SUPERCRITICAL CARBON DIOXIDE FRACTIONATION OF PALM KERNEL OIL AND FORMULATION OF COCOA BUTTER REPLACERS FAT

By

MD. ZAIDUL ISLAM SARKER

Thesis submitted in fulfillment of the requirements for the degree of

Doctor of Philosophy

July 2003

Dedicated To

My revered parents

Alhajj Mohd. Ezahar Ali Sarker & Mrs. Zobeida Begom

And my parents-in-law

The Late Nazir Hossain & The Late Amina Begom

ACKNOWLEDGEMENTS

I am especially grateful to my supervisor, Professor Ir. Dr. Mohd. Omar bin Ab. Kadir of the School of Industrial Technology, and my co-supervisor, Associate Professor Dr. Nik Norulaini Nik Abd. Rahman of the School of Distance Education, Universiti Sains Malaysia (USM) for their enthusiasm and invaluable guidance, which has brought this project into a reality. I would also like to thank them for appointing me as a research fellow under IRPA grant No: PTEKIND/305/610636.

My thanks are also due to the vice chancellor of USM for providing my tuition fees under the Federation of Universities of the Islamic World (FUIW) fellowship scheme and also to the Dean of the School of Industrial Technology, USM, Associate Professor Dr. Wan Rosli Wan Daud, to the deputy deans, Associate Professor Dr. Abd. Karim Alias and Associate Professor Dr. Rozman, to other lecturers especially to Dr. Azlan Ariffin and staff of School of Industrial Technology, USM, for their direct or indirect help in making this project successful.

I would like to express my gratitude and thanks to Associate Prof. Dr. Ataharul Islam of the School of Mathematics, USM for his cordial help in statistically analysing the experimental data of this thesis.

Cooperation from laboratory assistants, from my colleagues especially from Anuar Omar, and from my friend Md. Fazlul Bari, PhD student from the School of Chemical Sciences, is also gratefully appreciated.

I would like to thank all Bangladeshi and foreign students for their encouragement within the small international community in USM, especially to Md. Mahmudul Hassan (PhD student of the Center for Policy Research, USM) and Mrs. Salma Mahmud (Master Student of the same center, USM) for their suggestions and moral support.

I would like to gratefully thank Malpom Sdn. Bhd. (Nibong Tabat, Penang), Wilmer Edible Oil Company (Butterworth, Penang) and Acidchem International (Seberang Prai, Penang) for providing samples for this research.

The special gratitude and thanks go to my all teachers, especially Mohd. Abd. Latif Prodhan and Dr. Mohd Shahjahan (Dept. of Biochemistry, Rajshahi University, Bangladesh) and to villagers, relatives and friends left back at home for their encouragement and support. I thank my elder brother Dr. Azim Uddin Ahmed who is waiting to call me Dr., and also my younger brother Md. Shahidul Islam Sarker and my sisters especially Mrs. Jahanara Begom for their encouragement.

Finally, I gratefully acknowledge the moral support and perseverance of my dear wife and my best friend Mrs. Nazmin Begom who took special care of my physical and spiritual comfort throughout the study. Her continuous support and enthusiasm for my career is most invaluable.

TABLE OF CONTENTS

CHAPTER 4

LIST OF TABLES

LIST OF FIGURES

Captions | Page no.

LIST OF ABBREVIATIONS

ANOVA	=	Analysis of variance
AR	=	Analytical reagent
CP	=	Critical point
CB	=	Cocoa butter
CBR	=	Cocoa butter replcers
CBE	=	Cocoa butter equivalent
CBS	=	Cocoa butter substitute
CBe	=	Cocoa butter extender
CO_2	=	Carbon dioxide
C8	=	Caprylic acid
C10	=	Capric acid
C12	=	Lauric acid
C14	=	Myristic acid
C16	=	Palmitic acid
C18	=	Stearic acid
C18:1	=	Oleic acid
C18:2	=	Linoleic acid
FFA	=	Free fatty acid
FID	=	Flame ionization detector
FAME	=	Fatty acid methyl ester
GC	=	Gas chromatography
GLC	=	Gas liquid chromatography

GC-MS	=	Gas chromatography mass spectroscopy
IV	=	Iodine value
MPa	=	Mega Pascal
NMR	=	Nuclear magnetic resonance
PO	=	Palm oil
PKO	=	Palm kernel oil
PKO-SC1	=	Palm kernel oil-first fraction
PKO-SC2	=	Palm kernel oil-second fraction
PKO-SC3	=	Palm kernel oil-third fraction
PKO-SC4	=	Palm kernel oil-fourth fraction
Palmac 760	=	Oleic acid
Palmac 98-18	=	Stearic acid
PORIM	=	Palm oil research institute Malaysia
ppm	=	parts per million calculated as w/w
Psi	=	Pound per square inch
Pc	=	Critical pressure
SFs	=	Supercritical fluids
SFE	=	Supercritical fluid extraction
SEM	=	Scanning electron micrograph
SC-CO_2	=	Supercritical carbon dioxide
SMP	=	Slip melting point
SFC	=	Solid fat content
Tc	=	Critical temperature

ABSTRACT

The extraction of dehulled ground palm kernel using supercritical carbon dioxide (SC-CO_2) as a solvent at temperatures of 40 and 80°C and pressures of 20.7, 27.6, 34.5, 41.4 and 48.3 MPa was studied. Continuous extraction was fractionated into four fractions and each fraction was collected for every 10 minutes. Thus the total extraction process was performed for 40 minutes extraction time. Solubility of palm kernel oil (PKO) increased and total use of carbon dioxide (CO_2) decreased with increase in temperature from 40 to 80°C and pressures at 34.5, 41.4 and 48.3 MPa. At lower pressures of 20.7 and 27.6 MPa solubility of oil decreased and use of CO_2 increased with increase in temperature (80°C) and this was due to the retrogradation characteristic of supercritical fluid at lower pressure. The highest yield was obtained at 48.3 MPa and 80°C with minimum CO_2 used. For fractionations the first fraction gave the highest yield and then gradually decreased till to the last fraction for both temperatures 40 and 80°C. That was due to the low solubility of PKO in the later fraction. This trend had been observed for all the pressures except 20.7 and 27.6 MPa. At 20.7 and 27.6 MPa the yield was found to be almost the same for all fractions. This is due to lower amount of oils were extracted at 20.7 and 27.6 MPa. The three-way analysis of variance (ANOVA) on the yield as a function of pressure, temperature and fractionation showed that the yields were found to be significantly different at different pressures and temperatures indicating a significant difference between the population means (p value ≤ 0.01). Also the yield at different fractions was significantly different ($p \leq 0.01$).

The different fatty acid compositions caprylic (C8), capric (C10), lauric (C12), myristic (C14), palmitic (C16), stearic (C18:0), oleic (C18:1) and linoleic (C18:2) acid in the SC-

CO_2 fractionated PKO were found to be different at various pressures and temperatures. More C8, C10, C12 extracted in the first fractions, which gradually declined as extraction period extracted into second, third and fourth fractions. C14 remained unvaried over the extraction period showing no remarkable difference among the fractions but at 48.3 MPa and 80°C it reduced remarkably in the last fraction. On the other hand more C16, C18 and unsaturated C18:1 and C18:2 were present in the latter fractions. This trends were found to increase with increase in pressures and temperatures but the difference were not remarkable at lower pressure 20.7 and 27.6 MPa for both temperatures. However, the compositions were found to be statistically significant for pressure and fraction times by type of fatty acid.

Blending of palm oil (PO), 4th fraction of palm kernel oil extracted using SC-CO_2 at 48.3 MPa and 80°C (PKO-SC4), palmac 98-18 (C18 based) and palmac 760 (C18:1 based) at different ratios were carried out to obtain blends of cocoa butter replacers (CBR). There were 13 blends performed through out the study. Blends 1 to 10 were recommended as cocoa butter substitutes (CBS) and blends 11 to 13 were referred as cocoa butter equivalent (CBE) fats. The slip melting point (SMP) for blends 11, 12 and 13 were found to be 33.3, 35.1 and 35.3°C, respectively, whereas for the CB it is 35°C. Also the solid fat content (SFC) of these CBE were found to be higher at 20°C and 0 percent within 37.5°C. This trend is quite similar to commercial cocoa butter (CB). The iodine value, saponification value and acid value of these CBE were found quite similar to CB. Thus it proposed to substitute CB at maximum level of substitution. However, the physico-chemical properties of blends 1 to 10 were not found to be as close as CB but it could be proposed as CBS for low level of substitution of CB.

ABSTRAK

Pemeringkatan Lampau Genting Karbon Dioksida Minyak Isirung Sawit Dan Formulasi Lemak Pengganti Mentega Koko

Kajian ke atas pengekstrakan isirung sawit tanpa testa hancur menggunakan lampau genting karbon dioksida (LG-CO_2) sebagai pelarut pada suhu 40 dan 80°C dan tekanan 20.7, 26.7, 34.5, 41.4 dan 48.3 MPa. Pengekstrakan berterusan dipemeringkatkan kepada empat pemeringkat dan setiap pemeringkat dikumpul selama 10 minit dengan jumlah proses pengekstrakan dibuat selama 40 minit masa pengekstrakan. Kelarutan minyak isirung sawit (PKO) meningkat dan jumlah karbon dioksida (CO_2) yang digunakan menurun dengan meningkatnya suhu dari 40 ke 80°C dan tekanan pada 34.5, 41.4 dan 48.3 MPa. Pada tekanan rendah 20.7 dan 27.6 MPa kelarutan minyak menurun dan penggunaan CO_2 meningkat dengan meningkatnya suhu (80°C) dan ini disebabkan oleh ciri kesan undur (retrogradation) bendalir lampau genting pada tekanan rendah. Penghasilan minyak tertinggi diperolehi pada 48.3 MPa dan 80°C dengan penggunaan CO_2 yang minima.

Pemeringkatan untuk pemeringkat pertama memberikan hasil yang tertinggi dan kemudian perlahan-lahan menurun pada pemeringkat yang terakhir untuk kedua-dua suhu 40 dan 80°C. Ini disebabkan oleh kelarutan PKO yang rendah dalam pemeringkat yang terakhir. Tren ini diperhatikan pada kesemua tekanan kecuali tekanan 20.7 dan 27.6 MPa. Didapati pada 20.7 dan 27.6 MPa hasil yang diperolehi adalah sama pada kesemua pemeringkat, ini disebabkan oleh kurangnya amaun minyak yang diekstrak pada 20.7 dan 27.6 MPa.

Analisis varian 'three-way' (ANOVA) ke atas hasilan sebagai fungsi tekanan, suhu dan pemeringkatan menunjukkan bahawa hasilan berbeza seca signifikan pada tekanan berbeza dengan suhu di antara min populasi (p value $\leq$ 0.01). Begitu juga hasilan pada pemeringkat berbeza berbeza secara signifikan (p value $\leq$ 0.01).

Didapati komposisi asid lemak kaprilik (C8), kaprik (C10), laurik (C12), miristik (C14), palmitik (C16), stearik (C18:0), oliek (C18:1) dan linoliek (C18:2) berbeza di dalam pemeringkatan PKO LG-CO_2 pada tekanan dan suhu berbeza. Asid lemak C8, C10 dan C12 banyak diekstrak pada pemeringkat pertama dan semakin perlahan-lahan berkurangan pada pemeringkat kedua, ketiga dan keempat. Komposisi asid lemak C14 kekal tidak berubah pada kesemua pemeringkat sepanjang tempoh pengekstrakan tetapi pada tekanan 48.3 MPa dan suhu 80°C ianya berkurangan dengan banyak pada pemeringkat terakhir. Sebaliknya, terdapat peningkatan C16, C18:0 dan C18:1 dan C18:2 tidak tepu pada pemeringkat terakhir. Tren ini meningkat dengan meningkatnya tekanan dan suhu tetapi perbezaannya tidak begitu ketara pada tekanan rendah 20.7 dan 27.6 MPa pada kedua-dua suhu. Walau bagaimana pun, didapati komposisi signifikan secara statistik untuk tekanan dan masa pemeringkat mengikut jenis asid lemak.

Adunan minyak sawit (PO), PKO-SC4, palmac 98-18 (asas C18) dan palmac (asas C18:1) pada nisbah berbeza dibuat untuk memperolehi adunan pengubah mentega koko (CBR). Terdapat 13 adunan yang dibuat untuk kajian ini. Adunan 1 hingga 10 dicadangkan sebagai gantian mentega koko (CBS) and adunan 11 hingga 13 dirujuk sebagai lemak setara mentega koko (CBE). Takat cair lolos untuk adunan 11, 12 dan 13 ialah masing-masing

33.3, 35.1 dan 35.3°C manakala untuk mentega koko (CB) ialah 35°C. Begitu juga kandungan lemak pepejal (SFC) gantian mentega koko ini tinggi pada 20°C dan 0 peratus diantara 37.5°C dan tren ini adalah hampir menyamai mentega koko komersial. Nilai iodin, nilai saponofikasi dan nilai asid gantian mentega koko ini juga hampir menyamai mentega koko. Oleh itu ianya dicadangkan untuk gantian mentega koko pada paras maksima penggantian. Walau bagaimana pun, cirri fisiko-kimia adunan 1 hingga 10 didapati tidak menghampiri mentega koko tetapi ianya dicadangkan sebagai gantian mentega koko pada paras terrendah penggantian mentega koko.

CHAPTER 1

INTRODUCTION

1.1 Preamble

Supercritical fluid extraction (SFE) is a new and powerful technique in separation processes and an alternative processing method. Carbon dioxide (CO_2) is employed as a supercritical fluid because it has a low critical temperature (31.1°C) and critical pressure (7.28 MPa), which makes it an ideal solvent for extracting thermally sensitive materials. CO_2 is also nontoxic, nonflammable, and easily available and relatively low cost. In Malaysia the laboratory grade CO_2 is costs about US$ 0.66/ liter (dm^3). In addition, products obtained by SC-CO_2 extraction are completely free of solvent residues. On the contrary, conventionally solvent-extracted products must be desolventized before they are suitable for consumption. SC-CO_2 defatted meal can therefore be directly used in low-calorie foods. Also, crude oils obtained by SC-CO_2 extraction are generally more easily refined than conventionally extracted oils as they contain fewer impurities (Devittori et al., 2000, Bruno & Ely, 1991 & de Castro et al., 1994). The low critical temperature (31°C), non-toxicity and low cost have long rendered SC-CO_2 a suitable solvent for food products (McHugh & Krukonis, 1994 & Saldana et al., 1999). There has been renewed interest in the use and development of supercritical fluid extraction (SFE) methods for industrial applications. Several investigations have been made in recent years on probable industrial applications of the SFE, which offer some preferences over the conventional methods, such as separation by extractive solvents by distillation, especially in the areas of the food, pharmaceutical, chemical and oil industries (Bruno & Ely, 1991).

Several researchers have studied SC-CO_2 extraction of seed oil from a wide range of seed species such as corn germ (List et al., 1984a), cottonseed (List et al., 1984b), canola seed (Fattori et al., 1988), evening primrose (Lee et al., 1999), dehulled and undehulled ground palm kernels (Hassan et al., 1999 & Hassan et al., 2001) and palm kernel meal (Anuar, 2002). Palm kernel is a by-product of palm oil industry. It constitutes about 45 percent of palm nut of palm oil *Elaeis guineensis*. On a wet basis, the kernel contains about 45-50 percent of oil. Although it lies within the palm nut, palm kernel oil and palm oil differ greatly in their characteristics and properties. Palm kernel oil is rich in lauric acid, C12 (48.3%) and other major fatty acids are myristic, C14 (15.6%) and oleic acids, C18:1 (15.1%) (Rossell et al., 1985, Goh, 1993 & Omar et al., 1998). This gives the fats a solid consistency at cool ambient temperatures, but they nevertheless melt below 30°C (Rossell, 1985). Although SC-CO_2 extraction of lipids has been extensively studied in the laboratory very few studies were reported on solubility of palm kernel oil (PKO) in SC-CO_2. Hassan (2000), Hassan et. al. (2000) and Anuar (2002) measured the solubility of PKO in SC-CO_2 within the pressure range of 27.6-48.3 MPa and the temperature range of 40-80°C. It is expected that the solubility of palm kernel oil will increase significantly higher at 27.6 MPa. In this study it is envisaged that palm kernel oil solubility of 4% could be achieved at a pressure of 34.5 MPa, which is lower than the pressure required in the extraction of non-lauric oils such as soyabean oil, and cottonseeds oil as conducted by Friedrich (U.S. Pat. No. 4,466,923) where a pressure of at least 55.2 MPa is required.

Palm kernel oil (PKO) is regarded as high quality oil for food use. It is a valuable component of margarine formulation, giving rapid melting character in the mouth. Its high solid content at 15-20°C, together with rapid melting point, makes it particularly useful in confectionary products. It is commercially fractionated into liquid olein and solid stearin, the latter being a premium product. Fractionation increases the lauric (C12) and myristic (C14) acid concentration in the resulting stearins, and leads to a corresponding fall in the levels of short-chain (C6-C10) and unsaturated (C18:1 and C18:2) fatty acids (Hassan et al., 2000). But there are no others alternative convenient and environmental friendly process available to reduce the short chain (C8 and C10) and medium chain (C12 and C14) and increasing the long chain (C18:0, C18:1 and C18:2) fatty acids in PKO which can be used in a large amount as cocoa butter replacers (Nik Norulaini et al., 2002a & 2003a) or confectionary fat as it is an expensive fat. Moreover, composition of the triglycerides with short and medium chain fatty acids (C8-C12) decreases with fraction of oil collected, while long chain triglycerides (C16-C18:2) increases (Hassan, 2000). The amount of the total extracted PKO using SC-CO_2 at 34.5-48.3 MPa and 40-80^{o}C is higher than the total oil extracted through conventional method (screw press) and the process removes free fatty acids (FFA) and deodorizes the oil (Hassan et al., 2000), and better quality palm kernel meal can be achieved as a by-product of PKO which can be used as poultry diet due to the reduced fiber content of the kernels using SC-CO_2 (Anuar, 2002).

Blending of single or straight chain oils and fats usually is unable to fulfill the complex technical specifications prescribed for a particular product application. O'Brien (1998) mentioned that vegetable oils are blended to meet both the composition and analytical consistency controls identified by the product developers and quality assurance. The consistency controls frequently include analytical evaluations for solids fat index, iodine value, melting points, fatty acid composition, and so on.

All the fats, which replace cocoa butter either partially or wholly in chocolate products, are generally known as cocoa butter replacer (CBR). It should also be cheaper than cocoa butter (CB) and it must serve the purpose of CB. Likewise, it would be able to be processed and it has to meet the legal requirements (some countries forbid the use of animal or synthetic fats in chocolate products). It should be based on easily available raw materials (Kheiri, 1982). CBR are further classified into the following three groups namely; cocoa butter equivalent (CBE), cocoa butter substitute (CBS) and cocoa butter extender. Cocoa butter equivalents (CBE) are fats, which behave like cocoa butter in all respects and are able to mix with cocoa butter in any proportion without altering the melting, rheological and processing characteristics similar to cocoa butter. There are no 100 percent CBE available in the market. It is designed to contain glyceride composition similar to that of cocoa butter. Their properties therefore are expected to be similar and are compatible with cocoa butter in mixtures for chocolate manufacture. Cocoa butter substitutes (CBS) are fats, which can be mixed with cocoa butter to a limited extent, without significantly altering its melting, rheological and processing properties. They do not necessarily have physico-chemical characteristics similar to cocoa butter. The amount of the CBS used depends on its degree of compatibility with cocoa butter and/or cocoa butter and vegetable fat blends. This degree of compatibility determines the quality and hence the price of the extender. Vegetable fats are used as extenders in those countries where partial replacement of cocoa butter is permitted. A good quality CBS is hard at ambient temperature, has sharp melting characteristics like cocoa butter and has a high degree of compatibility with cocoa butter and/or cocoa butter- milk fat blends. CBS are mainly used to make imitation products where the fat phase. CBS are fats, usually based on lauric acid, i.e. either originating from PKO or coconut oil. They have snap and melting properties similar to cocoa butter but differ in chemical composition. They are therefore not compatible with cocoa butter. Cocoa butter extender are fats which can not tolerate cocoa butter except in a very limited amount as mixing of these with cocoa butter adversely affects the rheological, melting and processing characteristics of the product.

1.2 Objectives of Research

- To study the extractability and solubility of palm kernel oil (PKO) and its components from palm kernel using supercritical carbon dioxide (SC-CO_2) at various temperatures and pressures.

- To fractionate the fatty acid components based on solubility at various pressures and temperatures and to identify the fraction, which has both the lowest lauric acid and the highest oleic acid content.

- To formulate blending ratios of the extracted fraction of PKO (with lowest lauric and highest oleci acid content) with palm oil and fatty acid supplements (stearic and oleic acids) to achieve the properties of some confectionary fats having cocoa butter quality.

- To analyze the physico-chemical properties i.e. fatty acid profiles, slip melting point, solid fat content, iodine value, acid value and saponification value etc of the components before and after blending compromising the cocoa butter quality fats.

CHAPTER 2

LITERATURE REVIEW

2.1 Historical Background of Supercritical Fluid Extraction

2.1.1 Discovery of supercritical fluid extraction

Buchner (1906) carried out studies over a wide range of temperatures and determined the solubilities of several solutes in supercritical fluid. He was the first to investigate the solubility of naphthalene in supercritical carbon dioxide and ethylene. Due to considerable experimental problems associated with working in the supercritical fluid region, further investigations on naphthalene solubility in supercritical fluid were hindered until Diepen & Scheffer (1948) began their extensive publications on the phase behaviour of naphthalene-supercritical ethylene system in late 1940s. For example, Diepen & Scheffer (1948) published the solubility and phase behaviour data of naphthalene in supercritical ethylene.

Phase behaviour and solubility of naphthalene in various other supercritical fluids were studied extensively in the early 1960s (McHugh & Krukonis, 1986). In 1954, Francis (1954) reported an extensive study on the phase behaviour of ternary systems containing liquid CO_2 and determined the solubility of 261 compounds in near-critical liquid carbon dioxide in a single paper. It is possible to formulate general rules on the solubility behaviour of compounds in supercritical carbon dioxide (SC-CO_2) using his data that is also applicable to the supercritical region.

In the last 30 years, there has been a tremendous interest in the use of supercritical fluid as solvents for extraction or separation purposes. Intensive study on extraction of food components using supercritical fluid began in the early 1970s. Many patents resulted from these studies such as for the extraction of hops, decaffeination of coffee and tea, tobacco and spices.

The first symposium on supercritical fluid (Stahl et al., 1988) was held in Essen, which was the result of rapid development of the SFE and separation methods. Stahl et al. (1988) initiated this symposium under the theme 'Extraction with Supercritical Gases' and since then supercritical fluid has been receiving increasing interest as a solvent for extraction of solids and liquids.

2.1.2 Development of supercritical fluid solvent technology

A supercritical fluid is a fluid that exists above its critical temperature and pressure. Figure 2.1.2.1 shows the schematic pressure-temperature phase diagram for a pure component (Rizvi et al., 1986). The supercritical region is denoted by the crosshatched area in the diagram. The region is above the critical temperature and pressure of the component. In this region, there is no phase change and there is continuous transition from liquid to supercritical fluid by increasing the temperature at constant pressure or from gas to supercritical fluid by increasing the pressure at constant temperature.

In the past decade or so, numerous industrial and academic research and development laboratories have investigated the underlying principles and process applications of supercritical fluids as solvents. This interest was due to the possibility of separation of multi-component mixtures using supercritical fluid solvents.

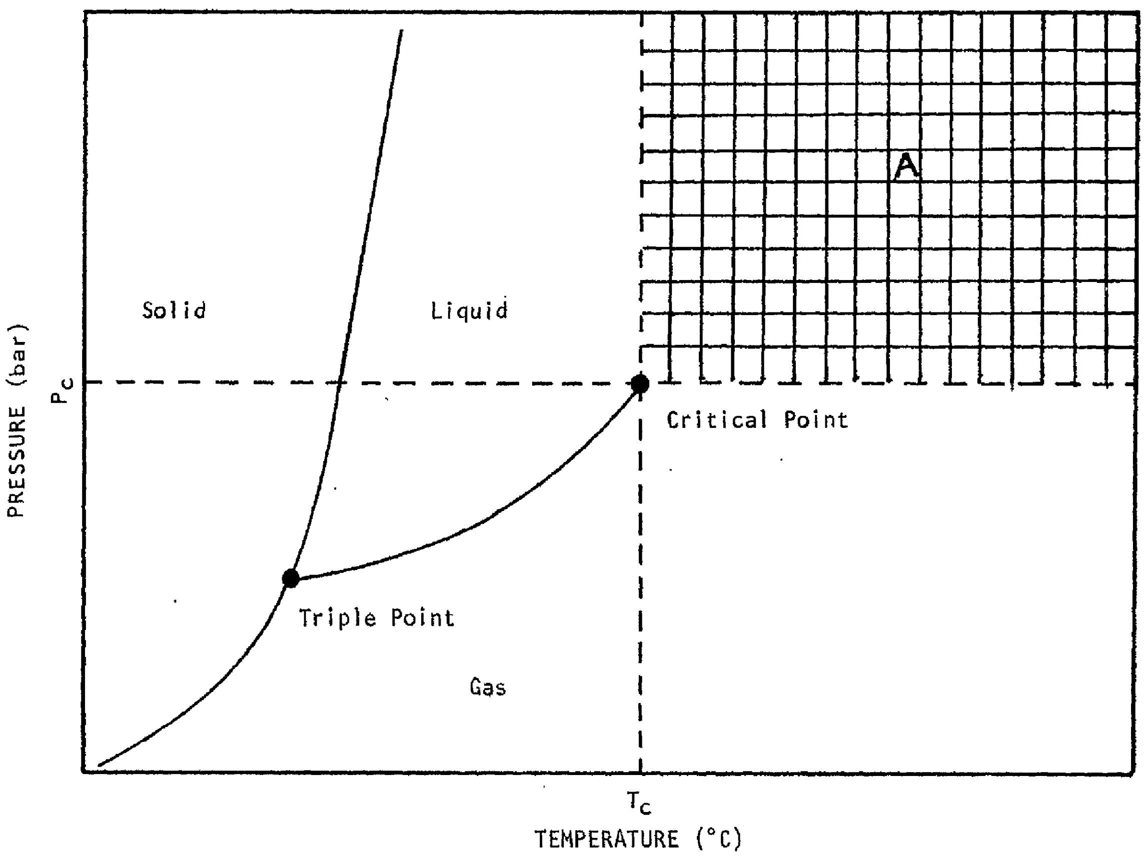

Figure 2.1.2.1 Typical Pressure-Temperature phase diagram for a pure compound showing the supercritical region, A (Rizvi et al., 1986)

Many gases were observed to exhibit enhanced solvent power when compressed to conditions above their critical parameters. The reasons for the motivation for development of supercritical fluids as solvents for viable extraction and separation are given below (McHugh & Krukonis, 1986):

(a) Cost of energy has increased sharply over the years and resulted in increased cost of traditional, energy -intensive separation techniques such as distillation.

(b) Increased awareness on environmental safety, which led to, increased governmental scrutiny and regulation of common industrial solvents such as the halogenated hydrocarbons, which made non-toxic, environmentally acceptable supercritical fluid solvents such as CO_2 very attractive as alternative industrial solvents.

(c) Pollution-control legislation has become more stringent which led to industries having to consider alternative means of waste treatments.

(d) Joint Association for the Advancement of Supercritical Fluid Technology (JAAST) was formed in the United States to develop and disseminate knowledge regarding the application of supercritical fluids for cleaning purposes in 1990 (Taylor, 1996).

(e) Increased demand for better performance materials, which traditional/conventional separation or extraction techniques cannot meet.

However, in truth, supercritical fluid technology has become an interdisciplinary field, utilized by chemical engineers, chemists, food scientists, materials scientists, agronomists, and researchers in biotechnology and environmental control. The last ten years have seen the emphasis in supercritical fluid expand from commodity chemicals and synthetic fuels toward more complex, highly specialized, and more valuable substances. Today supercritical fluids are being touted for sample preparation prior to trace analysis and for mobile phases in analytical and preparative scale supercritical fluid chromatography. In summary, the extensive use of supercritical fluid in both science and engineering laboratories in the near future is certain.

2.2 Properties of Supercritical Fluids

2.2.1 Density consideration with pressure and temperature

A phase diagram, as shown in Figure 2.2.1.1, described the physical stage of a substance of fixed composition. In this pressure-temperature (PT) diagram for CO_2 there are three lines describing the sublimation, melting, and boiling processes. These lines also define the regions corresponding to the gas, liquid, and solid states. Points along the lines (between the phases) define the equilibrium between two of the phases. The vapor pressure (boiling) starts at the triple point (TP) and ends at the critical point (CP). The critical region has its origin at the critical point. At this point we can define a supercritical fluid as any substance that is above its critical temperature (T_c) and critical pressure (P_c). The critical temperature is therefore, the highest temperature at which a gas can be converted to a liquid by an increase in pressure. The critical pressure is the highest pressure at which a liquid can be converted to a traditional gas by an increase in the liquid temperature. In the so-called critical region, there is only one phase and it possesses some of the properties of both a gas and liquid. Subcritical (liquid) CO_2 is found in the triangular region formed by the melting curve, the boiling curve and the line that defines the critical pressure (Brogle, 1982). Supercritical and liquid CO_2 can both be used as solvents. In contrast to sub critical CO_2 (i.e., liquid), the solvating power of the supercritical fluid is highly dependent on its temperature and pressure. Figure 2.2.1.2 illustrates that at low pressure solvent power of CO_2 surprisingly decreases with rising temperature; whereas at high pressures it increases in a straightforward fashion as measured by naphthalene solubility Figure 2.2.1.3. If the parameter "pressure" is replaced by the parameter "density," the solubility-temperature relationship becomes much simpler, as shown in Figure 2.2.1.4. This anomaly comes about

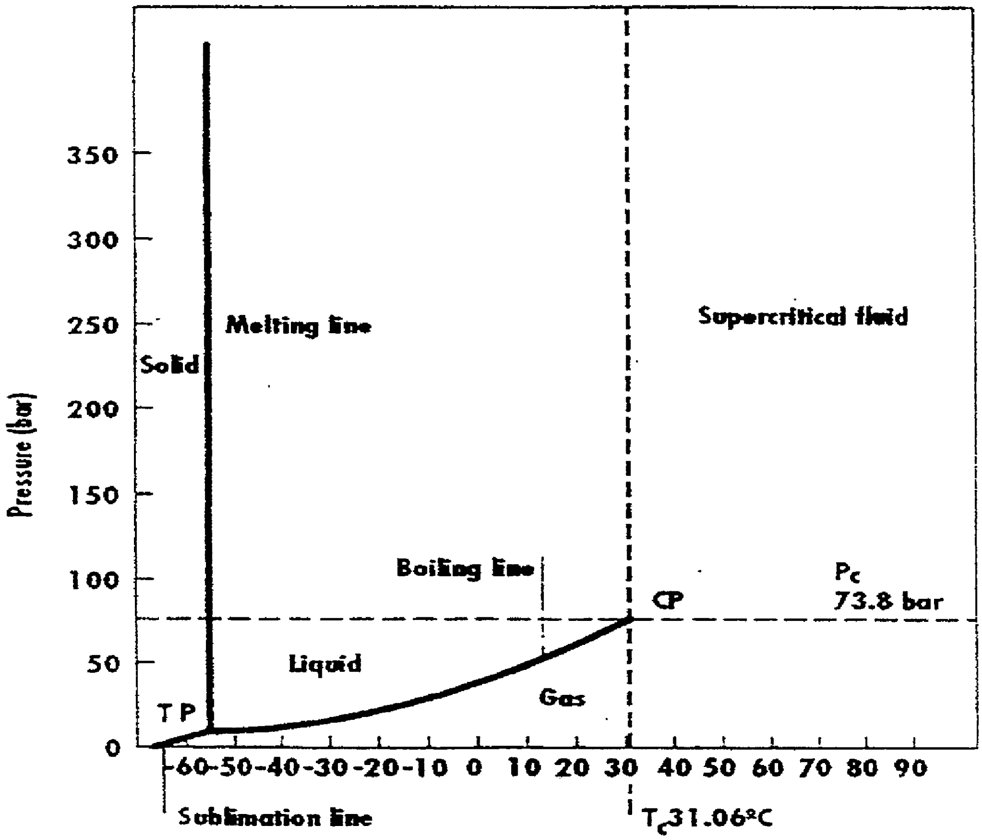

Figure 2.2.1.1 Phase (pressure-temperature) diagram for CO_2: CP=critical point, TP=triple point, P_c= critical pressure, T_c= critical temperature (Brogle, 1982)

because density decreases dramatically with an increase in temperature at low pressure; whereas at higher pressure, changes in temperature have much less effect on density. Thus density, not pressure, to a first approximation is proportional to the solvent power of the supercritical fluid. Figure 2.2.1.5 shows the density of CO_2 as a function of pressure at different temperatures (Fattori et al., 1988). This figure indicates that an increase in temperature results in a decrease in the density of carbon dioxide at a constant pressure. On parameter "pressure" is replaced by the parameter "density," the solubility-temperature relationship becomes much simpler, as shown in Figure 2.2.1.4. This anomaly comes about

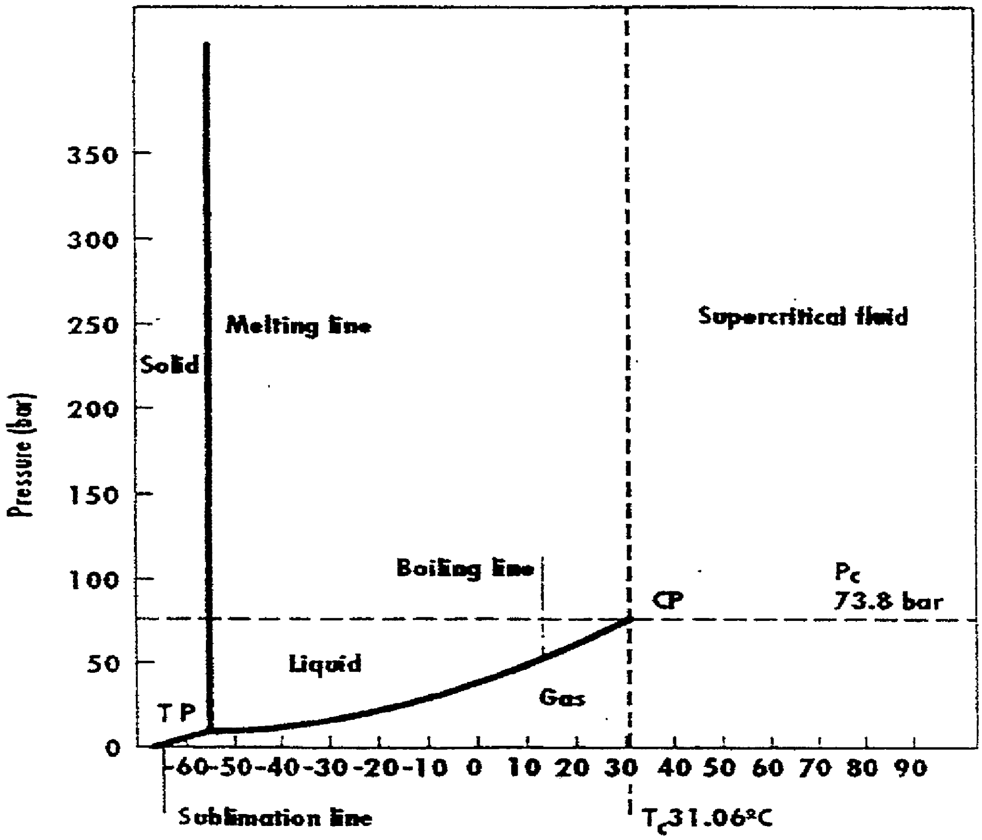

Figure 2.2.1.1 Phase (pressure-temperature) diagram for CO_2: CP=critical point, TP=triple point, P_c= critical pressure, T_c= critical temperature (Brogle, 1982)

because density decreases dramatically with an increase in temperature at low pressure; whereas at higher pressure, changes in temperature have much less effect on density. Thus density, not pressure, to a first approximation is proportional to the solvent power of the supercritical fluid. Figure 2.2.1.5 shows the density of CO_2 as a function of pressure at different temperatures (Fattori et al., 1988). This figure indicates that an increase in temperature results in a decrease in the density of carbon dioxide at a constant pressure. On

For a material at temperatures just above the critical temperature of the substance, liquid-like densities are rapidly approached with modest increases in pressure, (i.e., approximately 0.7-2 times the critical pressure). Higher pressures are required to attain liquid-like densities for temperatures further above the critical temperature. However, Brogle (1982) stated that Supercritical fluid technology would be better served if all scientists-discussed experiments in terms of density rather than pressure.

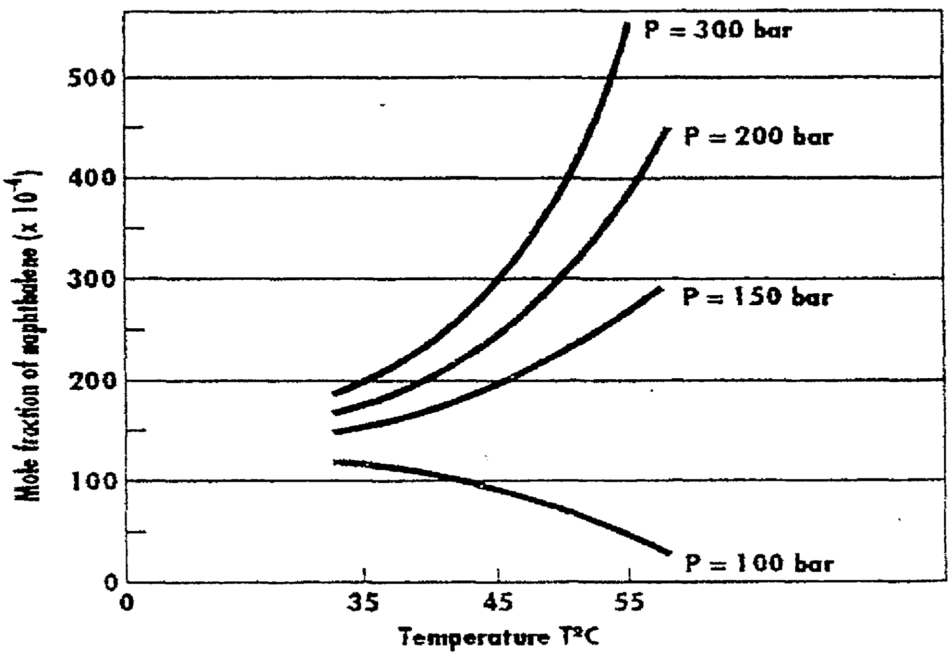

Figure 2.2.1.2 Solubility (mole fraction) of Naphthalene in CO_2 as a function of temperature at various pressures (Brogle, 1982)

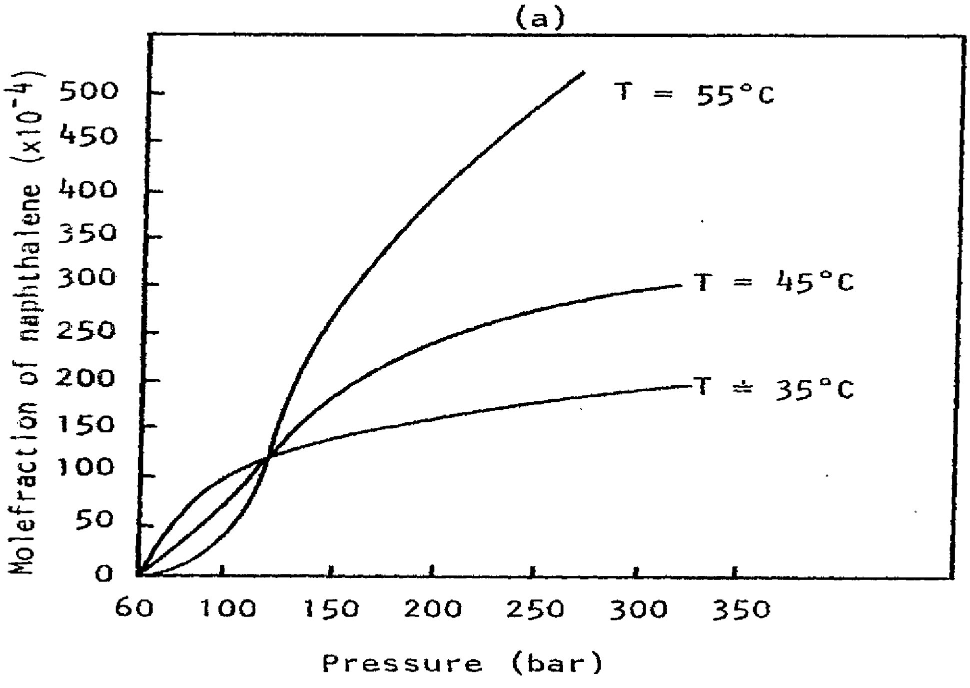

Figure 2.2.1.3 Solubility (mole fraction) of Naphthalene in CO_2 as a function of pressures at various temperatures (Brogle, 1982)

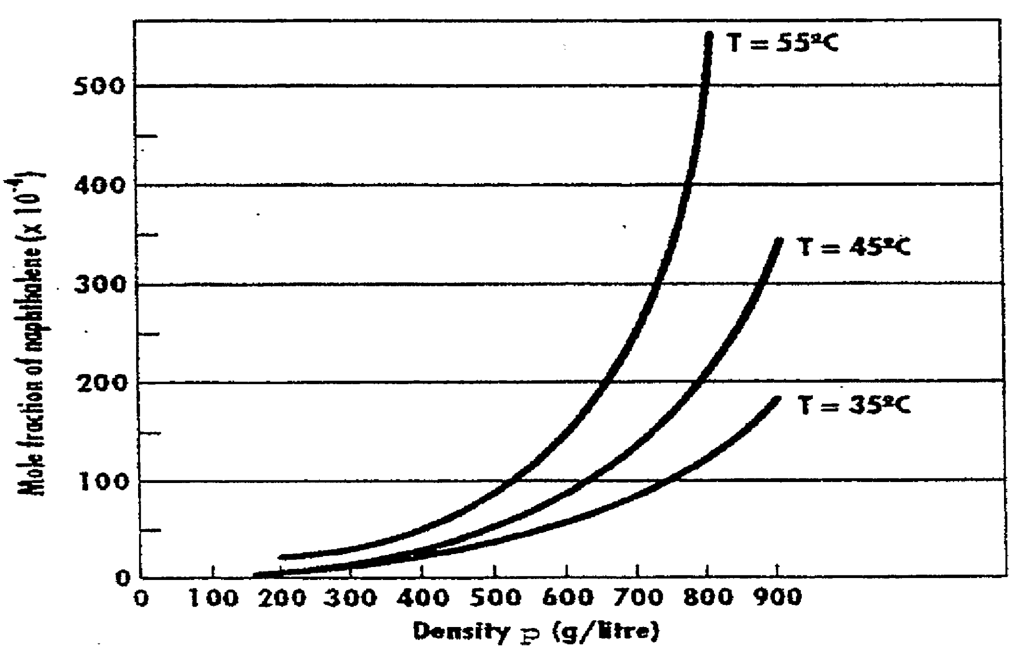

Figure 2.2.1.4 Solubility (mole fraction) of Naphthalene in CO_2 as a function of density at various temperatures (Brogle, 1982)

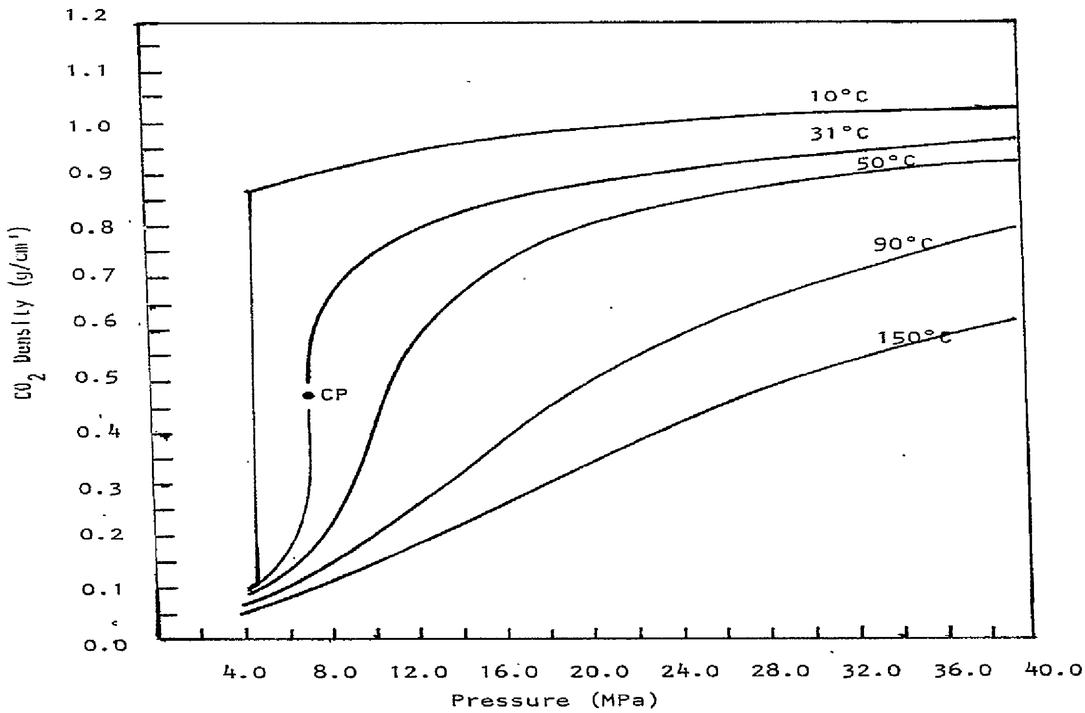

Figure 2.2.1.5 Density of carbon dioxide as a function of pressure at different temperatures. The critical point (Cp) of the CO_2 is indicated on the diagram (Fattori et al., 1988)

Table 2.2.1.1 Supercritical fluid carbon dioxide (SFC)/supercritical fluid extraction (SFE) pressure unit conversion factors (Taylor, 1996)

To convert pressure in one of these units......	... to pressure in one of these units, multiply by:				
	atm	bar	MPa	psi	kg/cm^2
atm.	1	1.0132	0.10132	14.696	1.0332
bar	0.98692	1	0.1	14.504	1.0197
MPa	9.8692	10	1	145.04	10.197
psi	0.068046	0.068948	0.0068948	1	0.7030
kg/CM^{2a}	0.96784	0.98067	0.098067	14.224	1

Source: Provided by Dionex Inc., Sunnyvale, CA.
[a] Strictly, kg/cm^2 is not a pressure unit. It use assumes standard acceleration of gravity

Table 2.2.1.1 contains conversion factors for other pressure units used in Supercritical Fluid Carbon dioxide (SFC)/Supercritical Fluid Extraction (SFE).

Table 2.2.1.2 Density-temperature-pressure (bar) relationship (Taylor, 1996)

Density	Temparature								
(g/mL)	40°C	50°C	60°C	70°C	80°C	90°C	100°C	110°C	120°C
1.00	526	618							
0.95	383	463	544	644	680				
0.90	281	350	420	489	518				
0.85	211	269	329	401	447				
0.80	164	213	264	314	365	416	467		
0.75	134	175	218	261	305	348	392	436	510
0.70	115	150	187	2 23	260	297	334	372	425
0.65	104	133	165	196	227	259	290	322	354
0.60	97	122	149	176	203	229	256	284	311
0.55	93	115	138	161	183	206	230	252	276
0.50	91	109	129	148	168	188	207	227	246
0.45	89	104	122	138	155	172	188	205	221
0.40	87	100	115	129	143	157	171	185	197
0.35	84	96	108	120	132	144	155	167	178
0.30	81	90	101	111	121	130	140	149	158
0.25	77	84	93	100	108	116	123	130	137
0.20	70	75	82	88	94	99	105	110	116

Source: Adapted from Hewlett Packard Co. (Wilmington, DE) literature.

Table 2.2.1.2 gives a more detailed listing of the pressure (in bar) requirements necessary to achieve specific CO_2 densities at various temperatures (Figure 2.2.1.4). The listing once again emphasizes the need for higher pressures at higher temperatures to achieve a specified density.

2.2.2 Diffusivity/Viscosity consideration

Taylor (1996) also stated that supercritical fluid exhibits physicochemical properties intermediate between a liquid and a gas. In addition to its relatively high, liquid-like density at high pressure, which affords good solvent power, mass transfer relative to a liquid is rapid in SFs. For pressures between 50 and 500 atm, diffusivity of supercritical CO_2 varies between 10^{-4} and 10^{-13} cm^2/sec. Similarly the viscosities of supercritical fluids mirror their diffusivities being 10-1000 times lower than liquids. The values for viscosity and diffusivity are dependent on temperature and pressure. The viscosity and diffusivity of the SF approach of a liquid as pressure is increased. Whereas an increase in temperature leads to an increase in viscosity of a gas, the opposite is true in the case of SFs. Diffusivity, on the other hand, will increase with an increase in temperature. The properties of gas-like diffusivity and viscosity, coupled with liquid-like density, combined with the pressure-dependent solvating power of SFs have provided the impetus for applying SF technology to analytical separation problems (Mc Hugh & Krukonis, 1986). Finally, the low (essentially zero) value of surface tension of SFs allows better penetration into the sample matrix relative to liquid solvents. As evidenced by Figures 2.2.3.1 and 2.2.3.2, changes in viscosity and diffusivity are most pronounced in the region about the critical point (McHugh and Krukonis, 1986). Taylor (1996) found that at fixed density viscosity decrease and diffusivity increase with increase in temperature and at fixed temperature diffusivity decrease and viscosity increase with increase in density (McHugh and Krukonis, 1986).

Even at high pressures (300-400 atm), viscosity and diffusivity of SFs differ by 1-2 orders of magnitude from normal liquids. A review of these important points follows:

- Fixed density, temperature ↑, Diffusivity ↑, viscosity ↓

- Density ↑, fixed temperature, diffusivity ↓, viscosity ↑

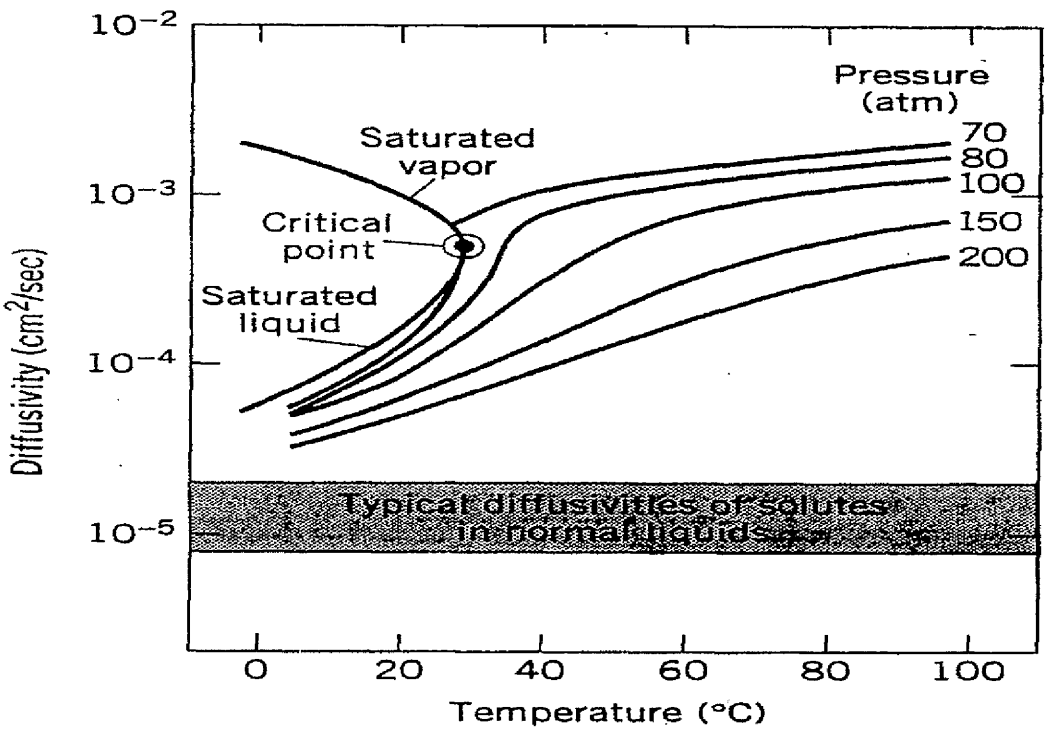

Figure 2.2.2.1 Diffusivity of CO_2 versus temperature at various pressures (McHugh & Krukonis, 1986)

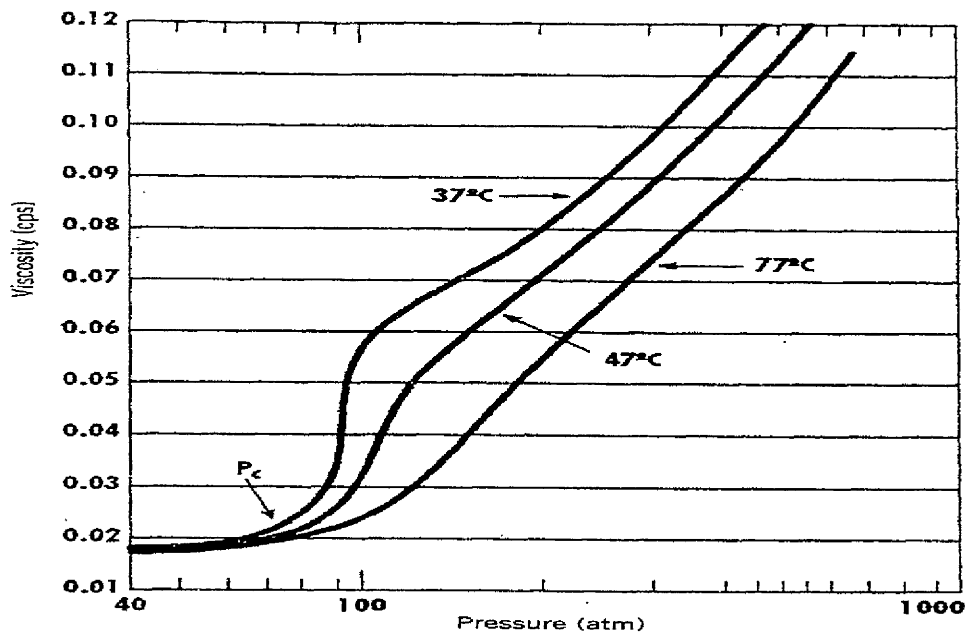

Figure 2.2.2.2 Viscosity behaviour of CO_2 at various temperatures and pressures (McHugh and Krukonis, 1986)

Table 2.2.2.1 Physical properties of gas, liquid and supercritical fluid, SFC (Rizvi et al., 1986)

Properties	Gas	SCF	Liquid
Density (gcm^{-3})	(0.6-2.0) X1-01	0.2-0.9	0.6-1.6
Diffusivity (CM^2sec^{-1})	0.1-0.4	(0.2-0.7) X10-1	(0.2-2.0) X10-1
Viscosity ($gcm^{-1}sec^{-1}$)	(1-3) $X10^{-4}$	(1-9) $X10^{4}$	(0.2-3.0) $X10^{2}$