the amount of trimesoyl

chloride had good mechanical properties but the walls were very permeable to the perfume (almost 54% of the perfume released in 3h in the leakage test experiment) so that they are not useful for the purpose of this research.

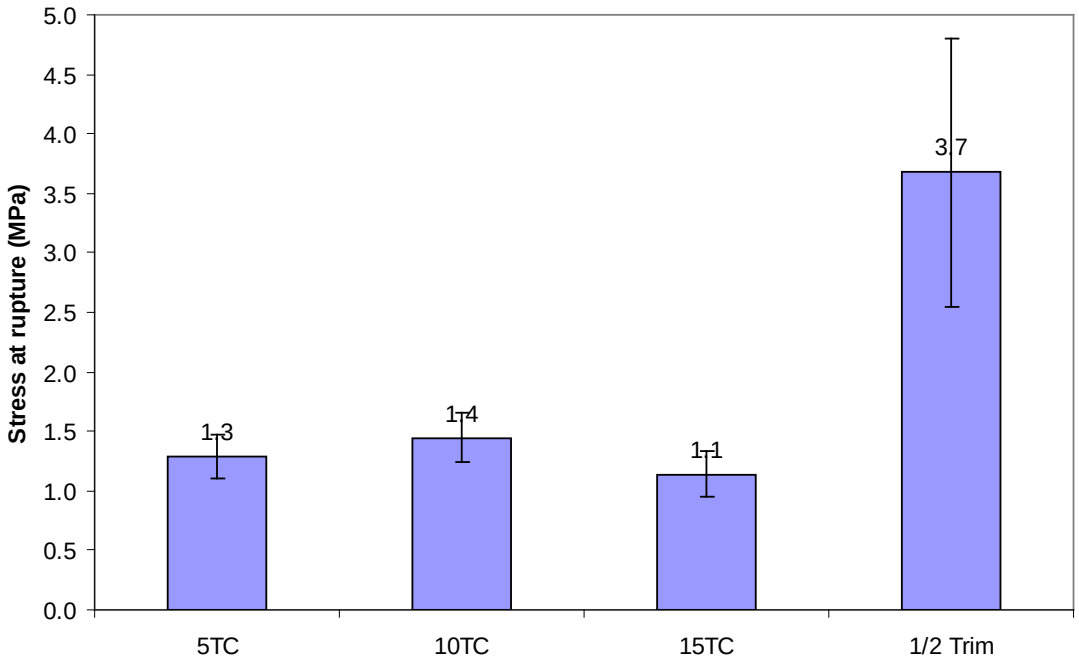


Figure 5.13. Nominal stress at rupture of capsules produced with different organic monomer concentrations.

5.4.1.3. Influence of aqueous monomers

Different aqueous monomers have been used to produce microcapsules. Capsules have been prepared using each aqueous monomer alone in the formulation with trimesoyl chloride as organic monomer and reacting at 0°C microcapsules (Figure 5.14). Capsules made with hexamethylenediamine presented the highest mechanical resistance and, as it was seen in the previous section, they formed the less permeable capsules. It is clear that the use of the cross-linking agent in the organic phase is much more efficient than their use in the aqueous one (Persico, 2005).

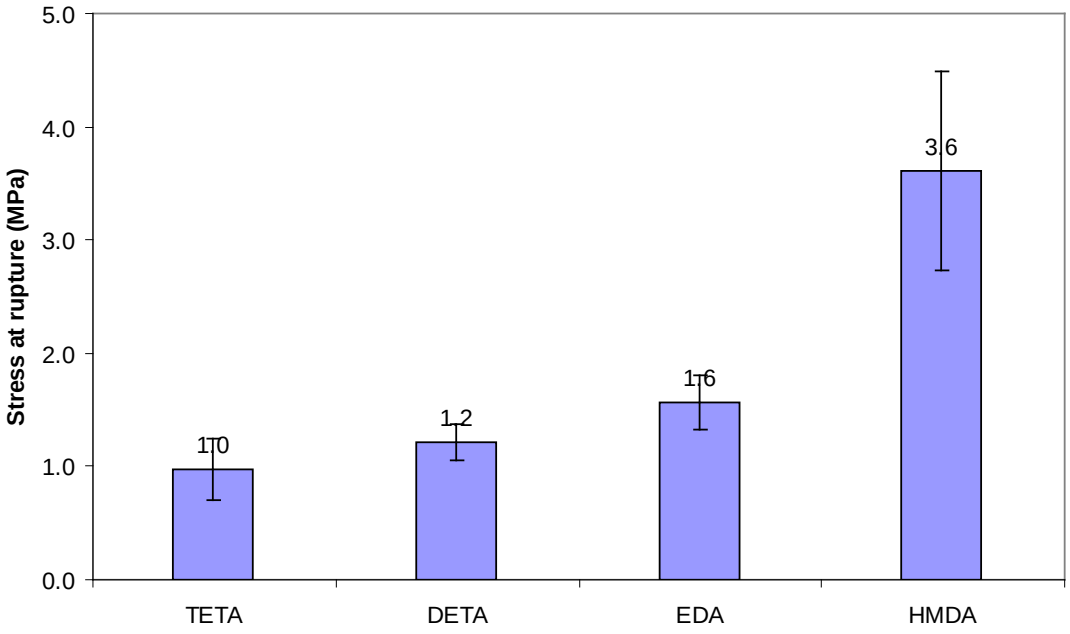


Figure 5.14. Nominal stress at rupture of capsules prepared with different aqueous monomers.

According to the method used to produce microcapsules it is possible to add different aqueous monomers at different times (it is not possible to do that with the organic monomer). Some formulations using different combinations of monomers and different addition times have been produced. Their nominal stress at rupture has been measured and it is shown in Figure 5.15.

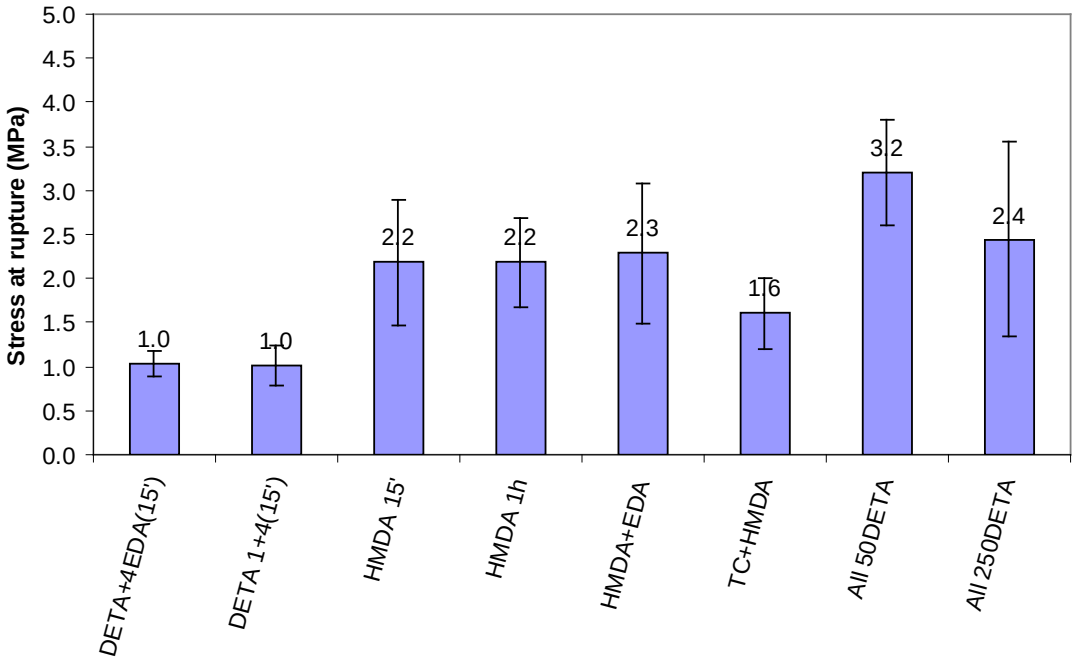


Figure 5.15. Nominal stress at rupture of capsules prepared with different monomers added at different times (see Table 4.1 for details of each formulation).

The results show that formulations where HMDA was added present higher resistance and that EDA does not improve the properties of the capsules in comparison with DETA. The best results have been obtained using a combination of organic and aqueous monomers, the formulation All50DETA.

In conclusion the formulation All50DETA has been the one with the best properties: high stress at rupture and lowest leakage.

5.4.1.4. Deformation at rupture

The deformation (displacement divided by the capsule diameter) at rupture of single microcapsules has also been calculated from micromanipulation data. Results are shown in Figure 5.16.

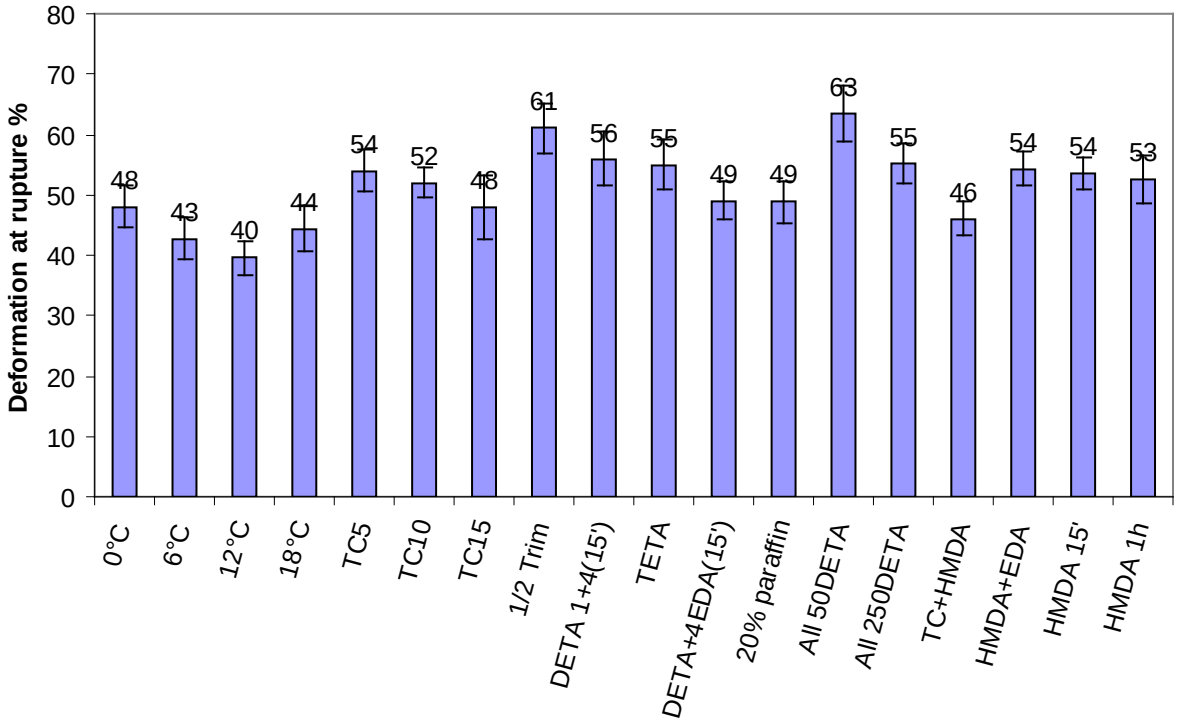


Figure 5.16. Deformation at rupture for all the samples compressed

As can been seen in the figure, all samples presented deformation values between 40 and 63%. There is not a clear relationship between the deformation at rupture and the sample composition and reaction conditions.

5.4.2. Viscoelasticity

Microcapsules with the best properties (All50DETA, highest nominal stress of rupture and lowest leakage) were further characterised. “Compress and hold” and “compress and release” experiments have been done to these microcapsules to characterize their viscoelastic properties if any.

The **compress and hold experiment** (Figure 5.17) shows that after initial compression, the force decreases when the capsule was held, the relative force relaxation (drop in force divided

by the peak force value) for each displacement is shown in Table 5.4. This result indicates that the capsule exhibited a visco-elastic behaviour. The figure also shows that the initial force versus time curves corresponding to displacements of 4μm (12%) and 7 μm overlap, which indicates the microcapsule was mainly elastic for the small displacement. However, the following consecutive compressions resulted in significantly different force versus time curves, which implies the microcapsule had undergone plastic deformation when the displacement exceeded a certain value (10% as seen in the compress and release experiments in the next section).

Displacement (μm)	4	7	10	13	16
Relative relaxation (%)	34	32	32	33	32

Table 5.4. Relative force relaxation for different displacements during the compression of a 32.6μm capsule at 2μm/s and then holding

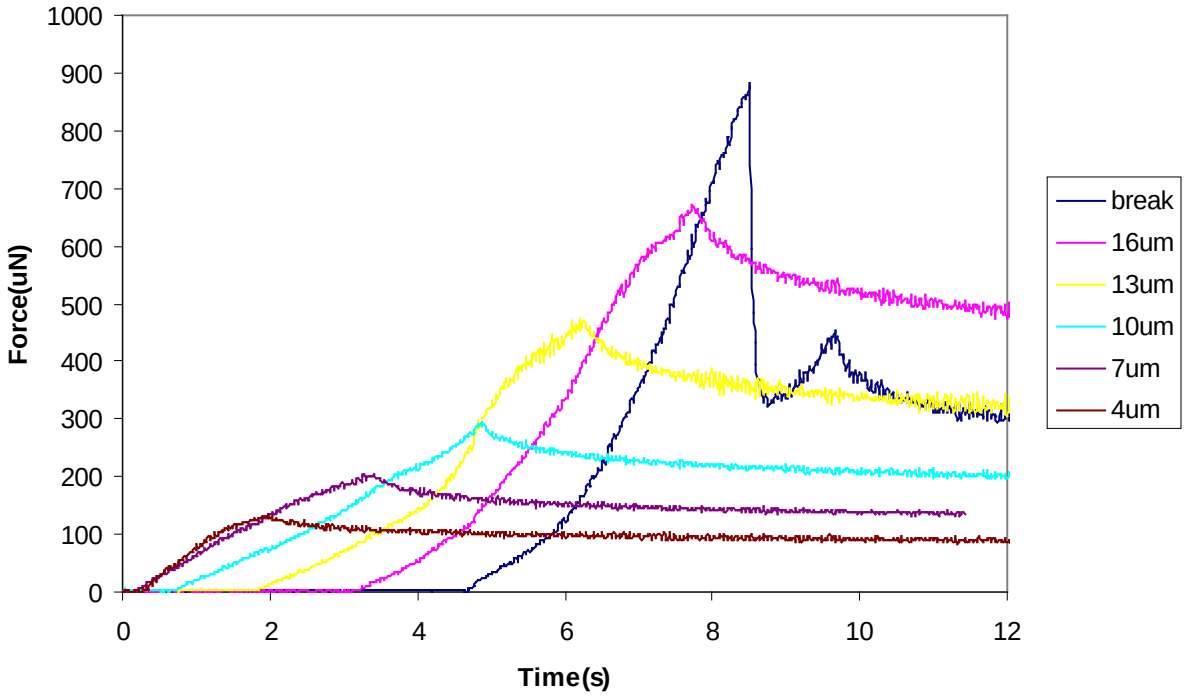
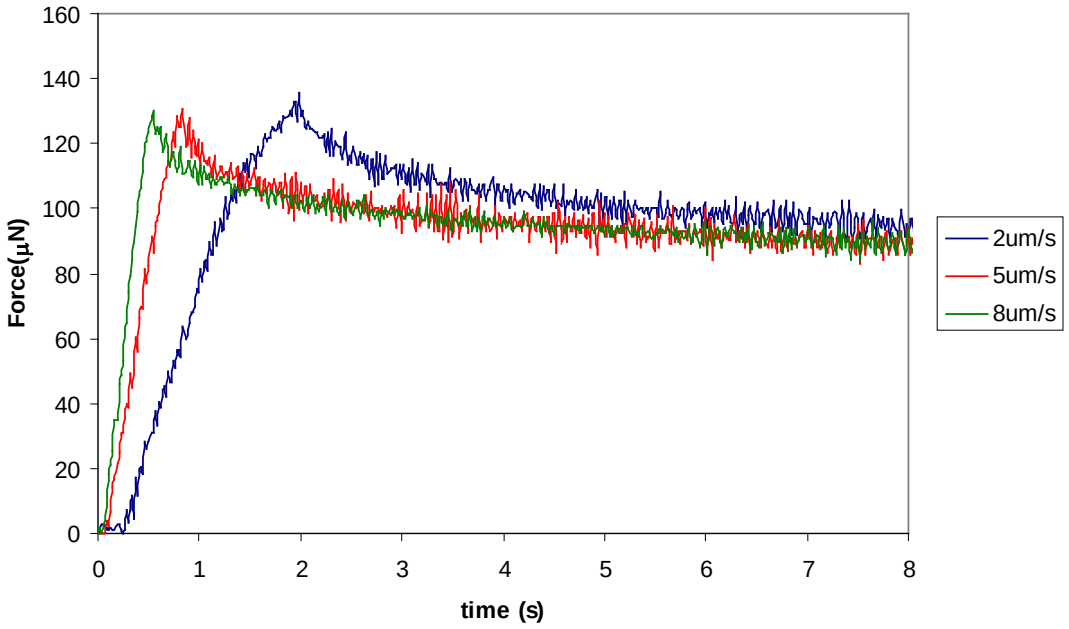
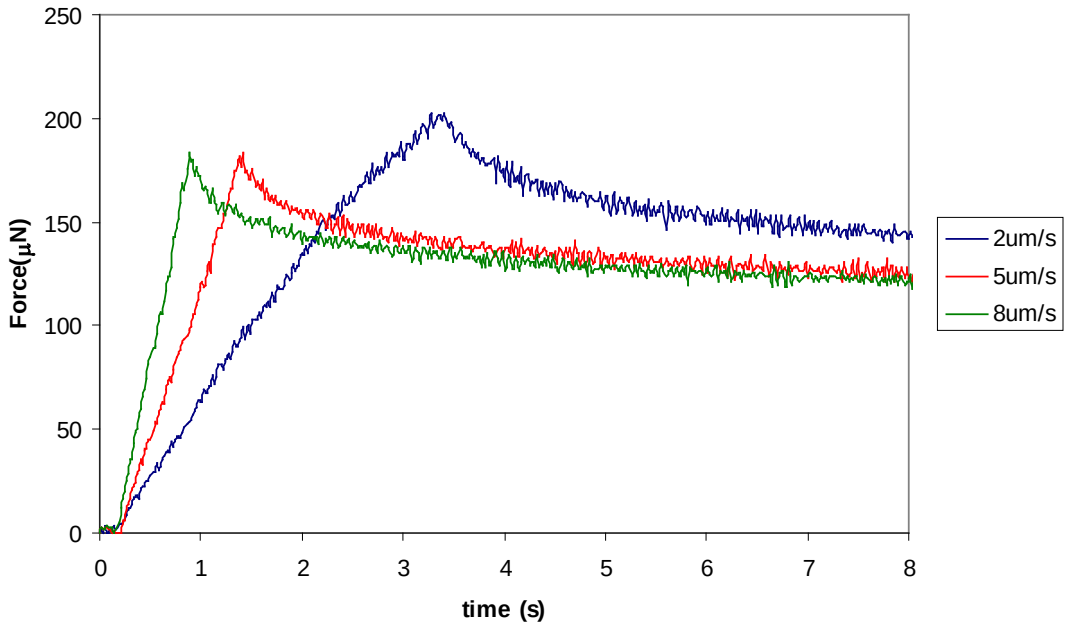


Figure 5.17. Force versus time data for compression of a 32.6μm microcapsule to different displacements at a speed of 2 μm/s and then holding.



(a)

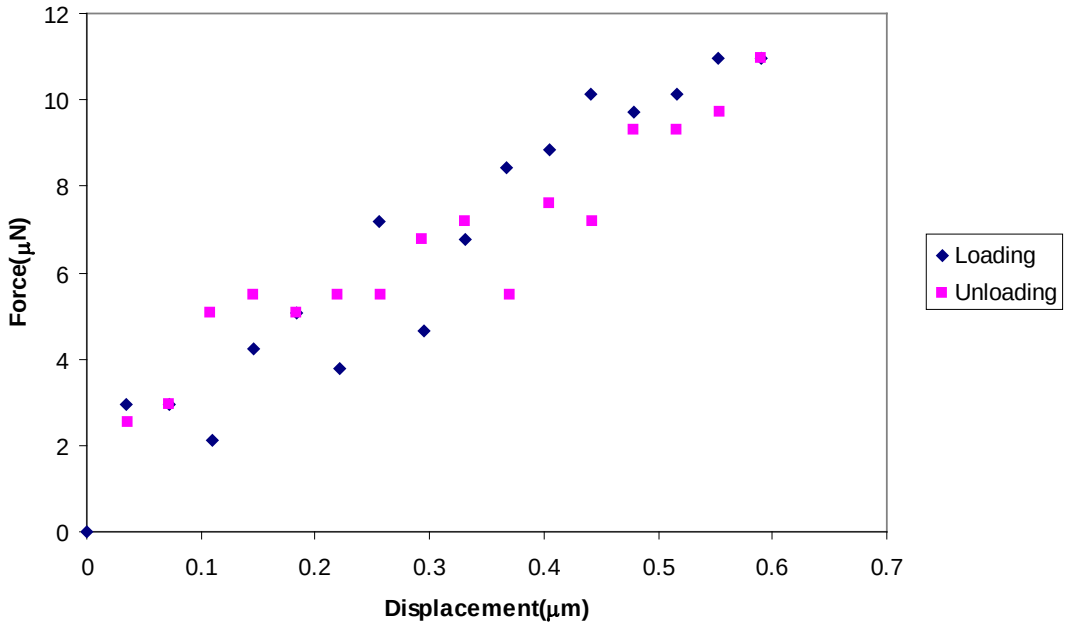


(b)

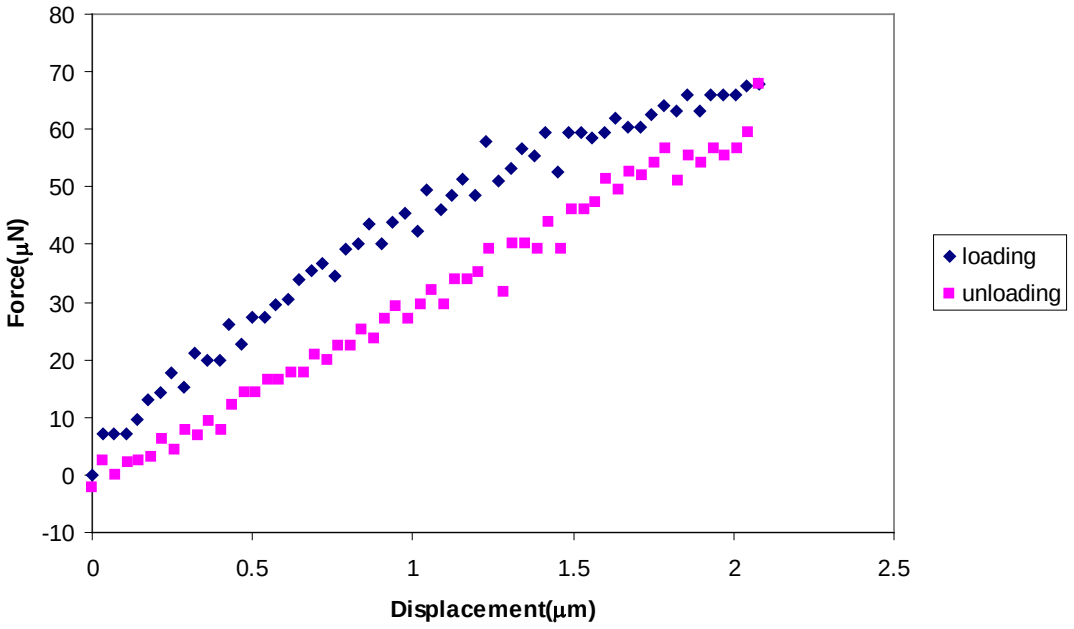
Figure 5.18. Compression of a 32.6μm microcapsule at different compression speeds and then holding. Two displacements are shown: (a) 4 μm and (b) 7μm.

Compression at different speeds and then holding has also been done (Figure 5.18).It is possible to see that in the mainly elastic region (4μm graph) the force measured at the 3 different speeds and the relative relaxation were very similar. Consecutive compressions made in the plastic region (7μm graph) show a slightly higher measured force (10%) for the first experiment done (2μm/s) than for the other two (5 and 8μm/s) although the relative relaxation observed was also very similar.

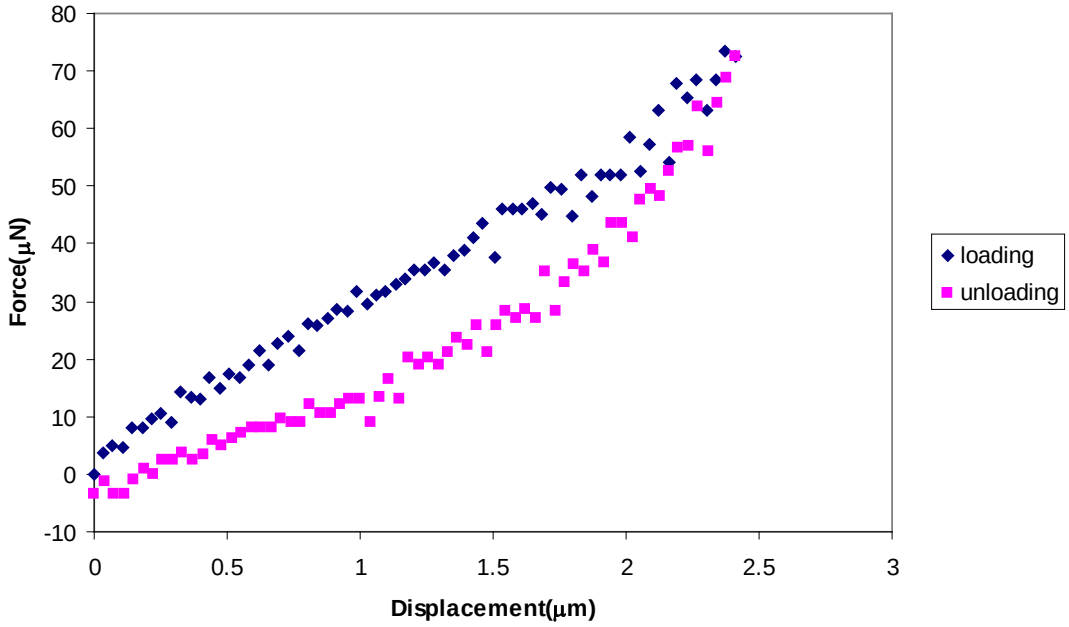
The **compress and release experiment** (Figure 5.19) confirms that at a low final deformation (3%) the wall presented elastic properties. When the deformation was increased (6%) a marginal hysteresis was found, which is a signal of visco-elastic behaviour. At a 10% deformation the wall started showing plastic behaviour and at 18% it is possible to see that there was a more profound hysteresis and that there is a clear plastic behaviour, the force corresponding to unloading has already reduced to 0 at a point where the probe is far away from the initial position.


12
10
8
6
4
2
0
Force(μN)
0
0.1
0.2
0.3
0.4
0.5
0.6
0.7
Displacement(μm)
Loading
Unloading

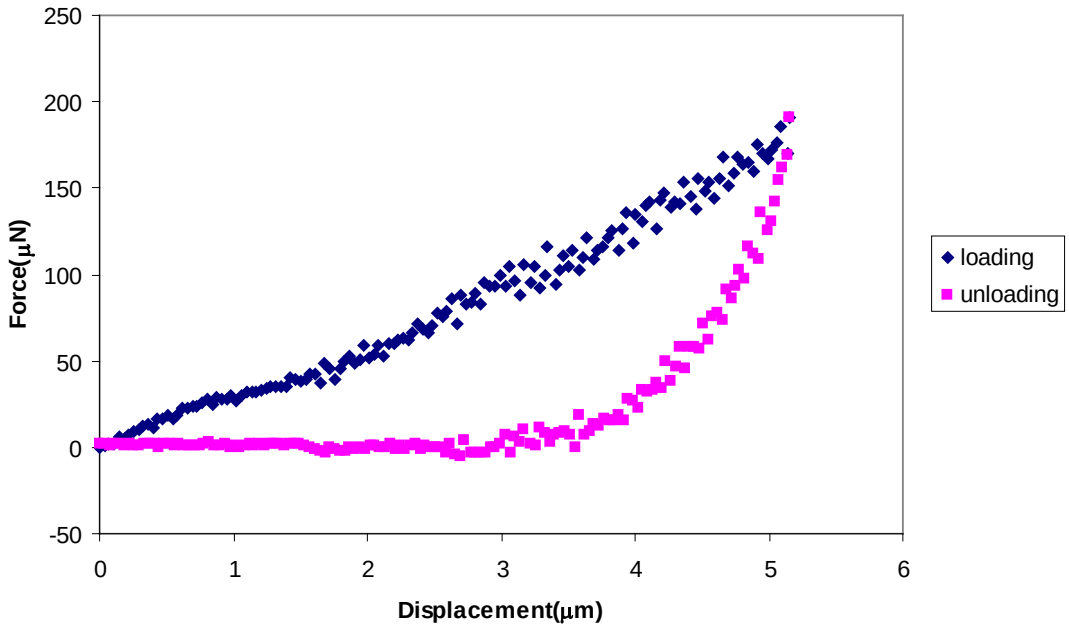
(a)


80
70
60
50
40
30
20
10
0
-10
Force(μN)
0
0.5
1
1.5
2
2.5
Displacement(μm)
loading
unloading

(b)



(c)



(d)

Figure 5.19 Loading and unloading of a single microcapsule at different deformations. (a) 3%; (b) 6%; (c) 10% and (d) 18%

In comparison with other polymer walls, polyamide presents a lower elastic limit, around 10%, in contrast with 19% of melamine-formaldehyde and 17% of urea-formaldehyde (Sun and Zhang, 2002).

5.5. Conclusions

Perfume microcapsules made in previous chapter have been characterized and results show that they can be of industrial interest. Using low temperatures of reaction and a correct combination of monomers and addition times of the aqueous monomers to the reaction system it is possible to obtain microcapsules with low leakage and high mechanical resistance.

The **loading** of the microcapsules produced was very high (83-97% depending on the formulation) due to the thin walls obtained using this method. The **encapsulation efficiency** was defined in two different ways:

- As the percentage of the total perfume that has been encapsulated, which in this case is called Total Encapsulation Efficiency and has a value between 93 and 99.8% depending on the formulation.
- As the percentage of the total perfume that it is possible to extract from the microcapsules, which in this case is called Useful Encapsulation Efficiency and has a lower value (specially for formulations where HMDA is present), between 74 and 98%.

The **temperature of reaction** has a big influence on the final properties of the capsules. It was only possible to obtain stable capsules when the reaction temperature was below 18-20°C, when trimesoyl chloride and diethylenetriamine monomers were used, and lower

temperatures were required for other aqueous monomers. However below this temperature limit there was not a big difference in the properties of capsules prepared at different temperatures although microcapsules were slightly stronger as the temperature of reaction was lower.

It has also been found that the monomers used in the formation of the capsule wall reacted with the perfume at room temperature. The organic monomer was dissolved at 1°C, the emulsion was formed in a short time and the reaction was carried out at low temperature to minimize the loss of monomer by reaction with the perfume.

The use of trimesoyl chloride as **organic monomer** (trifunctional aromatic monomer) highly improved the strength and reduced the leakage of the microcapsules. The explanation of this effect is that the use of a trifunctional organic monomer might lead to cross-linked walls, which provided a better compaction of the polymer wall.

In contrast with the important effect of the trifunctional monomer in the organic side of the reaction, the use of a multifunctional monomer in the **aqueous** side (diethylenetriamine and trietrhylenetetramine) led to worse capsules than the ones made with hexamethylenediamine. The microcapsules made with the last monomer had the highest mechanical resistance and lower leakage among all the microcapsules made with the single aqueous monomers studied. But it has been found that using a combination of organic and aqueous monomers and adding the aqueous monomers at different reaction times the properties of the capsules can be improved. The best formulation produced is the All50DETA (for d_{32}:24.7μm, wall thickness:

301 ± 18nm, loading: 92.9 ± 0.4%, total encapsulation efficiency: 99.7 ± 0.1%, useful encapsulation efficiency: 77.9 ± 0.1%, leakage at 3h: 2.7 ± 0.6%.

The **viscoelastic and plastic properties** of the microcapsules have been studied. “Compress and hold” and “compress and release” experiments have been done to the microcapsules and results show that the polymer wall presented viscoelastic (mainly elastic) behaviour at small deformations (with an elastic limit of approximately 10% deformation), and plastic behaviour at higher deformations.

CHAPTER 6:

ENCAPSULATION OF A WATER SOLUBLE ACTIVE: GLYCEROL

The most exciting phrase to hear in science, the one that heralds the most discoveries, is not "Eureka!" (I found it!) but "That's funny..."

Isaac Asimov

Summary

Glycerol microcapsules with polyamide walls have been produced. The effect of the oil used to prepare the emulsion and the addition of an electrolyte in the system to stabilize this emulsion has been investigated and the best conditions to form stable glycerol-in-oil emulsions selected.

The effect of the monomer formulation and the reaction conditions on the particle size distribution of the microcapsules has been studied. Finally some results on the mechanical strength and the encapsulation efficiency of the encapsulation process are presented.

6.1. Introduction

Polyamide microcapsules have been demonstrated to have desirable properties for the encapsulation of perfume oils. To prove their flexibility in contrast to other technologies (like in-situ polymerisation) a water-soluble system has also been selected as internal phase. The water phase selected was glycerol.

6.1.1. Glycerol

Glycerol (also called glycerine or glycerin) is a colourless, odourless, and viscous liquid (Morrison, 2000). It has three hydrophilic hydroxyl groups that are responsible for its solubility in water and its hygroscopic nature. Glycerol is non toxic and has slightly sweet taste. It has also the capacity of dissolving flavours and dyes, it has plasticizer and antioxidant properties, it is a known antiseptic (it kills bacteria, yeast and fungi by contact, by drowning out water from them) and is easily biodegradable (ABG, 2008). Its chemical structure is shown in Figure 6.1.

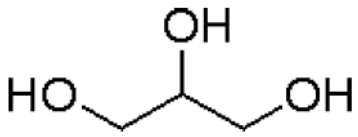


Figure 6.1. Chemical structure of glycerol.

Glycerol has been historically a byproduct of the soap-making industry (saponification of fats) and over the last years it has also become a byproduct of the production of biodiesel (transesterification). In both cases the product obtained presents many impurities (water, salts, methanol, fatty acids), which is called “crude glycerol” and needs further purification for most of its uses. It can also be produced from petroleum derivatives but this route has been almost abandoned due to the increase of the biodiesel production, since it is no longer profitable except for very high quality products.

Glycerol is sold in crude or refined form. There are three basic grades of refined glycerol (generally sold as 99.5% and 99.7% purity) differentiated by potential end uses (ABG, 2008):

1. Technical grade: used as a building block in chemicals, not for food or drug formulations.
2. Pharmaceutical grade: obtained from animal fat or vegetal oil sources and suitable for food products and pharmaceuticals.
3. Kosher grade: obtained from vegetal oil sources and suitable for use in kosher foods.

The properties of glycerol create a versatile product that can be put towards many end-uses. In fact, there are over 1,500 end-uses for the chemical. In most products, however, it is only used in very small portions. There are only a few end-uses which require a significant amount of glycerine in their formulation.

The main uses of refined glycerol (see Figure 6.2) are:

- Food products: In foods and beverages, glycerol serves as a humectant, solvent and sweetener, and may help preserve foods. It is also used as a filler in commercially prepared low-fat foods and as a thickening agent in liqueurs. As a sugar substitute, it has approximately 27 calories per teaspoon and is 60 percent as sweet as sucrose. Although it has about the same food energy as table sugar, it does not raise blood sugar levels, nor does it feed the bacteria that form plaques and cause dental cavities. Glycerol can also be used as a lubricant in food manufacturing facilities.
- Personal care products: The properties of glycerol are ideal ingredients in many personal care products, mostly helping to prevent moisture loss. Thus, glycerol is used as an emollient in skin creams, lotions, shaving creams, makeup and deodorant.
- Oral care products: Glycerol is commonly found in toothpastes, mouthwashes and sugar-free gum, giving these products a sweet taste without contributing to tooth decay. Gel toothpastes generally contain more glycerol than traditional toothpastes because it helps to provide a smooth appearance.
- Tobacco humectant: Glycerol is often sprayed on leaves before processing to prevent crumbling and dehydration. It is used as a plasticizer in cigarette papers as well as a sweetener in chewing tobacco.
- Polyether polyols for urethanes: Glycerol provides one of the basic chemical building blocks for the construction of rigid polyurethane foams.
- Pharmaceuticals: Glycerol provides lubrication and smoothness to many cough syrups and elixirs. It can be used as a plasticizer in gel caps and is an active ingredient in the emergency heart medicine, nitroglycerine.

- Miscellaneous: About 3% of refined glycerin is used for the formulation of alkyd resins. Alkyd resins are used as protective surface coatings, components of plastics, and paints. Glycerin is also a component of nitroglycerine explosives.

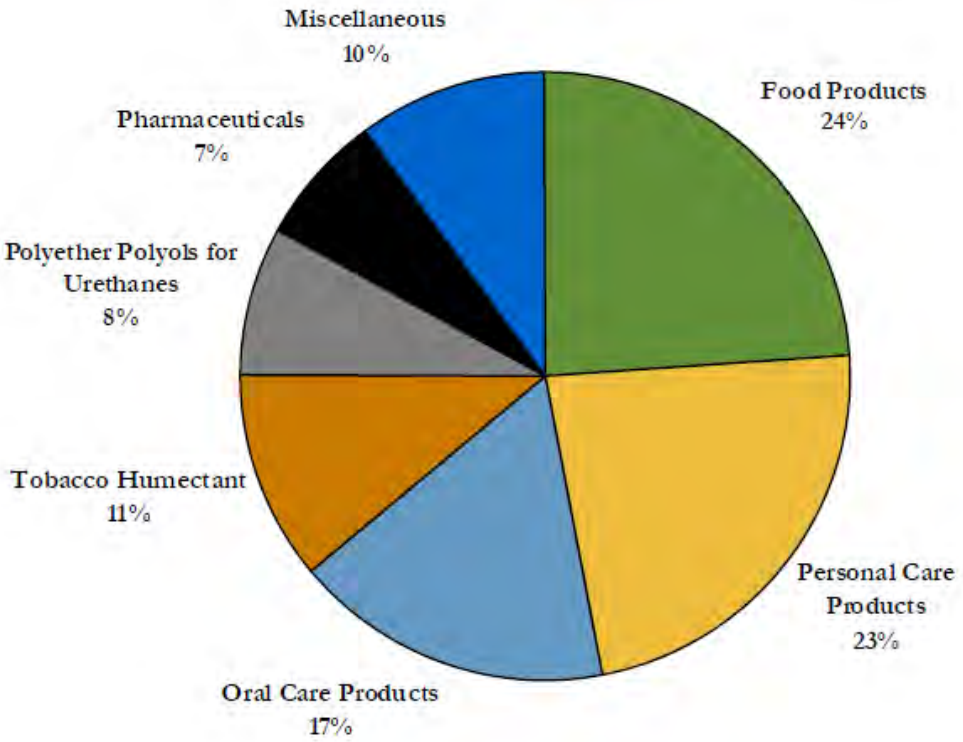


Figure 6.2. End use of refined glycerol. Adapted from ABG (2008).

Crude glycerol can be refined to be used as described before or it can be used as energy source for incineration. Glycerol burns well at high temperatures (it is toxic at 392-572F) and is useful for heating industrial boilers. This is one use of glycerol that consumes large amounts and does not require refining. However, this is considered the lowest valued use of glycerol, as it adds little value to biodiesel producers when the glycerol is sold for waste byproduct prices.

Over the last years a great deal of research has been conducted to make value-added products from the increasing amounts of crude glycerol produced from the biodiesel industry (usually crude glycerol produced is 10% of the biodiesel). Some of these new outlets are in development phase (Claude, 2009; ABG, 2008):

- Second generation biofuels: One of the most exciting developments on the horizon is research related to converting glycerol into renewable fuels, known as second generation biofuels. These fuels would improve the yield of the biodiesel process, resulting in less glycerol production to either market or dispose of. It has also been identified that there is a strain of *E.Coli* that converts glycerol to ethanol in an anaerobic environment (ABG, 2008).
- Livestock feeds: Another potential use of glycerol is corn replacement in cattle, swine and poultry feed.
- Industrial chemicals: There are opportunities to use glycerol as building blocks of industrial and organic compounds. New processes are being developed to produce valuable products from crude glycerol:
    - Epichlorohydrin: Solvay Chemicals has patented a process (ABG, 2008) which converts glycerol from biodiesel into epichlorohydrin. This compound is then used as a component of UV coatings, resins, and paper reinforcement.
    - 1,3-propanediol: Researchers have identified several strains of bacteria which are capable of converting glycerol into 1,3-Propanediol. This compound, much like epichlorohydrin, is useful in UV curing, adhesives, polyesters and laminates.
    - Hydrogen: Although hydrogen fuel cells are very much in their developmental stage, glycerol processing often yields fuel-grade hydrogen as a co-product.

The refined glycerol market is relatively small, with a global production of around 900,000 ton annually (2004) and a market value of $1 billion worldwide (ABG, 2008). But as pointed before the tax incentives and subsidies given to the production of biodiesel are increasing the production of glycerol. By the year 2020 the generation of crude glycerol from biodiesel is projected to reach 2.7 million ton (ABG, 2008), creating an excess of it in the market and subsequently a decrease on the prices. It is expected (Global Industry Analysts, 2010) that the global market of glycerol will reach 2 million ton by the year 2015, mainly driven by growing demand from the oral and personal care market, specially in developing countries including Asia-Pacific, Latin America, Middle East and Africa.

As stated above, glycerol is a basic chemical product used in many fields including in personal care products (moisturizers, creams and lotions, soaps, deodorants and make up products such as mascara and lipstick) because of its humectant properties. Our industrial partner is interested in studying the possibility of encapsulating glycerol to include it in lipsticks formulations.

Lipsticks are made with waxes and oils where glycerol is not really soluble which make glycerol to leak (it is liquid at room temperature) if it is added in high concentrations, creating an undesired lipstick appearance and making them difficult to use. The encapsulation of glycerol will make the formed glycerol microcapsules to be easily dispersed in the wax, preventing these undesired effects and making it possible to release the glycerol on the lips after the lipstick is applied by a mechanical force.

In addition, over the last years the increase in the production of biodiesel has provoked a huge increase in the production of glycerol, which enables supply of cheap glycerol for new applications as the demand is lower than the offer. The formation of glycerol microcapsules should expand its applications in industry and provide a new block to build new products of interest.

6.1.2. Glycerol encapsulation

The only reference found in literature on glycerol microencapsulation (Newell, 1980) is a patent describing the encapsulation of a Lewis acid-glycerol complex catalyst using polyurethane walls (formed with isocyanates and epoxy resins). There has been no report on the use of polyamide walls to encapsulate glycerol.

One of the most important steps in the preparation of glycerol microcapsules using interfacial polymerisation techniques is the formation of a stable glycerol-in-oil emulsion. To stabilize a water-in-oil emulsion it is required to add an electrolyte in the water phase (Kanouni *et al.*, 2002). It has been suggested in the literature (Leal-Calderon *et al.*, 1997; Aronson and Petko, 1993) that 2% wt of $MgSO_4$ should be used in the dispersed phase to stabilize glycerol-in-water emulsions. According to these authors the addition of some electrolytes ($MgSO_4$ is preferred) prevents the emulsion coarsening, therefore they favour the stability of the emulsion and the formation of microcapsules.

6.1.3. Glycerol measurement

Glycerol concentration can be measured using different techniques. The first procedure used was the periodate method (*e.g.* ISO 2879:1975). When biodiesel started being popular more

different methods were developed, firstly based on GC (*e.g.* EN 14105:2003) and later on enzymatic analysis (*e.g.* BQP 02 kit from Sigma Aldrich). Other methods used the periodate reaction to form formaldehyde from glycerol and measured formaldehyde by HPLC (Wu *et al.*, 2003). Bondioli and Della Bella (2005) developed a new method based on the periodate reaction, but once formaldehyde was produced, they reacted the formaldehyde with acetylacetone in the presence of ammonium acetate, leading to the formation of 3,5-diacetyl-1,4-dihydrolutidine, generally known as Hantzsch's reaction. This compound presents a very high specific absorption at 410nm, which makes it ideal to be detected by spectrophotometry even at low glycerol concentrations.

Bondioli's method has been the one selected to be used in this work due to their simplicity and the availability of the spectrophotometer in the lab. This method is explained in detail in 6.3.4.

In this chapter a process to encapsulate glycerol and the effect of different parameters on the stability and properties of these new microcapsules are presented.

6.2. Materials and methods

6.2.1. Chemicals

Isopropyl myristate (IPM), span 85 (sorbitan trioleate), glycerol, magnesium sulphate heptahydrate ($MgSO_4 \cdot 7H_2O$), hexamethylenediamine (HMDA), diethylenetriamine (DETA) and trimesoyl chloride (Trim) were supplied by Sigma-Aldrich (Dorset, UK) and were used as received without further purification.

The reactants for glycerol measurement: glycerol, ethanol 95%, acetic acid, ammonium acetate, sodium periodate and acetyl acetone were also supplied by Sigma-Aldrich and used without further purification.

6.2.2. Interfacial polymerisation

The basic steps of the process used to produce glycerol microcapsules are shown in Figure 6.3.

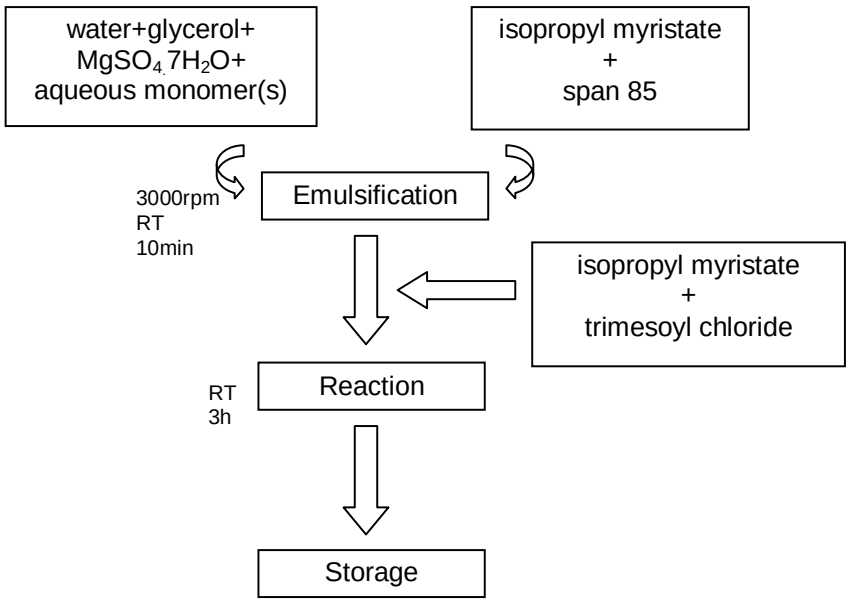


Figure 6.3. Illustration of preparation steps of the interfacial polymerisation method.

Firstly a 60% (vol.) solution of glycerol in water was prepared by mixing 7.57g of glycerol and 4g of water in a 25ml beaker. To this solution were added 10meq of NH_2 functions in the form of HMDA (0.58g), DETA (0.34g) or a combination of both and 1g of $MgSO_4 \cdot 7H_2O$. The solution was well mixed and the salt dissolved with the help of a magnetic stirrer. (Solution 1)

In a 250ml beaker with 4 glass blades built-in (by the Glass blower in the School of Chemistry of the University of Birmingham) 5ml of Span® 85 and 60g of IPM (approx. 70ml) were added. The solution was homogenized with a mechanical stirrer. (Solution 2)

A third solution with an organic monomer is prepared. 4.5g of trimesoyl chloride (50meq COCl functions) were dissolved in 8.7g IPM with the aid of a magnetic stirrer at room temperature. (Solution 3)

A Silverson homogenizer (Silverson L4RT, Silverson, Chesham, UK) was used to prepare the emulsion. It was placed in the 250ml beaker (solution 2) at 3,000rpm and the glycerol solution (solution 1) was added to it dropwise. After 10min emulsification the homogenizer was stopped and the beaker was placed under mechanical stirring using a 4cm diameter Rushton turbine (1,000rpm at the beginning, 500 after10min). Solution 3 was added to it and the reaction between the two monomers quickly started. After 3h reaction at RT (because of the heat generated during the emulsification and reaction, the maximum reaction temperature achieved was 38°C) the stirrer was stopped and the resulting microcapsule suspension was stored at room temperature in a glass bottle.

Other continuous phases (paraffin oil, mineral oil) were tried but it was not possible to form a stable emulsion of glycerol in them due to their high viscosity. When paraffin or mineral oil was used the temperature increase during emulsification was also very high reaching the oil temperatures of more than 65°C after 10min at 4,000rpm. Isopropyl myristate was found to have lower viscosity than them which made it possible to create this emulsion. The addition

of magnesium sulphate in the system stabilized greatly the glycerol-in-oil emulsion and permitted to obtain small microcapsules.

6.2.3. Particle measurement

An optical microscope (Leica DMRBE, Leica GMBH, Cambridge, UK) equipped with a camera was used to take micrographs of the samples and its software for photographic analysis (Leica QWin Standard v2.8) was used to measure the size of the glycerol microcapsules.

Mastersizer was also tried to measure particle size (like for the perfume microcapsules), but when glycerol microcapsules were added to the water in the sampler unit they aggregated and the data obtained was not representative of the real size distribution. It would have been needed to work with an organic phase instead of water in the sampler unit.

6.3.4. Glycerol analysis

Glycerol concentration has been measured with a spectrophotometric method (Bondioli and Della Bella, 2005) based on periodate oxidation of glycerol to formaldehyde and its reaction with acetylacetone in the presence of ammonium acetate to form 3,5-diacetyl-1,4-dihydrolutidine, which is easy to measure with the aid of a spectrophotometer at 410nm.

Some working solutions should be prepared before the analysis:

- Acetic acid stock solution: a 1.6M (9.6g/100ml) aqueous solution was prepared.
- Ammonium acetate stock solution: a 4.0M (30.8g/100ml) aqueous solution was prepared. Both solutions were stable over time. Mixed in equal volumes, these solutions result in a buffer solution at pH 5.5.

- Acetylacetone solution, 0.2M: 200µl (195mg) of acetylacetone were dissolved in 5mL of acetic acid stock solution and 5ml of ammonium acetate stock solution. This reagent was prepared daily.
- Sodium periodate solution, 10mM: approx. 21mg of sodium meta periodate were dissolved in 5ml of acetic acid stock solution, it was needed to swirl to dissolve the periodate. After periodate was completely dissolved, 5ml ammonium acetate stock solution were added. This reagent was prepared daily.
- Working solvent: equal volumes of distilled water and 95% ethanol were mixed. This solvent was used for sample extraction, reaction and mother glycerol reference solutions.
- Glycerol reference stock solution: approx. 150mg of glycerol was weighted, and put into a 50-mL calibrated flask. It was dissolved with the working solvent and filled up to the mark. This solution contained approx. 3mg/ml of glycerol.
- Glycerol reference working solution: using a precision pipette, 1ml of glycerol reference stock solution was transferred to a 100-ml calibrated flask. It was diluted and filled up to the mark using the working solvent. This solution contained approx. 30mg/l of glycerol. Both solutions are stable for some weeks.

The calibration curve was prepared by adding in 5 10ml glass test tubes, 0, 0.5, 1, 1.5 and 2ml of the 30mg/l glycerol solution. Each tube was filled till 2ml with the working solvent. 1.2ml of 10mM sodium periodate solution was added to a tube and it was shaken for 30s. After that 1.2ml of 0.2M acetyl acetone solution was added to the tube and this was put in a water bath at 70°C for 1min, stirred manually. After the reaction time the sample was immediately cooled by immersing the tube in a beaker with tap water (always at 20-25°C). The procedure

was repeated for the rest of the tubes and the samples were read in a spectrophotometer at 410nm. The resulting curve is presented in Figure 6.4. The slope of the curve, used in the following calculations was 0.108±0.003. It was supposed that the volumes of the different reactants were additive, resulting in a final volume of 4.4ml for each sample, which was used to calculate the final glycerol concentration in the tubes: 0, 3.5, 6.9, 10.4 and 13.8 mg/l respectively.

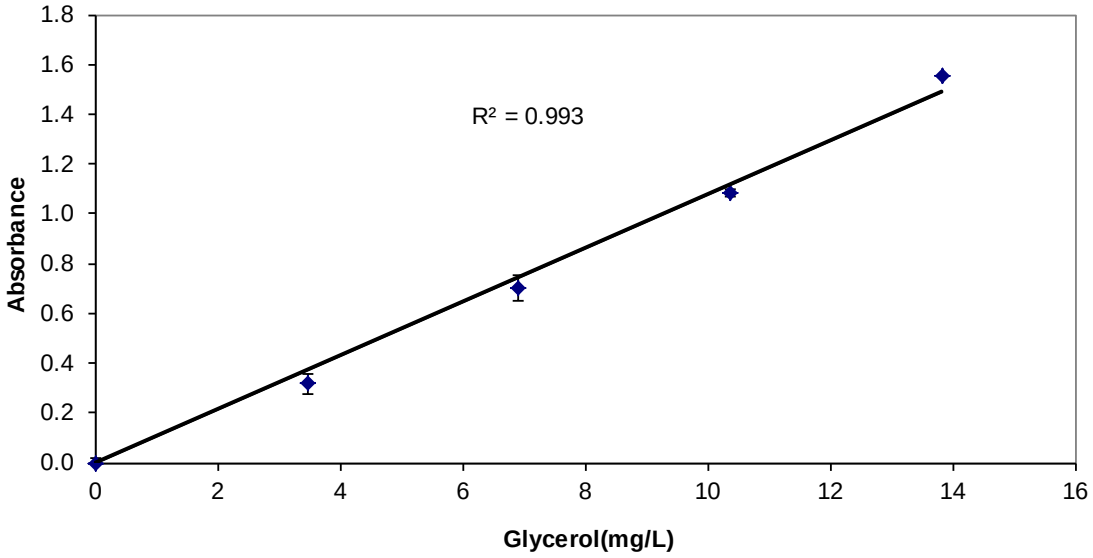


Figure 6.4. Calibration curve for glycerol using Bondioli's method at 410nm.

Once the calibration curve was obtained, it was used for measuring the encapsulation efficiency of the glycerol encapsulation process. 1g of slurry was weighted in a 15ml Falcon tube and 4ml of working solvent was also added. The tube was placed in a Vortex mixer at 3,000rpm for 5min to break the capsules and release the glycerol from the IPM phase to the ethanol/water one. After this time the tube was centrifuged at 2,000rpm (460g) for 15min. After centrifugation the top phase (IPM with broken capsules) was removed with a Pasteur pipette and exactly 0.5ml of the lower phase was transferred into a 10ml glass test tube. 1.5ml of working solvent was added to it. 1.2ml of 10mM sodium periodate solution was added to

the tube and it was shaken for 30s. After that 1.2ml of 0.2M acetyl acetone solution was added to the tube and this was put in a water bath at 70°C for 1min, stirred manually. After the reaction time the sample was immediately cooled by immersing the tube in a beaker with tap water (always at 20-25°C). The sample was read in the spectrophotometer at 410nm, against a blank sample processed in the same way after addition of 2ml of working solvent to the tube.

6.2.5. Mechanical characterisation of microcapsules

5ml of slurry from a batch was taken with an automatic pipette and transferred to a 15ml plastic tube. The sample was centrifuged at 500rpm (30g) for 10minutes and a phase separation (microcapsules at the bottom, organic layer at the top) was obtained. The top organic layer was removed and 5ml hexane were added to the plastic tube, then microcapsules were carefully resuspended with a 1ml automatic pipette. A sample from the hexane-microcapsules mixture was placed on a glass slide and left to dry at room temperature. Microcapsules from the slide were compressed using the micromanipulation technique described in Chapter 3.

6.3. Results and discussion

6.3.1 Experimental formulations

Five different experimental formulations have been studied. Details of them can be found in Table 6.1.

	Formulation (meq COCl or NH_2)				
Experiment	Trim	DETA	HMDA	T(°C)	Stirring rate(rpm)
I	100	-	10	RT (38°C)	3,000
II	50	10	-	RT (35°C)	3,000
III	50	5	5	RT	3,000
IV	50	10	-	5°-RT	3,000
V	50	10	-	RT (30°C)	1,500

Table 6.1. Formulation of the glycerol microencapsulation experiments.

The effect of the temperature of reaction on the quality of the microcapsules formed was first studied. In experiments I, II, III and V the temperature was not adjusted (but it was measured), the temperature at the beginning of the reaction was slightly higher (30-38°C) due to heat dissipation during emulsification and the heat released during reaction. After these first minutes of reaction the temperature was room temperature (RT) for the rest of the process. However experiment IV was done in an ice bath, the emulsification and the temperature of reaction during the first 30min were kept at around 5°C , but no more ice was added and the temperature went to RT after that.

The influence of the stirring rate during emulsification on the final particle size was also investigated. In experiments I to IV a stirring rate of 3,000rpm was used during emulsification, while in experiment V only 1,500rpm was used.

6.3.2. Size distribution

In preliminary experiments other organic phases were used (mineral oil, paraffin oil), but due to their high viscosity no glycerol emulsions were prepared. Glycerol formed a continuous phase inside the viscous oil, not forming droplets therefore avoiding the production of microcapsules.

When IPM was used as organic phase, it was found that it was possible to form a glycerol-IPM emulsion and following the procedure described in section 6.2.2 (although no salt - $MgSO_4$- was added yet) glycerol capsules of more than 1mm diameter (using 4,000rpm as stirring rate during emulsification) were obtained (Figure 6.5). In other experiment the reaction was carried out directly in the Silverson homogenizer (instead of moving the beaker to a mechanical stirrer) at 4,000rpm (instead of 1,000rpm) for the first 10min of reaction (reducing the stirring rate to 1,000 after this time) and glycerol capsules of a smaller diameter (minimum obtained was around 150μm) were obtained (Figure 6.6).

Both capsules were separated by filtration and stored in dry conditions. Over time it was possible to see that glycerol was leaking from them.

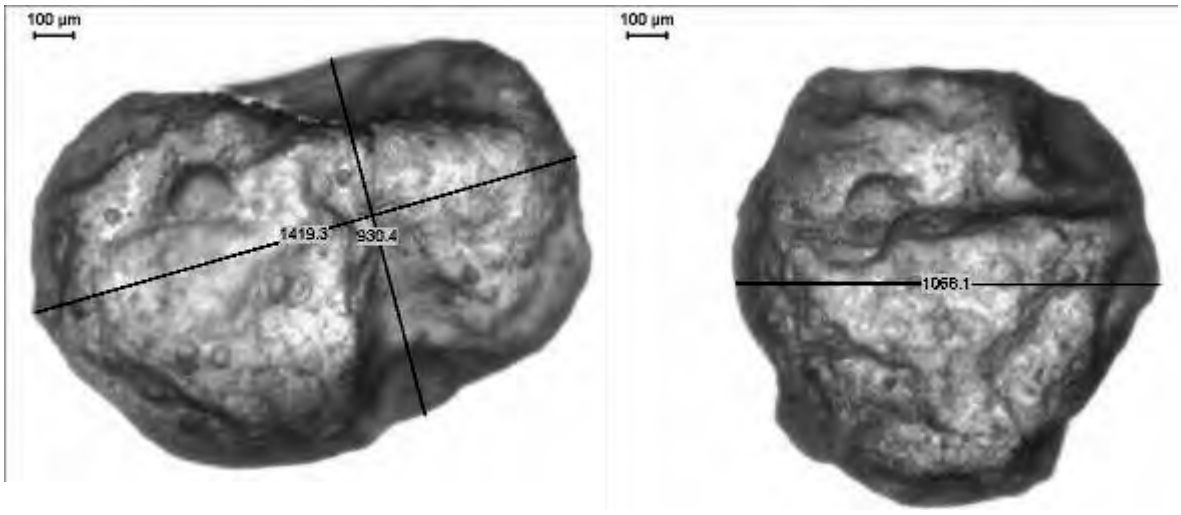


Figure 6.5. Polyamide-glycerol capsules prepared following the procedure without using salt.

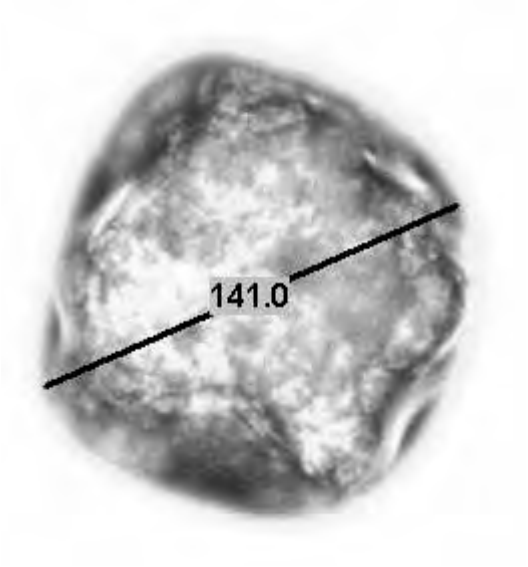


Figure 6.6. Smallest polyamide-glycerol microcapsule prepared using the homogenizer but without salt.

From these experiments it was possible to see that the glycerol-in-IPM emulsion formed was not stable over time. The smaller droplets formed during emulsification at 4,000rpm (they would have the same size as the microcapsules formed directly in the homogenizer) coalesced during the short time period from when the homogenizer was stopped until the acid chlorides were added to the system (the beaker was moved from the homogenizer to the mechanical stirrer) and formed bigger droplets which were the templates for the bigger microcapsules made under mechanical stirring.

The addition of the salt ($MgSO_4$) to the system stabilized greatly the emulsion and permitted the production of much smaller microcapsules (Figure 6.7). The average diameters of the microcapsules produced in experiments I to V are shown in Table 6.2. To calculate these diameters micrographs were taken and analyzed with photographic software (the number of microcapsules measured was 105, 106, 85, 61 and 267 respectively), the size distribution of the different samples was also calculated (Figure 6.8).

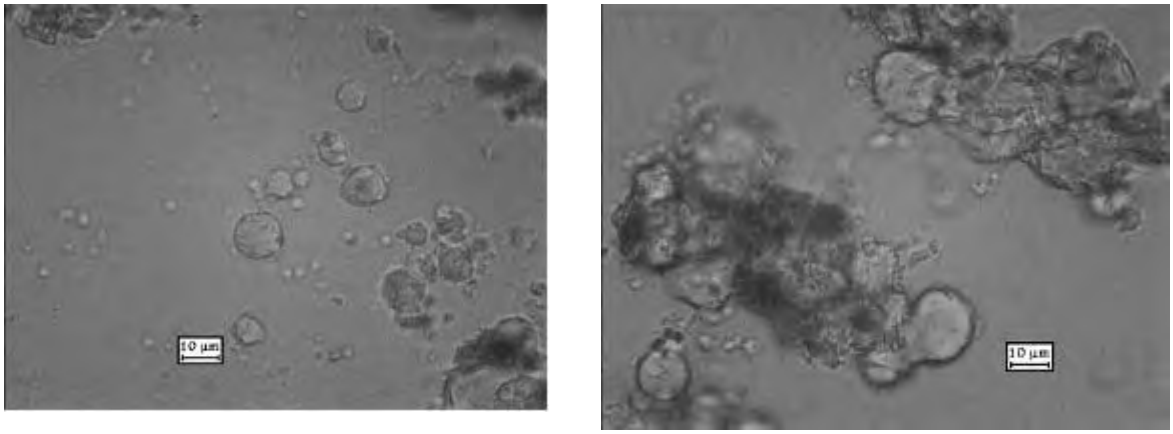


Figure 6.7, Polyamide-glycerol microcapsules prepared using $MgSO_4$ as stabilizer.

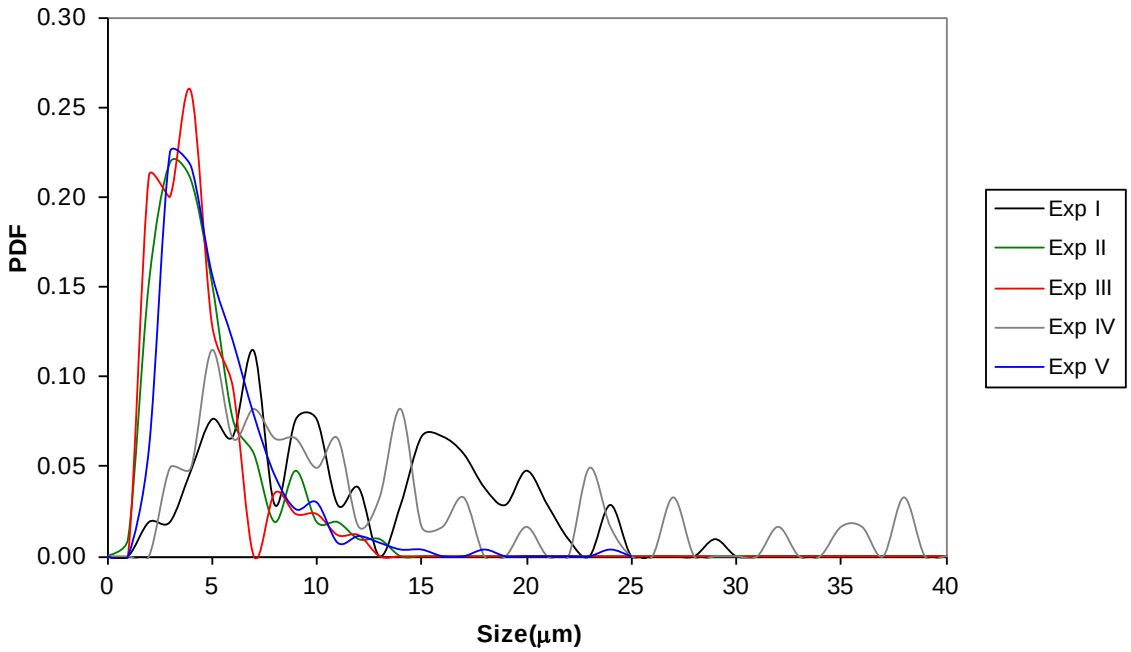


Figure 6.8. Size distribution of glycerol microcapsules prepared under different conditions, see Table 6.1.

It is possible to see that the size distribution of microcapsules prepared by experiments II, III and V was very similar and all of them present a clear peak at around 5µm. In experiment IV a lower temperature was used and the microcapsules had a wider size distribution. Experiment I also generated a wider size distribution of microcapsules, which might be due to the use of a different amine.

Sample	d_{10}(μm)	d_{32}(μm)
I	12.2±0.6	17.5
II	5.2±0.3	8.1
III	4.8±0.2	7.0
IV	13.2±1.2	25.8
V	5.7±0.2	9.0

Table 6.2. Number based average diameter and Sauter diameter of polyamide-glycerol microcapsules.

It has been observed that adding $MgSO_4$ to the system made it possible to obtain microcapsules in the range of 1 to 10μm in diameter, more than 100 times smaller than capsules prepared without using the salt. It is also possible to see that the size of the microcapsules prepared using 1,500rpm during emulsification (experiment V) was almost the same as the size of the capsules made at 3,000 rpm using the same monomer compositions and temperature of reaction (experiment II). It is clear that the addition of the salt in the system stabilized the emulsion and it seems that it also controlled the capsule size, although more experiments using different salt concentrations should be done to validate it.

6.3.3. Stability of the microcapsules

It has been found that glycerol microcapsules produced following this method (Figure 6.9) were very weak and most of them broke when the oil they were suspended in was removed and new fresh oil added, releasing the glycerol to the media (Figure 6.10). This effect was highly magnified if the oil was removed and water was added to the system (Figures 6.11 and 6.12). When water was added to the glycerol microcapsules, an unexpected phenomena was observed: The released glycerol was not dissolved in water, and instead it formed glycerol droplets suspended in water. If the sample was centrifuged, the glycerol was concentrated at the bottom of water forming one phase, but if the tube was shaken they formed an emulsion of small glycerol droplets in water (Figure 6.13, after 10 seconds at 3,000rpm vortex shaking).

This phenomenon may be due to the effect of the surfactant and the sodium sulphate, which stabilize the emulsion, although not a complete explanation has been found.

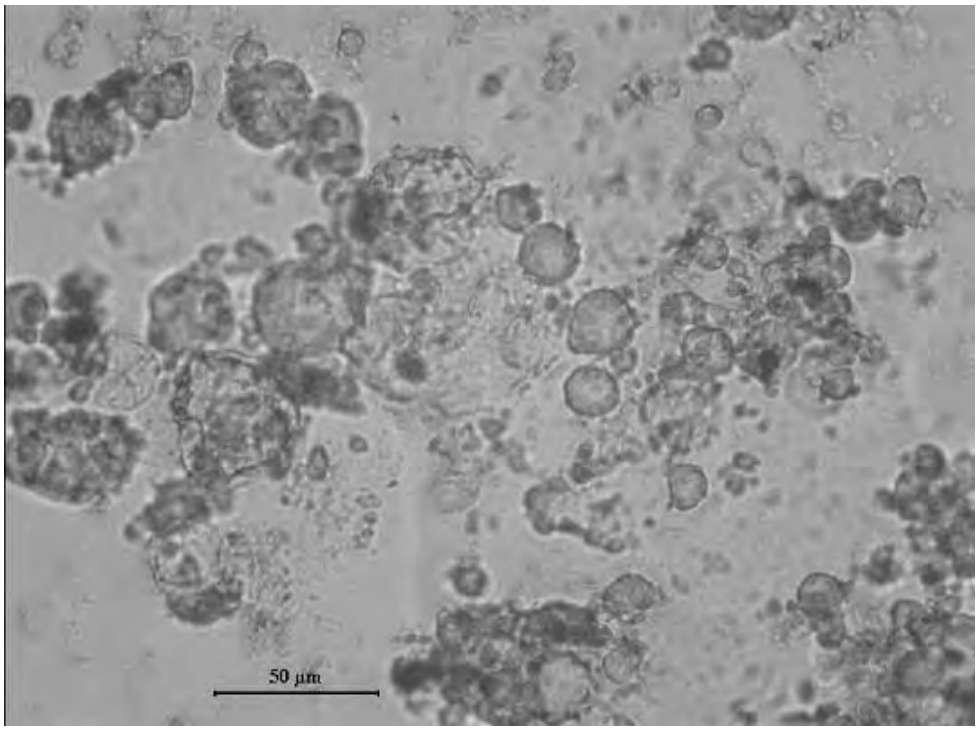


Figure 6.9. Glycerol microcapsules suspended in IPM (sample IV).

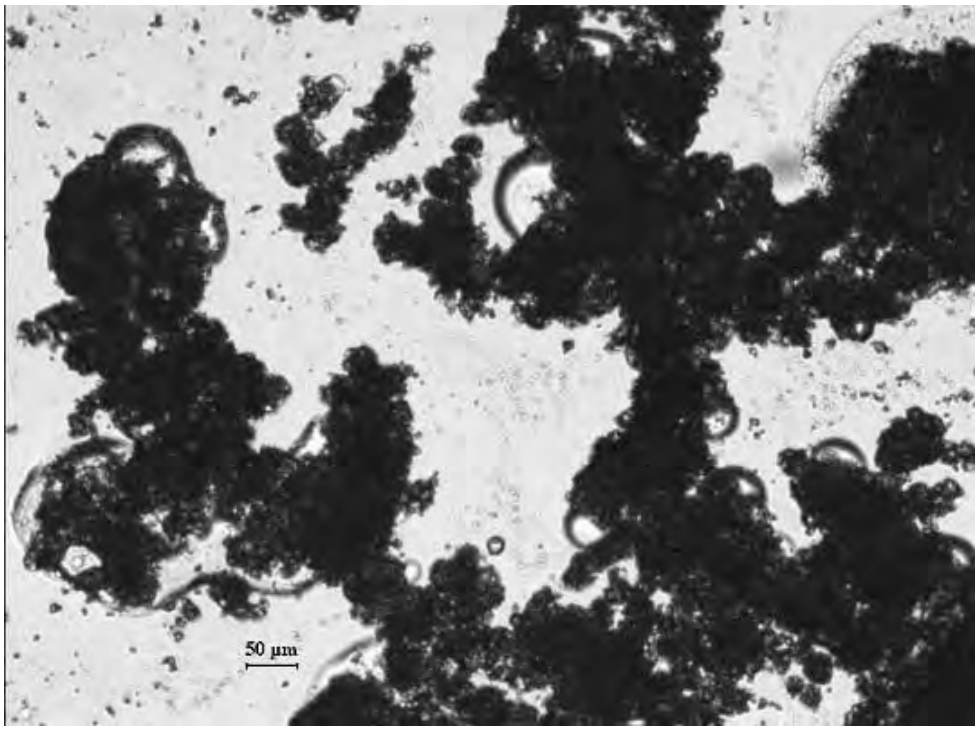


Figure 6.10. Glycerol leaking from microcapsules after changing the oil.

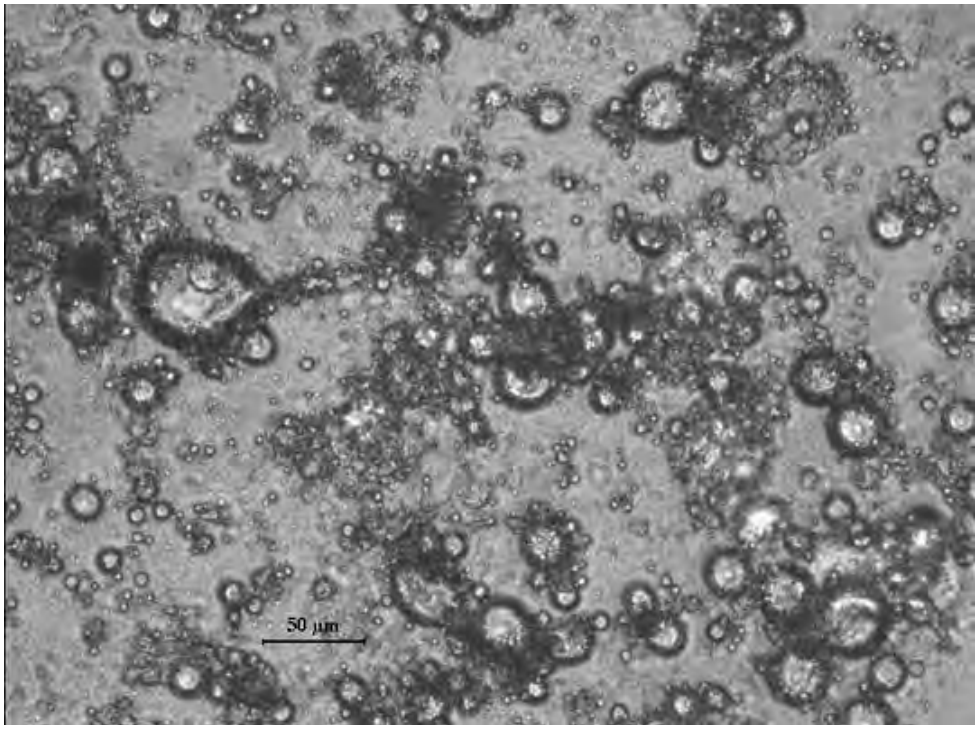


Figure 6.11. Glycerol released after changing the oil continuous phase for water

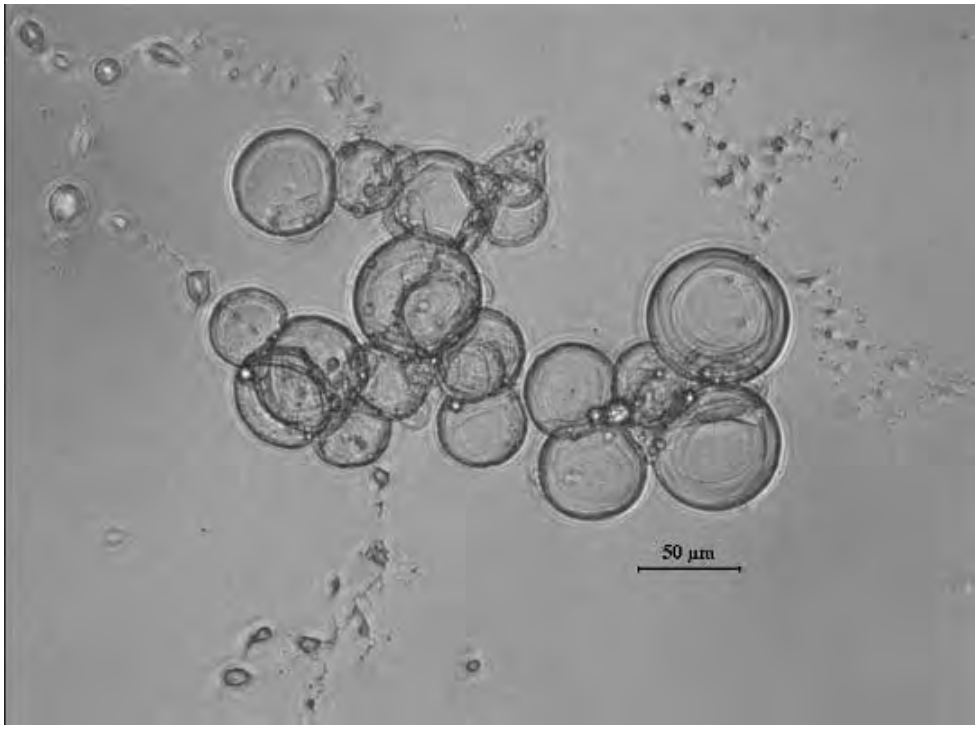


Figure 6.12. Glycerol droplets released from the microcapsules dispersed in water.

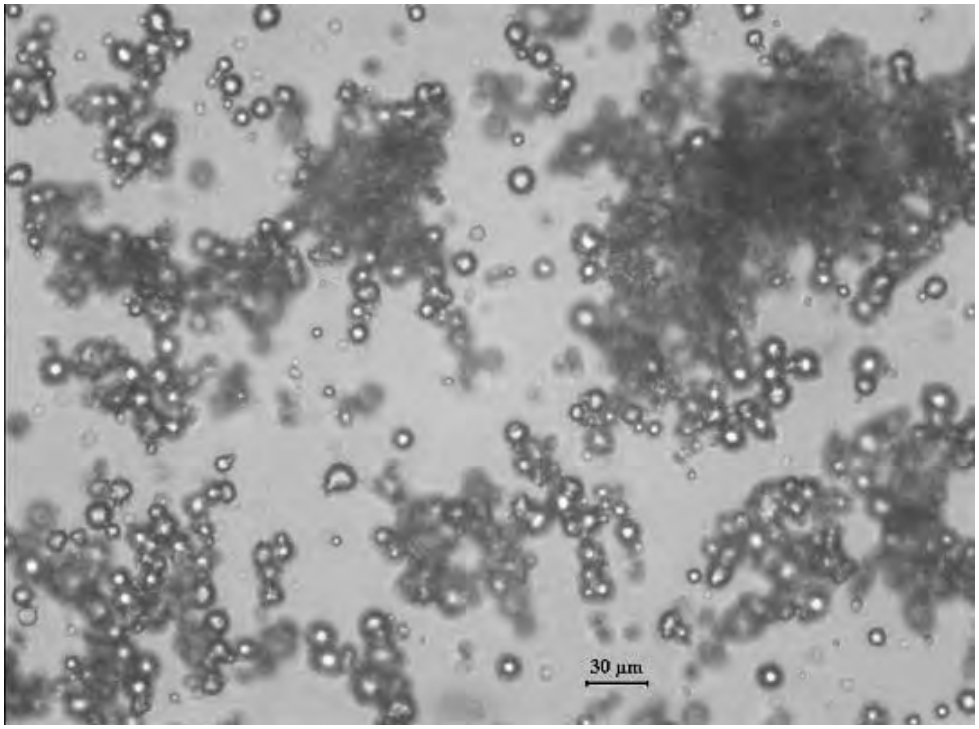


Figure 6.13. Glycerol-water emulsion after shaking glycerol microcapsules in water.

Corresponding to this behaviour it was very difficult to find microcapsules to compress for measuring their mechanical strength using the micromanipulation rig, as mainly glycerol droplets were found on the glass surface not offering any resistance to the compression. Sample IV was compressed but only four microcapsules (less than 10% of the compressed "particles", 90% were glycerol droplets) were found and really compressed. Results of the compressions are shown in Table 6.3.

Diameter(µm)	Force (µN)	Nominal Stress (MPa)	Displacement (µm)
6.3	551	17.7	1.1
10.5	169	2.0	0.9
11.6	892	8.4	1.0
13.7	1078	7.3	0.7

Table 6.3. Mechanical properties (force, nominal stress and displacement at rupture) of glycerol microcapsules.

It was also found that microcapsules had several breakages during the compression (the data shown in Table 6.3 were based on the first breakage point) as it is possible to see in Figure 6.14.

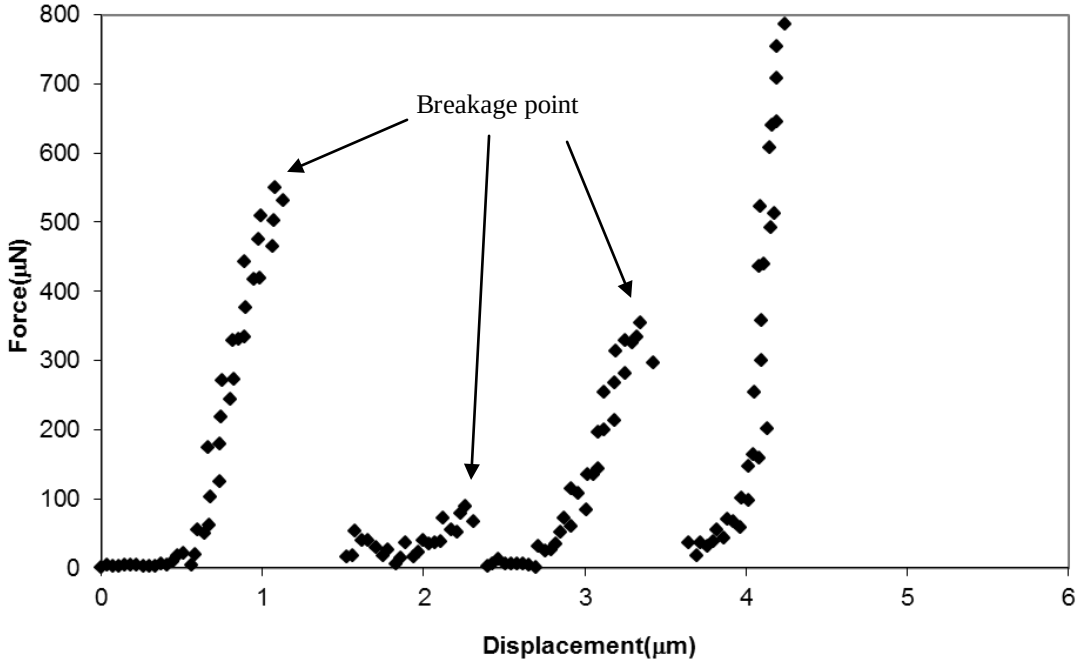


Figure 6.14. Compression of a microcapsule of 6.3μm diameter.

In addition, the data in Table 6.3 suggest that the displacement at rupture was independent of the capsules size. The microcapsules seemed to break when they were compressed by 1μm in all cases. It is however difficult to extract conclusions from these few successful compressions. More work should be done in the future.

6.3.4. Effect of the monomers

Following the previous work developed in perfume microencapsulation (where it was found that trifunctional organic monomers gave a higher stability and better mechanical and leakage properties to the microcapsules) only the trifunctional organic monomer was used to prepare

glycerol microcapsules. But two different amines have been used as aqueous monomers: diethylenetriamine (DETA) and hexamethylenediamine (HMDA).

It has been found that the nature of the amine used had a huge influence on the final properties of the microcapsules formed. When HMDA was used as unique aqueous monomer (Experiment I) the slurry obtained gelled after some weeks during storage while when DETA was added to the system (alone or in combination with HMDA) the slurry formed did not gel and microcapsules were able to move in it, although they settled at the bottom of the bottle.

This behaviour may be explained by considering that DETA is a trifunctional monomer while HMDA is a bifunctional one. It is suggested that DETA may form cross-linked walls resulting in microcapsules more resistant to leakage and permitting them to move in the oil phase. The use of HMDA, on the other side, may produce microcapsules weaker and less resistant to leakage, releasing the glycerol during storage. And it is suggested that when glycerol reached the organic phase it reacted with the excess of trimesoyl chloride present therefore forming a polymeric gel (polyester) in the bulk. This behaviour is opposite to the one found with perfume microcapsules, where the use of HMDA reduced greatly the release rate of perfume from inside the microcapsules.

6.3.5. Encapsulation efficiency

The method developed by Bondioli and Della Bella (2005) to measure glycerol in biodiesel was tried on the glycerol-microcapsule slurry but the readings obtained in the UV spectrophotometer were much higher than the ones calculated for 100% glycerol recovery suggesting that undesired reactions took place during the sample preparation for analysis

and/or that the presence of the monomers had a dramatic effect in the process. Therefore no quantitative method has been used to measure encapsulation efficiency.

From micrographs shown before it is possible to see that microcapsules (Figure 6.9) had an irregular shape and are opaque while glycerol droplets were almost spherical and transparent (Figures 6.11, 6.12, and 6.13). Therefore it is supposed that the encapsulation efficiency was very high, close to 100% as no glycerol droplets can be seen in Figure 6.9 and other microcapsule micrographs. This high value of encapsulation efficiency is supported by our previous experiments on perfume microcapsules where encapsulation efficiencies of almost 100% were obtained.

6.4. Conclusions

- Glycerol microcapsules with polyamide walls have been successfully prepared. A method to produce them has been developed and the best working conditions have been selected.
- A continuous organic phase with low viscosity has to be used to produce capsules. The use of mineral or paraffin oil produced a polymer in the bulk but not capsules.
- The use of an electrolyte in the water phase stabilized greatly the emulsion formed in the homogenizer, permitting the production of much smaller and regular microcapsules. Capsules of glycerol of around 1mm were obtained without salt, when $MgSO_4$ was used the microcapsules obtained had a size of around 10μm.
- The size of the microcapsules produced was governed by the electrolyte, not by the stirring rate during the emulsification step.

- The use of HMDA as aqueous monomer alone (without DETA) produced microcapsules but the slurry gelled during the first weeks of storage. Microcapsules prepared with DETA did not show this behaviour, forming microcapsules that were able to move freely in the oil phase.
- Microcapsules produced were very weak and most of them broke when the liquid they were suspended in was removed, releasing the glycerol. The addition of a water phase on the microcapsules favoured the releasing of the glycerol and formed an emulsion of glycerol droplets in water instead of dissolving the glycerol in the water (perhaps because of the effect of the surfactant or salts added). Glycerol droplets slowly coalesced and formed bigger droplets, but never a continuous phase, unless they were centrifuged.
- Studies on monomer types and concentrations, temperatures of reaction and type and concentration of electrolytes are proposed for future work. The development of an analytical method to measure glycerol release is also suggested.

CHAPTER 7:

CONCLUSIONS AND RECOMMENDATIONS

I have had my results for a long time:

but I do not yet know how I am to arrive at them.

Karl Friedrich Gauss

7.1. Conclusions

7.1.1. Perfume microcapsules

Perfume microcapsules are a new way of dosing perfume in cleaning products. The main advantages of their use are the increase of the perfume efficiency (less perfume is needed in the final product for the same odour performance) and the controlled release of the perfume from the microcapsules at the desired time, which make it possible to reduce raw materials (less cost and less waste) and achieve a sustained perfume release desired by customers.

Current perfume microcapsules in the market are based on in-situ polymerisation techniques, in which aldehydes (usually formaldehyde) are used as crosslinkers to improve the mechanical resistance and impermeability of the walls. But formaldehyde is a known carcinogenic compound and its concentration in final products is highly regulated by law. Due to this restriction new formaldehyde-free perfume microcapsules have been developed.

Interfacial polymerisation techniques have been widely used to encapsulate herbicides and insecticides (which require porous walls) and recently also phase change materials (which require impermeable walls). In this last case usually polyurethanes, polyureas or polyamides have been used. Polyurethane and polyurea are formed using isocyanates, a toxic compound, while polyamide formation does not require the use of any toxic or carcinogenic chemical, therefore polyamide has been selected as suitable wall material for making perfume microcapsules in this project.

Polyamides can be formed at room temperature by the reaction of highly reactive acid chlorides and amines and as a result peptide bonds are formed and hydrochloric acid is

released. The properties of the final polymer obtained depend on many factors and in this work the effect of each of them has been studied.

Due to the release of HCl during the reaction it was possible to monitor the **reaction kinetics** during the process. The effects of the temperature of reaction, the surfactant concentration and the type of aqueous monomer used on the reaction kinetics were studied. As expected it was found that the higher the temperature was the faster the reaction took place. It was also found that the reaction was faster when a lower surfactant concentration was used. This second finding was unforeseen as it was expected that due to the smaller capsules formed when a higher surfactant concentration was used (and smaller capsules have a higher surface to volume ratio) the reaction should be faster. In this case it seems that the surfactant accumulated at the interface and made more difficult the diffusion of the monomers through the membrane. Finally the effect of using three different aqueous monomers (DETA, HMDA and EDA) was studied. It was found that at a constant temperature (0°C) the reaction was mush faster when EDA was used, followed by HMDA and by DETA. This suggests that the smaller the molecule is (EDA) the faster the reaction takes place because it is easier for the molecule to diffuse through the membrane to react at the other (organic) side.

Related to the reaction kinetics, it has been found that perfume microcapsules were only formed under certain conditions. There was a **critical reaction temperature** for each aqueous monomer over which the microcapsules formed were very weak (if formed) and broke when they were dried. The reaction temperature had to be maintained below 20°C when working with Trim and DETA in order to obtain capsules which were strong enough for further processing, such as drying and mechanical characterisation. A lower temperature was used for

other aqueous monomers. It has also been found that the **type of organic monomer** used was crucial in the formation of the microcapsules. Three different organic monomers were tried (using DETA as aqueous monomer in all cases), two aromatic ones (Trim and TC) and one aliphatic (SC), and microcapsules were only produced when the aromatic ones were used. If SC was used, a mass of polymer in the bulk was obtained, but no single microcapsules were formed. It was also found that there was a minimum amount of organic monomer that was needed to be used to form microcapsules.

The **chemistry of the wall** was investigated with a FTIR instrument and results showed that no (or very little) NH_2 remained unreacted and that an amide bond has been formed during the reaction. Polyamide has been produced.

Different **microscopy techniques** have been used to look at the microcapsules formed. *Optical microscopy* showed that the microcapsules produced were not completely spherical, some of them were oval (like a coffee bean) and when they were observed closely it was found that their surface was not regular, with most of them presenting scrapes, protrusions and valleys. When *SEM microscopy* was used it was observed that microcapsules shrank due to the vacuum produced by it and that they showed edges and a not very smooth surface on which it was possible to see some pores. Photographs taken using *TEM microscopy* permitted to see the inner structure of the wall produced and to measure their thickness. The morphology of the wall was very different depending on the aqueous monomers used in the reaction: EDA produced a thin and smooth wall, DETA produced a thicker one and HMDA produced a thick wall with a core material occluded in it, forming “bubbles” of perfume surrounded by polymer next to the wall. Wall thickness was found to be from 100 to 700nm

depending on the formulation. It has been found that when the same monomers were used the average wall thickness increased as the temperature of reaction decreased. The addition of bifunctional organic monomer (TC) in the system increased the average thickness when added in small amounts (10% wt) and decreased it when added in bigger amounts (18 and 25% wt), and in all cases it increased the minimum thickness. Walls obtained with EDA were thinner than the ones obtained with DETA and the ones made with HMDA presented very irregular thicknesses but the minimum thickness was always larger than the one for DETA walls.

The **size and size distribution** of the microcapsules produced have been measured and correlated with theoretical models and the parameters in the equations calculated. The Sauter mean diameter (d_{32}) was well correlated by the Hinze and Kolmogoroff model and the size distribution data has been successfully fitted to a log normal distribution when the Silverson homogenizer was used. When a Rushton turbine was used the size distribution obtained approached a bimodal distribution. The correlated equations were compared with the ones obtained by Calabrese (1986), Brown and Pitt (1972) and Pacek (2002). The distribution obtained in this work is wider than the ones described by other authors, this may be due to the higher coalescence in the system studied in this work and to the different geometry and process conditions.

The **loading** of the microcapsules was calculated with the wall thickness data obtained from the TEM micrographs. The loading of the capsules is high, from 83 to 97% vol. depending on the formulation, which means that the wall relative volume is from 3 to17%.

Perfume concentration has been measured using UV spectrophotometry. The total amount of perfume present, encapsulated, non-encapsulated and the rate of perfume release from microcapsules in a water-hexane system has been calculated.

Due to the peculiarities of the membrane walls created when HMDA was used as aqueous monomer, two different **encapsulation efficiencies** have been defined:

- The Total Encapsulation Efficiency: It is the percentage of the total perfume that has been encapsulated. It had a value between 93 and 99.8% depending on the formulation.
- The Useful Encapsulation Efficiency: It is the percentage of the total perfume that it was possible to extract from the microcapsules. It had a lower value (especially when HMDA was used as aqueous monomer), between 74 and 98%.

The organic and aqueous monomers used in the formulation not only reacted between them but also with the perfume if the reaction is done over room temperature. The **reactivity** at room temperature of both monomers with the perfume used in the encapsulation process was measured using gas chromatography. Results showed that 16% of the perfume reacted with trimesoyl chloride and 31% of it with diethylenetriamine. To eliminate the loss of perfume the organic monomer was dissolved in the perfume in a water bath (at around 2°C) and when the aqueous monomer was added to the system it reacted first with the organic monomer and it may not be able to pass through the membrane to react with the perfume after the capsule wall was formed, as the high values of encapsulation efficiency shown before indicates.

The effects of the different formulations and reaction conditions on the **leakage** of perfume from the microcapsules to a water-hexane system have also been discussed. The partition coefficient of the perfume in a water-hexane system was measured firstly. Leakage results showed that the *temperature of reaction* has not an effect on the perfume release from the microcapsules. The *type of monomer* used had a much higher influence on the leakage of perfume from the formed microcapsules. It has been found that the use of Trim instead of TC as organic monomer and of HMDA instead of DETA, EDA or TETA as aqueous monomer reduces greatly the perfume release. Some experiments were done using both organic monomers and adding different aqueous monomers at different reaction times and it was possible to enhance the wall impermeability. The formulation with the lowest leakage (called All50) was formed by a combination of Trim and TC and adding first DETA, 15min later HMDA and 1h later EDA (see Figure 5.6 for leakage data and Table 4.1 for formulation details). It has also been found that the presence of a *thickener* (paraffin oil) that doubled the viscosity of the encapsulated perfume phase did not have a significant influence on the leakage rate.

Apart from the encapsulation efficiency and release data the other important parameter for the microcapsules from an industrial point of view (for performance in final products) is their **mechanical strength**, which has been measured with a unique rig apparatus (see Figure 4.6). Single microcapsules were compressed until rupture and the forces required to break them measured. The influences of the different formulations and reaction conditions on the mechanical strength of the microcapsules have been discussed. As pointed before the *temperature of reaction* was crucial to the mechanical strength of the formed microcapsules. It has been found that the lower the temperature of reaction the stronger the microcapsules

formed, and the microcapsules made at 18°C were very weak and collapsed when the continuous phase evaporated. The *organic monomer* used in the formulation had a huge influence on the possibility of capsule formation. As discussed before no capsules were formed with the aliphatic monomer, only with the aromatic ones, and only when trimesoyl chloride was at least 70% of the total amount of organic monomer capsules were stable over time during storage. It was found that when some terephthaloyl chloride was added to the trimesoyl chloride (keeping the weight ratios 90/10 and 82/18) the mechanical strength of the capsules was slightly increased but when more TC (ratio 73/23) was added the capsules produced became weaker. Results show that the *aqueous monomer* used in the microencapsulation process had a higher influence on the mechanical strength of the particles. It has been found that microcapsules produced with HMDA were 3 times stronger than when DETA was used and when monomers were added at different times (formulation All50) the resulting strength was comparable to the use of HMDA alone.

The **viscoelastic and plastic properties** of the microcapsules have been studied. “Compress and hold” and “compress and release” experiments have been done to the microcapsules and results show that the polyamide formed presented elastic behaviour at very low deformations (3%), viscoelastic behaviour at low deformations (below 10%) and plastic behaviour at higher deformations.

Perfume has been successfully encapsulated using the interfacial polymerisation technique and the effects of different parameters on the final properties of the capsules have been studied. The best formulation has been selected and analysis data (d_{32}:24.7μm, wall thickness: 301 ± 18nm, loading: 92.9 ± 0.4%, total encapsulation efficiency: 99.7 ± 0.1%, useful

encapsulation efficiency: 77.9±0.1%, leakage at 3h: 2.7±0.6%, stress at rupture: 3.2± 0.6MPa) suggested that microcapsules may find industrial applications.

7.1.2. Glycerol microcapsules

Glycerol is a viscous water soluble liquid. Due to its hygroscopic nature it is highly appreciated in cosmetics, like lipsticks, although it is currently not too widely used because of its instability in final products. The technique developed to encapsulate perfume can be modified to encapsulate also water soluble actives and glycerol has been chosen as a model active. Glycerol microcapsules will permit to stabilize glycerol in final products and release it slowly when it is required. Production of glycerol is highly increasing over recent years and it is expected to continue growing in the future as it is a byproduct from other industries (detergents and biodiesel mainly) which enables supply of cheap glycerol for new applications.

The encapsulation process comprises the **formation of an emulsion** of the dispersed phase (glycerol/water) in a continuous organic phase. Different organic phases have been tried (paraffin oil, mineral oil and IPM) and only IPM provided the low viscosity needed to form an emulsion. A glycerol/IPM emulsion was formed but it was not stable and glycerol tended to coalesce fast, which made it impossible to obtain small microcapsules, the capsules obtained varied between 150 and 1500μm depending on the operating conditions, whilst the objective was to obtain capsules of less than 50μm. It has been found that the addition of magnesium sulphate stabilized the emulsion and glycerol microcapsules of a regular size of less than 10μm were obtained.

Once the problem of emulsion stabilization was solved 5 batches using different aqueous monomer compositions and process conditions (temperature and stirring rate) were made. The **size distribution** of the microcapsules formed was measured and it was observed that the stirring rate used was not as important as the salt addition to produce small microcapsules. If salt was added to the system there was almost no difference in the size distribution obtained when stirring rates of 1,500 and 3,000rpm were used.

Most of the microcapsules formed were very weak and it was not possible to measure their **mechanical properties** as they broke when the liquid around them was removed, *i.e.* after drying. Only 4 microcapsules from one sample were compressed and it was found that they broke at low displacements (5 to 17%), and they were fragile.

The **stability** of the microcapsules over time has also been studied. It has been found that when HMDA was used alone in the formulation the slurry formed completely gelled after a few weeks making it impossible to re-suspend the microcapsules anymore. When DETA was present in the system liquid was always evident in the slurry. This may suggest that HMDA microcapsules were weak and permeable and that they released the glycerol to the bulk where it reacted with the excess of trimesoyl chloride present there forming a gel. In the case of DETA the microcapsules formed were more resistant to leakage and glycerol was not released.

In all the cases studied an interesting behaviour was found: if the organic phase was removed and replaced with water the glycerol was not dissolved in the water, it formed a dispersion of

small glycerol droplets in water, probably stabilized by the salt. If the water was evaporated under the microscope the small droplets joined together and formed bigger ones.

7.2. Recommendations for future work

7.2.1. Perfume encapsulation

In this work perfume has been successfully encapsulated with polyamide using an interfacial polymerisation technique. The microcapsules produced have been characterized and the effects of different parameters on their final properties have been discussed. But it is possible to go further and study different compositions and reaction conditions:

- It is suggested that other amines and acid chlorides be used, like poly(oxypropylene)diamine (Soto-Portas, ML *et al.*, 2003), silane coupling agent (Mathiowitz and Cohen, 1989a), or many other listed in the literature (Argillier *et al.*, 2002). Silane coupling agent was found to reduce the leakage from polyamide microcapsules for example.
- It is also possible to add the aqueous monomers at different times from the ones studied here.
- In our system the minimum reaction temperature was determined by the freezing temperature of the water phase (around -1.5°C, depending on the surfactant concentration), it would be possible to modify the system to be able to work at lower temperatures (adding an alcohol or glycol, different emulsifiers or salts, for example).

The main problem found during this work was the separation and purification of the microcapsules formed. Filtering was tried but it was not possible to resuspend again the solids recovered. This means that the microcapsules have been stored in the reaction media which

present an excess of amines. It would be good to develop a method to remove the amines from the system and wash the microcapsules formed.

It would also be interesting to use cryo-SEM to see the aspect of a section of the membrane wall and measure its thickness since the preparation of the samples for TEM is aggressive, and could have a side effect on the results obtained, for example during the preparation of the weaker samples (the ones with TETA) it was found that the capsules did not survive the process and they were broken.

It would be needed to do more experiments on microcapsule compression (loading-holding and loading-unloading) at low deformations to calculate more precisely the elastic, plastic and viscoelastic property parameters of the microcapsule walls made of the polymer.

It is highly recommended to study the use of polyureas or polyurethanes (Su *et al.*, 2006) as polymeric walls. In theory they should create more impermeable walls although they use isocyanates in their formulation, which are toxic substances.

7.2.2. Glycerol encapsulation

Glycerol has been successfully encapsulated, but there was no time for optimizing the formulation and process conditions.

It has been found that microcapsules formed were very weak therefore it is recommended to use a higher monomer concentration and lower reaction temperature. The increase of the monomer concentration may cause some amine monomer to remain unreacted inside the

capsule. This presence of unreacted amine should be measured. The reduction of the temperature of reaction will increase the viscosity of the system which may result in bigger microcapsules for the same formulations and emulsification conditions, even if the microcapsules formed in this work were very small and were stabilized by the presence of salt in the system. There is still a scope to change processing conditions to form microcapsules of the desired size. The use of different salts to stabilize the emulsion can be further studied.

The technique attempted in this study to measure glycerol concentration has not provided satisfactory results. It seems that some of the other products in the formulation interfered with the method and gave a much higher reading than the expected one. It will be required to develop a new method to measure glycerol concentration, which permits to determine the encapsulation efficiency and the leakage of glycerol from the microcapsules precisely.

Once strong glycerol microcapsules are developed they would be mechanically characterized and the rupture force, nominal stress at rupture and displacement at rupture would be measured. The intrinsic properties of the wall polymer (elastic, plastic and viscoelastic property parameters) should also be determined.

Like those for perfume capsules, other monomers can be studied to make polyamide microcapsules and other polymers can be formed (polyurea or polyurethane).

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