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FACULDADE DE ENGENHARIA DE ALIMENTOS
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"DESACIDIFICAÇÃO DO ÓLEO DE FARELO DE ARROZ POR EXTRAÇÃO LÍQUIDO-LÍQUIDO"

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"Somos feitos do mesmo material de que são feitos os sonhos"
(William Shakespeare)

Edson e Thiago, dedico a vocês a realização deste sonho.

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"Feliz aquele que transfere o que sabe e aprende o que ensina".

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Resumo

Este trabalho de pesquisa teve como objetivo o estudo do processo de desacidificação do óleo de farelo de arroz por extração líquido-líquido (ELL), em equipamento contínuo, utilizando como solvente etanol azeotrópico, bem como a determinação experimental e modelagem do equilíbrio de fases do sistema óleo de farelo de arroz / ácido graxo / solvente alcoólico. Este processo alternativo de desacidificação foi investigado sob a ótica de sua capacidade em minimizar a perda de óleo neutro, capacidade de extrair os ácidos graxos livres e capacidade de minimizar a perda de antioxidantes naturais, γ-orizanol, tocoferóis e tocotrienóis.

Para se atingir os objetivos propostos, um amplo estudo do equilíbrio de fases de sistemas compostos por óleo de farelo de arroz e solvente alcoólico foi realizado. Os dados de equilíbrio foram correlacionados por modelos semi-empíricos descritivos (NRTL e UNIQUAC) e os parâmetros de interação binária obtidos foram utilizados para a predição do equilíbrio de fases e na simulação do processo de extração em equipamento contínuo. Dados de equilíbrio também foram analisados por metodologia de superfície de resposta (MSR). Esta análise permitiu o conhecimento da influência do teor de acidez no óleo e do teor de água no solvente etanólico sobre as perdas de óleo neutro, dos compostos nutracêuticos, e sobre a transferência dos ácidos graxos livres, do óleo para a fase extrato, no estágio de equilíbrio. Finalmente, o óleo de farelo de arroz desacidificado por extração líquido-líquido foi extensivamente caracterizado para se avaliar o impacto do processo de extração na qualidade do óleo desacidificado.

O processo de desacidificação por ELL de um óleo de farelo de arroz com 8,5% de acidez possibilitou a obtenção de um óleo com baixo nível de ácidos graxos livres (menor que 0,30% em massa, conforme o recomendado pela legislação), com altos teores de compostos nutracêuticos (cerca de 580 ppm de tocoferóis totais e 1,3 % de γ-orizanol), além de gerar uma baixa perda de óleo neutro (menor que 4%).

Abstract

The objective of this work was to evaluate the rice bran oil deacidification process by liquid-liquid extraction (LLE), in a continuous equipment, using aqueous ethanol as solvent. The alternative process was evaluated in terms of: ability to extract the free fatty acids; ability to minimize the neutral oil loss and, ability to preserve nutraceutical compounds, γ-oryzanol, tocopherols and tocotrienols, in the deacidified oil.

The present work reports experimental equilibrium data for fatty systems containing rice bran oil, free fatty acids, γ-oryzanol, tocols, ethanol and water. These experimental data were used to adjust the interaction parameters of the NRTL and UNIQUAC models. The adjusted parameters allowed a quite successful prediction of liquid-liquid equilibrium for systems containing crude rice bran oil plus ethanolic solvent.

The process variable influence on the losses/transfers of fatty compounds during the deacidification process were also investigated using the response surface methodology. Finally, the performance of a perforated rotating disc contactor was evaluated in the deacidification process. The mass transfer coefficients were obtained and the simulation of extraction process presented good agreement with experimental data. A extensively characterization of the oil deacidified by liquid extraction was done to evaluate the impact of extraction process on the quality of product obtained.

The rice bran oil deacidified by LLE presented low level of free fatty acids (lower than 0.30 %, in mass) and high levels of nutraceutical compounds (580 ppm of tocopherols/tocotrienols and 1.3 % of γ-oryzanol, in mass). The losses of neutral oil (lower than 4%, in mass) and nutraceutical compounds were significantly lower than the results reported in the literature for alkali or physical refining of rice bran oil. These results indicate the feasibility of the total deacidification of edible oils by solvent extraction.

Introdução

Os óleos vegetais comestíveis constituem uma importante fonte de energia, ácidos graxos essenciais, vitaminas e antioxidantes lipossolúveis. O processo de refino ao qual é submetido um óleo bruto, para que este atinja o grau comestível, é decisivo na qualidade (odor, sabor, cor), na funcionalidade (composição em ácidos graxos, vitaminas e antioxidantes) e no custo (perda de óleo neutro, equipamentos e custo energético) do produto final.

O óleo de farelo de arroz destaca-se entre os outros óleos comestíveis devido a apresentar, em sua composição, um alto nível de substâncias com valor nutracêutico, tais como o γ-orizanol, os tocoferóis e os tocotrienóis, os quais têm sido citados como poderosos agentes antioxidantes e eficientes na prevenção de doenças degenerativas. No entanto, o óleo de farelo de arroz é produzido atualmente no Brasil em níveis bem inferiores ao seu potencial devido, principalmente, à alta acidez no óleo bruto que dificulta o refino por meio dos métodos usuais, refino químico e físico, ocasionando grande perda de óleo neutro e perda das características nutracêuticas. Desta forma, o estudo de um processo alternativo de desacidificação do óleo de farelo de arroz torna-se relevante.

A desacidificação de óleos vegetais por extração líquido-líquido (ELL) têm-se mostrado como rota alternativa na obtenção de óleos vegetais com teores aceitáveis de ácidos graxos livres. A razão potencial deste novo processo está no fato de consumir menor quantidade de energia em relação ao refino físico, pois é realizado a temperatura ambiente e pressão atmosférica, e no fato de não gerar sabões e minimizar a perda de óleo neutro, se comparado ao refino alcalino.

Este trabalho de tese teve como objetivo o estudo do processo de desacidificação do óleo de farelo de arroz por extração líquido-líquido, em equipamento contínuo, utilizando solvente alcoólico, bem como a determinação experimental e modelagem do equilíbrio de fases do sistema óleo de farelo de arroz / ácido graxo / solvente alcoólico. Este processo alternativo de

desacidificação foi investigado sob a ótica de sua capacidade em minimizar a perda de óleo neutro, capacidade de extrair os ácidos graxos livres e capacidade de minimizar a perda de antioxidantes naturais, γ-orizanol, tocoferóis e tocotrienóis.

Para se atingir os objetivos propostos, um amplo estudo do equilíbrio de fases de sistemas compostos por óleo de farelo de arroz e solvente alcoólico foi realizado. Os dados de equilíbrio foram correlacionados por modelos semi-empíricos descritivos (NRTL e UNIQUAC) e os parâmetros de interação binária obtidos foram utilizados para a predição do equilíbrio de fases e na simulação do processo de extração em equipamento contínuo. Dados de equilíbrio também foram analisados por metodologia de superfície de resposta, permitindo o conhecimento da influência do teor de acidez no óleo e do teor de água no solvente etanólico sobre as perdas de óleo neutro, dos compostos nutracêuticos, e sobre a transferência dos ácidos graxos livres, do óleo para a fase extrato, no estágio de equilíbrio. Finalmente, o óleo de farelo de arroz desacidificado por extração líquido-líquido foi extensivamente caracterizado para se avaliar o impacto do processo de extração na qualidade do óleo desacidificado.

Os capítulos que compõem esta tese de doutorado são, em sua maioria, artigos publicados ou submetidos à publicação em revistas científicas durante o desenvolvimento da pesquisa. Os assuntos abordados em cada capítulo estão resumidos a seguir:

Capítulo 1: Este capítulo apresenta uma breve revisão da literatura acerca do óleo de farelo de arroz, seus principais componentes minoritários, γ-orizanol e vitamina E, e os tipos de refino aos quais este óleo pode ser submetido para tornar-se comestível. Este capítulo apresenta, também, tópicos pertinentes à extração líquido-líquido tais como o estudo do equilíbrio de fases.

Capítulo 2: Este capítulo apresenta dados de equilíbrio líquido-líquido para o sistema óleo de farelo de arroz + ácidos graxos livres + etanol + água bem como a correlação destes dados utilizando as equações NRTL e UNIQUAC.

Capítulo 3: Neste capítulo são apresentados dados experimentais de partição dos compostos γ-orizanol e tocoferóis totais presentes no óleo de farelo de arroz, em sistemas de duas fases líquidas, compostos por óleo de farelo de arroz refinado, com adição de ácido graxo comercial, ou óleo de farelo de arroz bruto e solvente etanol aquoso. Os dados de equilíbrio líquido-líquido foram correlacionados pelas equações NRTL e UNIQUAC e os parâmetros de interação binária obtidos foram testados na predição do equilíbrio de fases de sistemas graxos.

Capítulo 4: Este capítulo apresenta a influência das variáveis de processo nas perdas/transferências dos compostos graxos durante o processo de desacidificação do óleo de farelo de arroz por extração líquido-líquido. A influência das variáveis do processo foi analisada por metodologia de superfície de resposta, objetivando maximizar a transferência dos ácidos graxos livres, do óleo a ser desacidificado para a fase extrato, e minimizar as perdas de óleo neutro e compostos nutracêuticos.

Capítulo 5: O estudo do processo contínuo de desacidificação do óleo de farelo de arroz por extração líquido-líquido em uma coluna de discos rotativos perfurados (PRDC) é apresentado neste capítulo. A influência do tipo de pré-processamento do óleo sobre a transferência de massa foi avaliada. Além disso, o processo de desacidificação foi simulado computacionalmente e o óleo de farelo de arroz degomado, desacidificado por extração líquido-líquido, foi extensivamente caracterizado para se conhecer o impacto da técnica de refino sobre a qualidade do produto obtido.

CAPÍTULO 1

Estado da Arte

1.1 Introdução

Os óleos vegetais comestíveis fazem parte da dieta tradicional da maioria dos povos. Além de conferirem cor e sabor e melhorarem consideravelmente a aparência e palatabilidade dos alimentos, os óleos vegetais são provedores de energia e fonte de ácidos graxos, por exemplo o ácido linoléico, essencial ao bom funcionamento do organismo humano. Além dos ácidos graxos essenciais, os óleos vegetais contém componentes minoritários insaponificáveis com importantes características antioxidantes e vitamínicas. Um óleo vegetal rico nestes componentes minoritários é o óleo de farelo de arroz.

A fração insaponificável do óleo de farelo de arroz, cerca de 4% do óleo bruto (Orthoefer, 1996), contém um complexo único de substâncias antioxidantes de ocorrência natural constituído pela vitamina E (tocoferóis e, principalmente, tocotrienóis) e o γ-orizanol (Hu *et. al*., 1996). O γ-orizanol tem sido descrito como poderoso agente antioxidante e hipocolesterolêmico (Seetharamaiah e Chandrasekhara, 1993; Sugano e Tsuji, 1997; Deckere e Korver, 1996; Rong *et. al*., 1997) e os tocotrienóis, que apresentam, também, propriedade antioxidante, têm sido citados em recentes pesquisas como eficientes na prevenção de doenças cardiovasculares e algumas formas de câncer (Eitenmiller, 1997; Tomeo *et. al*., 1995; Qureshi *et. al*., 1997).

Apesar destas características nutricionais positivas, o óleo de farelo de arroz é produzido atualmente no Brasil em níveis bem inferiores ao seu potencial devido a dificuldades encontradas em seu processamento, ocasionadas principalmente por enzimas, particularmente lipases, que aumentam o teor de ácidos graxos livres após a extração do óleo do farelo (Orthoefer, 1996; Scavariello e Barrera-Arellano, 1998; Bhattacharyya *et. al*., 1987). A alta porcentagem de ácidos graxos livres, gerada pela alta atividade enzimática, ocasiona grande perda de óleo neutro nos processos de refino, tanto no processo físico, quanto no químico. Além disso, o uso

destes processos tradicionais de refino também afetam o conteúdo de γ-orizanol e tocotrienóis no óleo (Orthoefer, 1996).

Desta forma, para que estes componentes, γ-orizanol e tocotrienóis, os quais apresentam propriedades fisiológicas benéficas, sejam mantidos no óleo refinado em quantidades apreciáveis e, principalmente, para que este tipo de óleo seja refinado a grau comestível com mínima perda de óleo neutro, é necessário que o processo tradicional de refino do óleo de farelo de arroz bruto seja modificado.

Uma alternativa de modificação para o processo de refino é a substituição da etapa de desacidificação convencional por um processo contínuo de desacidificação por extração líquido-líquido. Este tipo de processo foi investigado enquanto sua capacidade em minimizar a perda de óleo neutro, de minimizar a perda de antioxidantes naturais, γ-orizanol, tocoferóis e tocotrienóis, e de extrair os ácidos graxos livres, para obtenção de óleo com teor de acidez livre menor que 0,3% (em massa), conforme o recomendado pelo Codex Alimentarius (1993).

1.2. Óleos Vegetais

1.2.1. Composição dos Óleos Vegetais

Os óleos vegetais são substâncias líquidas, lipofílicas, formadas predominantemente por produtos de condensação entre o glicerol e ácidos graxos, os chamados triacilgliceróis.

Os triacilgliceróis constituem cerca de 95% do óleo vegetal bruto, enquanto que os constituintes minoritários são formados por produtos de decomposição dos triacilgliceróis, mono, diacilgliceróis e ácidos graxos livres, fosfolipídios e ceras e, também, por substâncias genericamente denominadas de não-glicerídios ou insaponificáveis tais como esteróis, vitaminas, pigmentos, hidrocarbonetos e metais (Lawson, 1985).

1.2.2. O Óleo de Farelo de Arroz

Embora a produção de arroz represente ¼ da produção mundial de cereais, o óleo de farelo de arroz, um subproduto da indústria arrozeira, é atualmente produzido no Brasil em escala bem inferior ao seu potencial.

De acordo com a InfoArroz, a produção mundial de arroz em casca foi de 576 milhões de toneladas em 2002/03 (InfoArroz, 2003) e a projeção para o ano 2004 é de 591 milhões de toneladas, sendo que 91% da produção se concentra na Ásia, 3% na África, 3% na América do Sul e 3% em outras regiões (InfoArroz, 2003).

O Brasil, produzindo 11 milhões de toneladas/ano, é o principal produtor de arroz fora do continente asiático, ocupando a posição de 9º maior produtor mundial (InfoArroz, 2003). O brasileiro consome, em média, 70 quilos de arroz por ano, enquanto o argentino 11 quilos e o uruguaio 10 quilos, números muito distantes de países como Vietnã (175 kg/hab/ano), Indonésia (155 kg), China (103 kg) e Índia (85 kg).

O Rio Grande do Sul é responsável por 50% do arroz produzido no país e por 73,8% do arroz irrigado, sistema que também é empregado em larga escala em Santa Catarina, em boa parte de Mato Grosso, estado que tirou do Maranhão o posto de segundo produtor brasileiro de arroz, no Tocantins e em Goiás.

Atualmente, a produção anual mundial de OFA é cerca de 724 mil toneladas, porém, segundo cálculos da FAO, considerando um rendimento de 15% sobre 50% do farelo produzido, o potencial de produtividade do óleo é de 4,16 milhões de toneladas. Desta forma, sendo o Brasil um grande produtor de arroz, seu potencial de produção de óleo de farelo de arroz seria em torno de 84 mil toneladas ao ano (Anuário Estatístico Do Brasil, 1994), um volume significativo que ultrapassaria, por exemplo, a produção de óleo de milho brasileira, que atualmente é de 72 mil toneladas ao ano.

Apesar desta projeção otimista a balança do agronegócio brasileiro de dezembro de 2002 a novembro de 2003 mostra uma produção de farelo de arroz de apenas 132 toneladas (Balança Comercial do Agronegócio, 2003).

Nos Estados Unidos, a produção do óleo de farelo de arroz foi iniciada nos anos 50 e descontinuada nos anos 80. O interesse por esse óleo foi retomado na década de 90 devido às oportunidades de exportação, principalmente para o Japão, onde este tipo de óleo é largamente consumido (Deckere e Korver, 1996). Além disso, os resultados nutricionais positivos correlacionados ao consumo do óleo de farelo de arroz ajudaram a impulsionar a retomada de produção deste óleo nos EUA (Orthoefer, 1996).

O grão de arroz é constituído genericamente por 20% de casca, 70% de endosperma e 10% pelas camadas de farelo e germe, onde estão concentrados a maior parte dos lipídios do grão (15 a 20%) (Orthoefer, 1996; McCaskill e Zhang, 1999).

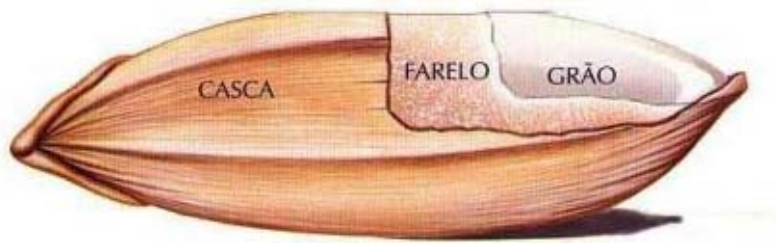

Figura 1.1. Componentes de um grão de arroz (Fonte da imagem: Josapar, 2004)

O beneficiamento do grão de arroz envolve etapas como a secagem do grão, descascamento, moagem e polimento (Wang e Luh, 1991). Durante as etapas de beneficiamento são separadas as diversas partes do grão dentre elas o farelo. O farelo de arroz, como as outras fontes vegetais de óleos, possui vários sistemas enzimáticos (α-amilase, β-amilase, catalase, lipoxigenase, peroxidade e lipases) (Luh *et al*., 1991).

Estas enzimas estão fisicamente isoladas do óleo enquanto o farelo permanece intacto no grão. Durante a etapa de moagem a estrutura do farelo é rompida e as enzimas são misturadas com o óleo conduzindo-o ao desenvolvimento da rancidez oxidativa e hidrólise. A hidrólise ocorre devido à presença das lipases que levam à produção de ácidos graxos livres. Já a rancidez oxidativa é gerada pela degradação dos lipídios em compostos químicos de cadeias curtas (Godber *et al*., 1993).

O teor de ácidos graxos livres (AGL) no óleo de farelo de arroz (OFA) bruto pode chegar até 70% em peso (Gupta, 1989), sendo que pode ocorrer um incremento diário de 4 a 5% no teor de AGL, devido às condições de armazenagem do farelo, tais como temperatura e umidade, que afetam largamente a taxa de hidrólise.

Muitos métodos tem sido desenvolvidos visando a inativação enzimática no farelo, para assim aumentar o tempo de vida da matéria-prima. Estes métodos incluem o uso da secagem do farelo, tratamento com irradiação gama e ou reagentes químicos (Salunke *et al*., 1991). Porém, a solução mais adequada seria a extração do óleo ou a estabilização do farelo imediatamente após a moagem, o que na maioria das vezes é impossível devido à capacidade reduzida dos moinhos e grande descentralização dos mesmos (Gupta, 1989).

O processo geral de extração do óleo à partir do farelo de arroz consiste das seguintes etapas:

- etapa de limpeza na qual são removidos materiais estranhos como casca, arroz quebrado, por meio de peneiras e aspiração (Kao e Luh, 1991);
- etapa de tratamento térmico, que tem como principal objetivo estabilizar o farelo por inativação térmica das lipases. Atualmente o cozimento por extrusão têm apresentado bons resultados (Saunders, 1986);
- etapa de extração do óleo do farelo por solvente. O hexano é o solvente mais utilizado, porém outros tipos de solventes mais polares tem sido estudados, tais

como o isopropanol (Hu *et al*., 1996; Xu e Godber, 2000) e o acetato de etila (Xu e Godber, 2000).

Os estudos envolvendo estes solventes mostram que o isopropanol possui menor eficiência de extração de óleo bruto, porém o óleo extraído apresenta maior conteúdo de vitamina E e quantidade similar de γ-orizanol (Hu et al., 1996). Estudos mais recentes realizados por Xu e Godber (2000) compararam a eficiência de extração de óleo e o teor de γ-orizanol no óleo bruto extraído com a utilização de solvente misto de hexano/isopropanol (50:50) e com a utilização de fluido supercrítico. Este último método apresentou alta produtividade de óleo com altos teores de γ-orizanol. A temperatura do solvente durante a extração do óleo do farelo afeta o conteúdo de ceras no óleo bruto (Kao e Luh, 1991). Menores temperaturas de extração fornecem óleos brutos com menores teores de ceras e constituídos principalmente de lipídios neutros (Orthoefer, 1996).

Krishna (1993) observou que as ceras podem formar miscelas ocluindo triacilgliceróis neutros em seu interior; desta forma, grande quantidade de óleo neutro ocluído é arrastada na etapa de deceragem. Ademais, altos teores de AGL também maximizam as perdas de óleo neutro no refino, encarecendo o processo.

A composição típica do óleo de farelo de arroz bruto é de 68 a 71% de triacilgliceróis, 2 a 3% de diacilgliceróis, 5 a 6% de monoacilgliceróis, 2 a 3% de ácidos graxos livres, 2 a 3% de ceras, 5 a 7% de glicolipídios, 3 a 4% de fosfolipídios e até 5% de matéria insaponificável (McCaskill e Zhang, 1999).

Quanto à composição em ácidos graxos têm-se que os ácidos oléico e linoléico constituem cerca de 80% dos ácidos graxos dos acilgliceróis (40% de oléico e 40% de linoléico). Este óleo contém, também, quantidades detectáveis do ácido graxo α-linolênico (1 a 3%) e a maior quantidade de ácido graxo saturado no óleo de farelo de arroz é o ácido palmítico (17%) (Sugano e Tsuji, 1997; McCaskill e Zhang, 1999).

1.2.3. Aspectos Nutricionais dos Óleos Vegetais Comestíveis

Há um consenso entre a comunidade científica de que um balanço entre a quantidade de ácidos graxos (saturados, monoinsaturados e ácidos graxos poliinsaturados ω3 e ω6) ingeridos na dieta diária é importante. O balanço certo entre os ácidos graxos protege o organismo humano contra doenças crônicas tais como doenças cardiovasculares (Deckere e Korver, 1996).

Muitos óleos vegetais tais como soja, girassol, canola, devido a apresentarem em sua composição quantidades apreciáveis de ácido linoléico (ω6), desempenham um importante papel no intuito de ajudar o organismo a atingir este balanço (Deckere e Korver, 1996). Além dos ácidos graxos essenciais, os óleos vegetais contém componentes minoritários insaponificáveis com características antioxidantes e vitamínicas. Um óleo vegetal rico nestes componentes minoritários é o óleo de farelo de arroz.

1.2.4. Aspectos Nutricionais da Matéria Insaponificável do Óleo de Farelo de Arroz

O óleo de farelo de arroz bruto apresenta, em média, 4% de matéria insaponificável dentre os quais 2% de γ-orizanol (Scavariello e Barrera-Arellano, 1998). Além do orizanol, o óleo de farelo de arroz é um dos únicos óleos vegetais, juntamente com o óleo de palma, que contém significantes níveis de tocotrienóis (cerca de 1000 ppm) (McCaskill e Zhang, 1999).

O γ-orizanol é uma mistura complexa de ésteres de ácido ferúlico com fitosteróis e álcoois triterpênicos (Scavariello e Barrera-Arellano, 1998). A alta capacidade antioxidante do γ-orizanol tem sido amplamente reconhecida e estudos tem mostrado vários efeitos fisiológicos relacionados a este composto.

A Tabela 1.1 mostra a solubilidade do γ-orizanol em diversos solventes (Tsuno Rice Fine Chemicals Co, 1995). Este composto possui solubilidade média em metiletilcetona e é muito pouco solúvel em água.

Tabela 1.1. Solubilidade do γ-orizanol em diversos solventes.

Solvente	Solubilidade a 20°C (g/l)
Água	0,1
Etanol	1,3
Metanol	1,4
n-Hexano	2,8
n-Heptano	4,0
Cloreto de etileno	20,0
Benzeno	40,0
Acetona	67,0
Clorofórmio	100,0
Metiletilcetona	200,0

Fonte: Tsuno Rice Fine Chemicals Co., 1995.

A presença de γ-orizanol no OFA confere a este óleo uma alta resistência à oxidação e deterioração. Estudos mostram que o material ativo é o ácido ferúlico (Kanno *et al*., 1985), porém a esterificação deste ácido com os esteróis das plantas, a forma do γ-orizanol, aumenta o potencial antioxidante (Graf, 1992; Sharma, 1976).

Atualmente têm-se relacionado ao γ-orizanol efeitos fisiológicos benéficos com a ingestão de OFA. Os estudos mostram a habilidade do γ-orizanol em reduzir os níveis de colesterol no plasma sangüíneo (Sugano e Tsuji, 1997; Deckere e Korver, 1996; Lichenstein *et al*., 1994), reduzir a absorção de colesterol pelo organismo (Rong *et al*., 1997), decrescendo a arteriosclerose precoce pela inibição da agregação de placas de gordura e aumento da excreção fecal de ácidos biliares (Seetharamaiah et al., 1990; Seetharamaiah e Chandrasekhara, 1990). A administração do γ-orizanol também é citada no tratamento de distúrbios nervosos e desordens decorrentes da menopausa (Nakayama *et al*., 1987).

As estruturas químicas da maioria dos componentes do γ-orizanol encontrados no OFA são similares à do colesterol. Esta similaridade pode ser responsável pelo efeito na absorção e síntese do colesterol (Rogers *et al*., 1993). A síntese de matéria insaponificável, como o γ-orizanol, nas plantas, apresenta etapas

iniciais similares às da síntese de colesterol no corpo humano, o que sugere que estes compostos podem alterar a síntese do colesterol e competirem na absorção.

De forma geral a síntese do colesterol no corpo humano é regulada pelo nível de colesterol no sangue, que por sua vez depende do colesterol exógeno proveniente da dieta. Provavelmente o γ-orizanol atua na inibição da enzima HMG-CoA reductase, a qual regula a síntese de colesterol endógeno. O organismo reconhece o γ-orizanol como se fosse colesterol, inibindo a ação enzimática da HMG-CoA reductase e, conseqüentemente, diminuindo a síntese de colesterol endógeno (Hegsted e Kousik, 1994).

Quanto aos tocotrienóis estes aparecem em quatro formas conhecidas (α, β, γ , δ) e são similares aos tocoferóis em estrutura química. O α-tocotrienol exerce 45% da atividade da vitamina E e a forma γ é a mais importante forma dos tocotrienóis, apresentando-se como a mais ativa no efeito de reduzir o nível de colesterol no plasma sangüíneo (Deckere e Korver, 1996; Qureshi *et. al*., 1997; Mccaskill e Zhang, 1999). Recentes pesquisas mostram, também, que os tocotrienóis podem suprimir o crescimento de células cancerígenas (Eitenmiller, 1997).

1.2.5. Processos de Refino de Óleos Vegetais

Refino é um termo genérico utilizado para denominar as etapas de purificação dos óleos vegetais brutos com o intuito de remover substâncias indesejáveis. A remoção dos ácidos graxos livres é o passo mais importante do processo de refino, principalmente porque o rendimento do óleo neutro nesta etapa tem um efeito significativo no custo do processo.

Além da remoção dos ácidos graxos livres os processos de refino visam a remoção de fosfolipídios, pigmentos, ceras e traços de metais. Entretanto, a permanência no óleo refinado de algumas substâncias tais como esteróis, tocoferóis e tocotrienóis, é altamente desejável devido a suas características antioxidantes e vitamínicas (Kim *et. al*., 1985).

A desacidificação, ou seja, a remoção dos ácidos graxos livres, tem sido realizada, predominantemente, por neutralização destes ácidos graxos livres com soda cáustica, no denominado refino químico, convertendo-os em sabões, os quais são insolúveis no óleo na temperatura de operação. Esta pode variar de 50 a 90°C dependendo da composição do óleo e do álcali utilizado (Hartman, 1971; Antoniassi *et. al*., 1998).

Além do refino químico, algumas refinarias tem utilizado o processo denominado refino físico para a desacidificação de óleos. Este processo baseia-se na diferença considerável entre os pontos de ebulição dos ácidos graxos livres e dos triacilgliceróis nas condições de operação, as quais requerem baixas pressões, 3 a 6 mmHg, e altas temperaturas, de 220 a 270 °C, dependendo do óleo (Hartman, 1971; Antoniassi *et. al*., 1998).

O refino químico é um processo versátil e bem conhecido que pode ser aplicado para qualquer óleo bruto; porém, quanto maior a porcentagem de ácidos graxos livres no óleo bruto, maior a perda de óleo neutro, a qual ocorre devido à saponificação e alta emulsibilidade do óleo no sabão formado (Antoniassi *et. al*., 1998). As perdas de óleo neutro, para óleos de milho bruto com conteúdos de ácidos graxos livres entre 8 e 14%, podem atingir de 15 a 25%, no refino alcalino, de acordo com Leibovitz e Ruckenstein (1983) e cerca de 14%, em refinarias brasileiras, para óleos com 4% de acidez (Antoniassi *et. al*., 1998).

De acordo com Orthoefer (1996) as perdas de óleo neutro no refino químico do OFA são consideravelmente altas (18 a 22%) se comparadas a outros óleos vegetais com conteúdos similares de AGL. Este fato é devido, principalmente, ao efeito sinergético das ceras e do γ-orizanol que maximiza a perda de óleo neutro (Mishira *et al*., 1988).

Krishna (1993) observou um efeito sinergético entre γ-orizanol, monoacilgliceróis e ceras no incremento da viscosidade do óleo de farelo de arroz bruto e, conseqüentemente, um incremento das perdas de óleo neutro no refino

alcalino. O autor sugere a eliminação dos monoacilgliceróis e γ-orizanol antes da etapa de deceragem, minimizando, assim, a perda de óleo neutro na etapa de desacidificação. Os monoacilgliceróis e o γ-orizanol dificultam a sedimentação das ceras na etapa de deceragem e as ceras, por sua vez, ocluem óleo neutro.

Além das grandes perdas de óleo neutro o refino químico ocasiona uma larga produção de sabão e poluição ambiental. Processos alternativos tem sido propostos por diversos autores, a fim de se obter alta produtividade de óleo de boa qualidade.

Processos baseados na reesterificação química ou bioquímica dos AGL são boas alternativas para OFA com alta acidez. A desacidificação enzimática de óleos vegetais através de lipases, o biorefino, tem apresentado bons resultados. Este processo baseia-se na capacidade das lipases de esterificar os AGL com os grupos hidroxila já presentes no óleo ou em grupos hidroxila de substâncias adicionadas, como o glicerol (Bhattacharyya e Bhattacharyya, 1989; Sengupta e Bhattacharyya, 1996; Kosugi *et al*., 1994). Este processo fornece óleo de boa qualidade por subseqüente refino alcalino, branqueamento e desodorização.

A reesterificação através de catalisadores ácidos e sais metálicos forneceu óleo de cor aceitável (Bhattacharyya e Bhattacharyya, 1987); além disso, monoacilgliceróis (De e Bhattacharyya, 1999) e diacilgliceróis (Kurashige, 1988) foram estudados como fontes de grupos hidroxila para esterificação dos AGL através de lipases. Em todos os casos, o principal inconveniente é o alto custo das enzimas, que impossibilita o processo industrial.

Já o refino físico pode ser economicamente vantajoso para óleos com altos teores de ácidos graxos livres, entretanto, este tipo de processo só é aplicável para óleos com baixo teor de fosfolipídios e para óleos onde as condições extremas de operação, baixas pressões e altas temperaturas, não tenham impacto negativo na qualidade final do produto (Antoniassi *et. al*., 1998). A desacidificação pelo método físico não é tão efetiva quanto a desacidificação alcalina (Carr, 1978) e a qualidade

do produto final é sempre determinada pela qualidade do óleo bruto (Cvengros, 1995).

Alguns autores propõem o refino físico como uma alternativa para a desacidificação do OFA (Kim *et al*., 1985; Tandy e McPherson, 1984; De e Bhattacharyya, 1998). Este processo remove os AGL juntamente com os compostos odoríferos pela purga de vapor saturado através do óleo a alta temperatura e alto vácuo, porém o OFA refinado fisicamente apresenta cor mais escura se comparado ao refino químico (Antoniassi *et al*., 1998).

O rendimento do refino físico depende da qualidade do óleo bruto. Quanto mais ácido e maior quantidade de ceras, orizanol e fosfolipídios o óleo bruto contiver, maior a perda de óleo neutro na destilação e pior a qualidade do produto final em termos de cor (Prabhakar *et al*., 1988).

Deve-se ressaltar que para ambos tipos de processos de refino, químico e físico, o conteúdo de antioxidantes naturais do óleo de farelo de arroz, tocotrienóis, tocoferóis e γ-orizanol, são afetados.

Considerando-se que o óleo de farelo de arroz bruto contém cerca de 2% de γ-orizanol, com a degomagem, etapa precedente à desacidificação que elimina, principalmente, os fosfolipídios, a quantidade deste antioxidante se reduz a 1,7%. O refino físico passa o teor de γ-orizanol no óleo para 1,0 a 1,5%, enquanto que no refino químico o teor de γ-orizanol cai drasticamente para 0,1% (Kim *et al*., 1985; Orthoefer, 1996).

1.3. Extração Líquido-Líquido

A extração líquido-líquido, também conhecida por extração líquida ou extração por solvente, é a separação dos constituintes de uma solução líquida, denominada alimentação, por contato íntimo com outro líquido apropriado, imiscível ou parcialmente miscível, denominado solvente, o qual deve ter a capacidade de extrair preferencialmente um ou mais componentes desejados

(soluto). Originam-se deste contato duas novas correntes, o refinado, que é a solução residual da alimentação, pobre em solvente, com um ou mais de um dos solutos removidos pela extração, e o extrato, rico em solvente, contendo o soluto extraído.

Nesta operação de transferência de massa as duas fases, alimentação e solvente, são quimicamente muito diferentes, o que leva a uma separação dos componentes de acordo com suas propriedades físico-químicas (Treybal, 1980).

1.3.1. Desacidificação de Óleos Vegetais por Extração Líquido-líquido

A desacidificação de óleos vegetais por extração líquido-líquido têm-se mostrado como rota alternativa na obtenção de óleos vegetais com teores aceitáveis de ácidos graxos livres. A razão potencial deste processo está no fato de consumir menor quantidade de energia, pois é realizado a temperatura ambiente e pressão atmosférica. Estas condições brandas de operação são, também, muito favoráveis para óleos que não aceitam as temperaturas altas do refino físico (220 a 270 °C) (Hamm, 1983). Além da economia de energia, a extração líquido-líquido apresenta vantagem em relação ao refino químico, pois não gera sabões e minimiza a perda de óleo neutro.

De acordo com Thomopoulos (1971) a desacidificação de óleos vegetais por extração líquido-líquido é baseada na diferença de solubilidade dos ácidos graxos livres e dos triacilgliceróis neutros no solvente e na diferença de ponto de ebulição do solvente e dos ácidos graxos livres. Este último aspecto torna muito fácil a recuperação posterior do solvente para sua reutilização.

Os trabalhos existentes na literatura sobre desacidificação de óleos vegetais por solvente são limitados. O primeiro trabalho reportado, de autoria de Fachini e Samazzi, data de 1925. Os autores estudaram a desacidificação de ázeites de oliva de alta acidez com etanol. Mais tarde, Schlenker (1931) estudou os equilíbrios de distribuição dos ácidos graxos entre as fases oleosa e alcoólica. Martinez-Moreno (1947) descreveu resultados de desacidificação de azeite de oliva em processo

contínuo em coluna empacotada usando etanol aquoso. Kim *et al*. (1985) estudaram o comportamento de metanol, etanol e isopropanol (94,7%) na desacidificação do OFA com 25% de acidez, em funil de separação. Em linhas gerais a acidez decresceu para 4% e as perdas de óleo neutro aumentaram com o aumento da razão solvente/óleo. Os três tipos de solventes apresentaram capacidades correlatas de desacidificação, porém, em termos de perda de óleo neutro, o metanol destacou-se, apresentando 15% de perda em comparação aos 85% apresentados pelo etanol e isopropanol.

Bhattacharyya *et al*. (1987) estudaram a eficiência do isopropanol como solvente para desacidificação do OFA por extração líquido-líquido, em batelada, como um pré-tratamento do óleo, com acidez de 20 a 56%, antes do refino alcalino. Os autores observaram que o isopropanol azeotrópico (91%) apresentou bons resultados, reduzindo de 56% de AGL para 7,2% com 3 contatos de extração, em funil de separação. Observou-se, porém, que este procedimento de desacidificação não afetou a cor do óleo.

Estudos com isopropanol hidratado foram realizados por Shah e Venkatesan (1989) e a extração dos AGL com a utilização de etanol hidratado na miscela foi realizada por Türkay e Civelekoglu (1991). Seus resultados mostram ser a extração líquido-líquido um processo promissor na desacidificação de óleos vegetais.

Monnerat (1994) determinou dados de equilíbrio líquido-líquido para sistemas do tipo triacilgliceróis / ácidos graxos / álcoois de cadeia curta (metanol, etanol, isopropanol). A autora observou ser o etanol hidratado o solvente mais adequado para ser aplicado no processo de desacidificação de óleos vegetais.

Antoniassi (1996) estudou o processo de desacidificação contínua por extração líquido-líquido do óleo de milho bruto, utilizando coluna de discos rotativos perfurados (PRDC). Pina e Meirelles (2000) realizaram estudos sobre a desacidificação do óleo de milho por extração líquido-líquido, utilizando etanol a 96% como solvente, em colunas de discos rotativos (RDC) e de discos rotativos

perfurados (PRDC). Bons resultados foram obtidos quanto a teores aceitáveis de ácidos graxos livres, menor que 0,3% para óleos com acidez de 3,5%, e perdas de óleo neutro de até 4,8%. A perda de óleo neutro pode ser considerada baixa se comparada às perdas em torno de 14% por refino químico, para o mesmo teor de ácido graxo no óleo bruto, obtidas nas refinarias de óleo de milho no Brasil (Antoniassi et al., 1998).

Em termos de retenção dos antioxidantes naturais no óleo, após a etapa de refino por extração líquido-líquido, Kim *et al*. (1985) mostraram que o γ-orizanol praticamente não foi afetado pelo refino com metanol, embora a quantidade de tocoferóis tenha sido diminuída consideravelmente.

1.3.2. Equipamentos para a Extração Líquido-líquido

No processo de extração líquido-líquido, as duas fases, alimentação e solvente, devem ser colocadas em contato íntimo com um alto grau de turbulência, garantindo altas taxas de transferência de massa. Desta forma, um equipamento de extração deve apresentar as funções de colocar os líquidos em contato, criar gotas da fase dispersa, afim de fornecer área interfacial para a transferência de massa, e separar os líquidos ao final da extração.

Industrialmente apresentam aplicabilidade os seguintes equipamentos de extração:

- misturadores-decantadores, extratores centrífugos, colunas não agitadas (spray, empacotadas, pratos perfurados) e colunas agitadas (Scheibel, Oldshue, Kuhni, RDC, ARDC).

Devido ao aumento da taxa de transferência de massa para o contato líquido-líquido quando a agitação mecânica é fornecida, as colunas agitadas são as mais comumente usadas. Um modelo popular para extração líquido-líquido é a coluna de discos rotativos (RDC), a qual oferece facilidade de projeto, construção, manutenção e flexibilidade de operação (Reman, 1951).

Estudos de desacidificação de óleos vegetais por extração líquido-líquido, realizados no LASEFI-FEA-UNICAMP, utilizando colunas de discos rotativos, original (RDC) e versão modificada com discos perfurados (PRDC), mostraram o bom desempenho deste tipo de equipamento no processamento de sistemas graxos nos quais uma das fases, a fase oleosa, apresenta alta viscosidade e deve ser introduzida no equipamento como fase dispersa (Antoniassi, 1996; Pina, 2001). Estes testes atestaram a adequabilidade deste tipo de coluna no processo de desacidificação do óleo de milho bruto pré-tratado, garantindo tanto um bom contato entre as fases, como o tempo necessário para sua separação na zona de decantação do equipamento.

1.3.3. Coluna de discos rotativos perfurados

A PRDC consiste de um cilindro vertical, no qual são introduzidos discos perfurados presos a um eixo central, ligado a um motor de velocidade variável, visando promover a dispersão e o contato entre as fases. As alimentações são introduzidas perpendiculares à direção do escoamento. Para reduzir o efeito do movimento dos líquidos e garantir a separação das fases, duas zonas mortas, uma abaixo e outra acima da região de extração, fazem parte do equipamento. A retirada de amostras da fase refinado é feita no compartimento inferior, e a da fase extrato na saída desta corrente no topo da coluna de extração (Pina, 2001). A Figura 1.2 mostra um esquema da PRDC utilizada neste trabalho de tese enquanto a Figura 1.3 mostra a PRDC existente no Laboratório de Separações Físicas.

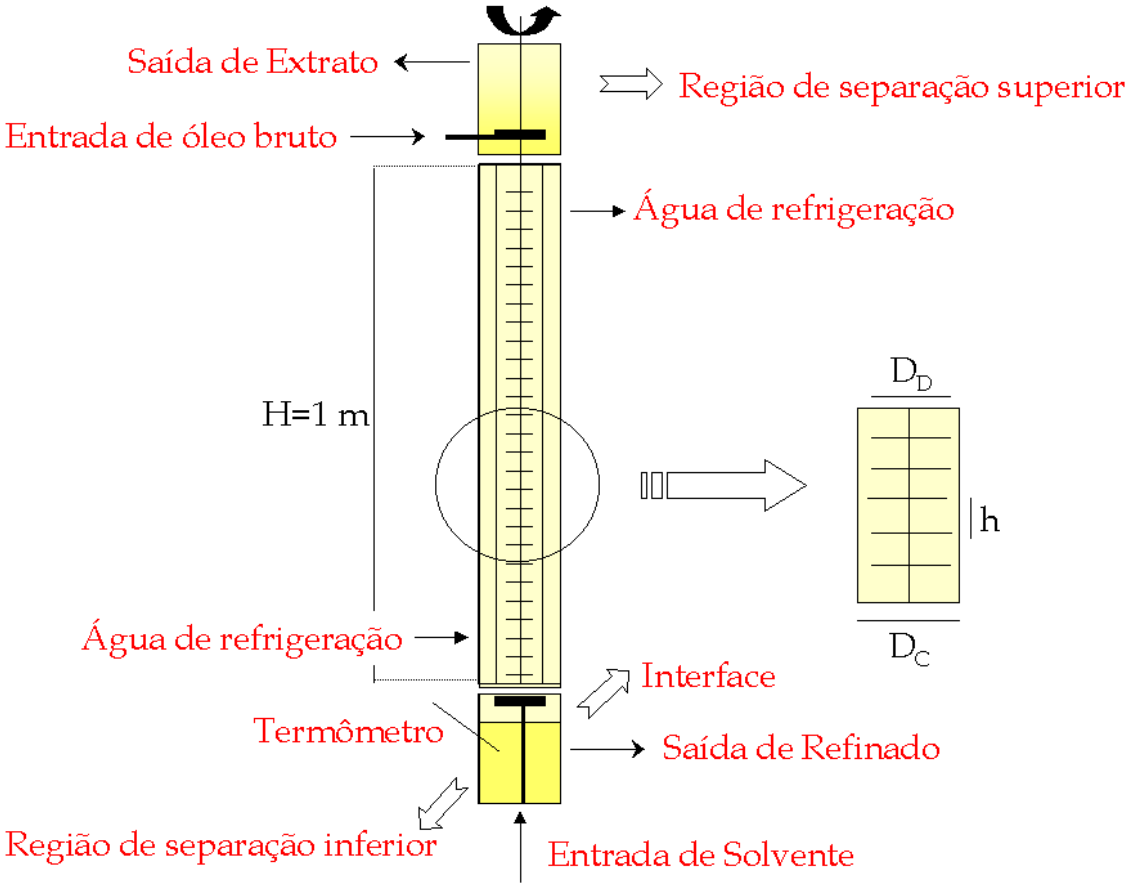

Figura 1.2. Diagrama esquemático da coluna: DC é o diâmetro interno da coluna, DD é o diâmetro do disco, h é a altura do compartimento e H é a altura da zona de extração.

Figura 1.3. Coluna de extração líquido-líquido (PRDC) existente no LASEFI.

Antoniassi (1996) estudou a desacidificação do óleo de milho, pré-tratado por diferentes processos, em uma coluna de discos rotativos (RDC) com área livre de escoamento de 4 e 20%. O óleo de milho branqueado foi considerado como o mais adequado para a alimentação da coluna de discos rotativos, pelo baixo conteúdo de umidade e fósforo (Antoniassi, 1996).

Pina (2001), estudou a eficiência de colunas de discos rotativos perfurados (PRDC) na desacidificação de óleo de milho refinado com adição de ácido oléico comercial, e concluiu que a PRDC sem chicanas foi a versão mais apropriada para a desacidificação de óleos vegetais.

Além da extração líquido- líquido, a PRDC também pode ser empregada com sistemas aquosos bifásicos (ATPS) para a extração e purificação de proteínas, por exemplo. De acordo com Cunha (2003), a PRDC é mais adequada para sistemas com baixa tensão superficial. Este autor estudou a extração de cutinase com sistemas aquosos bifásicos (ATPS) e comparou um sistema de extração em batelada com a extração contínua em PRDC. A extração contínua proporcionou uma capacidade de separação 2,5 vezes maior do que a extração em batelada (Cunha, 2003).

Os estudos de Carneiro-da-Cunha (1994) demonstraram a eficiência da extração da cutinase com coluna PRDC e sistemas micelares. Tambourgi *et al*. (1999) estudaram a extração das proteínas citocromo b5, protease e ascórbico oxirredutase utilizando o sistema bifásico polietileno-glicol (PEG) - fosfato de potássio, com uma coluna PRDC. Porto *et al*. (2000) trabalharam com a extração de albumina do soro bovino neste mesmo tipo de coluna. Sarubbo *et al*. (2003) também estudaram o mecanismo de transferência de massa da proteína de soro bovino em uma PRDC, utilizando um sistema aquoso bifásico composto de PEG-polissacarídeo (goma do cajueiro).

1.3.4. Equilíbrio de Fases

Os diagramas de equilíbrio líquido-líquido, os quais permitem a visualização da região onde existem as duas fases liquidas em equilíbrio, são apresentados a temperatura constante. A Figura 1.4 apresenta, como exemplo, um diagrama de equilíbrio constituído por óleo de farelo de arroz (diluente – w_1) / ácido oléico (soluto – w_2) / etanol+água (solvente – w_{3+4}). O diagrama pode ser considerado como composto por um par parcialmente miscível (solvente-diluente) e dois pares completamente miscíveis (soluto-diluente e soluto-solvente), apresentando então uma região de duas fases líquidas em equilíbrio, dependente da concentração de ácido graxo (w_2).

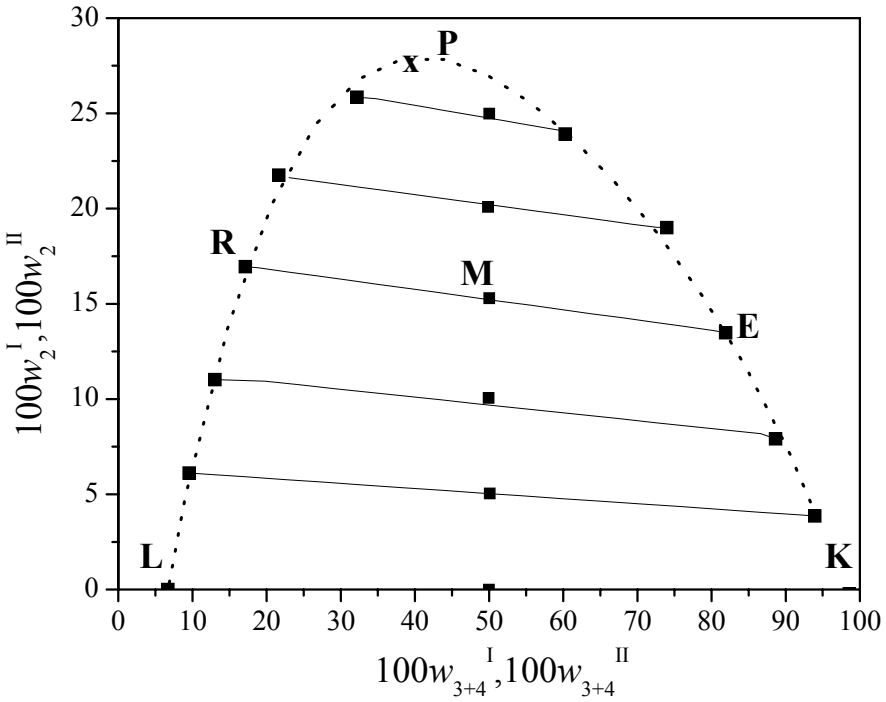

Figura 1.4. Diagrama de equilíbrio a 25,0 ±0,1°C para o sistema Óleo de Farelo de Arroz (1) / Ácido Oléico (2) / Etanol Hidratado com 9,92 ±0,01% de água em massa (3+4).

Neste tipo de sistema, o aumento da concentração do soluto aumenta a miscibilidade mútua entre o óleo e o solvente alcoólico, de modo que os três pseudo-componentes formam uma fase homogênea, desde que haja quantidade suficiente de ácido graxo no sistema. A nomeação dos componentes graxos como sendo pseudo-componentes é razoável, devido ao óleo vegetal ser uma substância composta de diversos triacilgliceróis.

Embora se trate de fato de um sistema multicomponente, os diversos triacilgliceróis presentes no óleo de farelo de arroz comportam-se, neste tipo de sistema, de forma muito similar, podendo, para fins práticos, serem aproximados por um triacilglicerol hipotético com a massa molar média do óleo. O mesmo procedimento pode ser considerado para o ácido oléico comercial.

Desta forma, trata-se de um sistema que pode ser bem aproximado por um pseudo-ternário. Embora a água tenha características bem diferentes do etanol, sendo do ponto de vista prático imiscível com os compostos graxos, o uso de um solvente alcoólico com baixo teor de água não muda, de forma significativa, as considerações apresentadas acima, permitindo, ainda, analisar o sistema em diagramas triangulares.

Nos diagramas de equilíbrio (vide Figura 1.4), os pontos localizados à direita (curva LRP) representam as concentrações obtidas experimentalmente para os componentes da fase alcoólica (fase II), enquanto que os pontos localizados à esquerda (curva PEK) representam as concentrações na fase oleosa (fase I). Os pontos no centro do diagrama referem-se aos pontos de mistura (M), obtidos pela concentração mássica inicial do sistema. O ponto L representa a solubilidade dos componentes (3+4) em 1 e o ponto K a solubilidade do componente 1 em 3.

A curva LRPEK é a curva binodal e apresenta a mudança de solubilidade das fases I e II com a adição do soluto. Qualquer mistura fora desta curva será uma solução homogênea de uma fase, enquanto que qualquer mistura dentro da curva, por exemplo a mistura M, formará duas fases líquidas imiscíveis com as composições indicadas em R (rica no componente 1) e E (rica nos componentes 3 e

4). A linha RE é a linha de amarração ou "*tie-line*", que deverá passar necessariamente pelo ponto M. O ponto P, conhecido como ponto crítico ou "*plait-point*", representa a última linha de amarração e o ponto onde as curvas de solubilidade das fases ricas nos componentes 1 e 3+4 se encontram.

A razão entre as concentrações (fração mássica) de ácido oléico w_2 na fase alcoólica (I) e na fase oleosa (II) representa o coeficiente de distribuição do ácido oléico o qual, também, pode ser visualizado pela inclinação das linhas de amarração no diagrama de equilíbrio (Figura 1.4). Matematicamente o coeficiente de distribuição (k) é representado pela equação 1.1 a seguir:

$$k_i = \frac{w_i^{II}}{w_i^{I}} \qquad (1.1)$$

No exemplo apresentado na Figura 1.4, a concentração de 2 na fase II é menor que na fase I; portanto, o coeficiente de distribuição será menor que 1.

Outra definição importante no estudo do equilíbrio de fases é a seletividade (S) do solvente (matematicamente representada pela equação 1.2) que mostra como um determinado solvente diferencia os ácidos graxos livres (componente 2) e os triacilgliceróis (componente 1) no processo de extração.

$$S = \frac{k_2}{k_1} \qquad (1.2)$$

Na desacidificação de óleos vegetais por extração líquido-líquido é desejável a utilização de solvente que possibilite k maior que a unidade e alta seletividade (S) aos ácidos graxos livres. Monnerat (1994) constatou que o etanol com 6 % de água em massa apresentou coeficiente de distribuição próximo a unidade e maior seletividade, se comparado ao etanol anidro, sendo, desta forma, um solvente adequado à desacidificação de óleos vegetais.

Para a simulação de processos químicos, como a extração, são requeridas estimativas de propriedades da mistura. Como não é possível obter todos os dados para a mistura particular nas condições de temperatura, pressão e composição

correspondentes às do estudo, é necessário, então, manipular os poucos dados experimentais de tal forma a obter-se a melhor interpolação e extrapolação. Isto é particularmente importante no caso do equilíbrio de fases de sistemas graxos que envolvem uma grande variedade de componentes e diferentes níveis de concentração.

A este respeito, deve-se considerar que as relações de equilíbrio são decisivas no cálculo da força motriz para transferência de massa e no cômputo das concentrações das fases que estão em contato no equipamento. É a partir do conhecimento destas composições que outras propriedades físicas importantes no processo (densidade, viscosidade, tensão interfacial, difusividade, etc.) podem ser estimadas.

Nas condições de equilíbrio termodinâmico de 2 fases líquidas, tem-se que:

$$f_i^I = f_i^{II} \tag{1.3}$$

onde: f_i^I - fugacidade do componente i na fase I

f_i^{II} - fugacidade do componente i na fase II

A equação (1.3) pode ser reescrita como:

$$x_i^I \gamma_i^I = x_i^{II} \gamma_i^{II} \tag{1.4}$$

onde: x_i^I - fração molar do componente i na fase I

γ_i^I - coeficiente de atividade do componente i na fase I

x_i^{II} - fração molar do componente i na fase II

γ_i^{II} - coeficiente de atividade do componente i na fase II

Muitas expressões semi-empíricas têm sido propostas na literatura para relacionar os coeficientes de atividade à composição e temperatura da mistura. Todas estas expressões contém parâmetros ajustáveis a dados experimentais, sendo que os principais modelos sugeridos para o equilíbrio líquido-líquido são as equações NRTL e UNIQUAC, cuja grande vantagem é permitir a extensão dos parâmetros obtidos pelo ajuste dos modelos a sistemas binários para o cálculo do

equilíbrio em sistemas multicomponentes contendo os mesmos constituintes. As equações para os modelos NRTL e UNIQUAC estão explicitadas no capítulo 3, equações 3.10 a 3.13 e equações 3.14 a 3.19, respectivamente.

Para sistemas ideais ou moderadamente ideais, o modelo NRTL não oferece muita vantagem sobre outros modelos, como Van Laar ou Margules-três sufixos, mas para sistemas fortemente não ideais esta equação pode fornecer uma boa representação dos dados experimentais, embora sejam necessários dados de boa qualidade para estimar os três parâmetros. O modelo NRTL também pode ser facilmente estendido para misturas multicomponentes (Renon e Prausnitz, 1968; Aznár, 2001).

O modelo UNIQUAC foi desenvolvido por Abrams e Prausnitz (1975), e é, de alguma maneira, uma extensão da teoria quase-química de Guggenheim para moléculas não-randômicas a misturas contendo componentes de diferente tamanho. Esta extensão foi chamada de Teoria Quase-química Universal, ou, pela sigla em inglês, UNIQUAC. A equação UNIQUAC consiste em duas partes: uma parte combinatorial, que descreve as contribuições entrópicas dos componentes, e que depende apenas da composição e do tamanho e forma das moléculas; e uma parte residual, que expressa as forças intermoleculares (de onde aparecem dois parâmetros ajustáveis a dados experimentais) devido às interações energéticas, e que são responsáveis pela entalpia de mistura.

Vale notar que a equação UNIQUAC dispõe de dois parâmetros ajustáveis para cada par de componentes presentes no sistema, ao invés de três parâmetros, como o modelo NRTL. Outro aspecto a ser mencionado é que, tanto a equação UNIQUAC quanto a equação NRTL, foram originalmente formuladas em fração molar, mas as equações apresentadas no capítulo 3 estão devidamente transformadas para unidades de fração mássica. Devido à grande diferença de massa molecular dos compostos que estarão presentes em nosso sistema ($M_{triacilgliceróis}$ = 833-912 kg/kmol; $M_{ácidos\ graxos}$ = 200-313 kg/kmol; M_{etanol} = 46

kg/kmol; $M_{água}$ =18 kg/kmol), trabalhar com fração mássica permite um ajuste mais preciso do modelo aos dados experimentais do que empregar fração molar.

No LASEFI - FEA - UNICAMP foram utilizadas as equações NRTL e UNIQUAC para modelar o equilíbrio líquido-líquido dos sistemas graxos óleo de canola/ácido oléico com diferentes solventes: metanol anidro, etanol anidro, isopropanol anidro e etanol azeotrópico (aquoso) (Batista e*t al.*, 1999), e óleo de milho / ácido oléico / etanol com diferentes graus de hidratação (Gonçalves *et. al.*, 2002).

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CAPÍTULO 2

Equilibrium Data for the System Rice Bran Oil + Fatty Acids + Ethanol + Water at 298.2 K

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Abstract

This work presents experimental data for the model system refined rice bran oil + commercial oleic acid + ethanol + water at 298.2 K. These data were correlated by the NRTL and UNIQUAC models, with a global deviation of approximately 0.7% for both models. The equilibrium of crude rice bran oil + aqueous ethanol was predicted with success using the adjusted interaction parameters, with deviation between calculated and experimental results not higher than 0.54%. The results showed that the addition of water to solvent increases the solvent selectivity, reducing the losses of neutral oil and nutraceutical compounds, and expands the region of phase splitting, allowing the refining of highly acidic crude rice bran oils by solvent extraction.

2.1. Introduction

Medical studies indicate the hypocholesterolemic effect of rice bran oil in humans and animals. The majority of such studies suggests that rice bran oil is more effective in decreasing serum and liver cholesterol concentrations than oils with similar fatty acid composition, such as groundnut oil.[1-3]

The lowering of cholesterol levels by rice oil may be attributed to its high level of unsaponifiable matter.[1,2,4] Crude rice bran oil may contain up to 5% of unsaponifiable matter. This level is reduced to values up to 1.5% in the refined rice bran oil. In contrast, most refined vegetable oils contains only 0.3-0.9% of unsaponifiable matter.[5] The crude rice bran oil unsaponifiable matter contains a unique complex of antioxidant compounds, such as (100 to 1000) ppm of Vitamin E (tocopherols and tocotrienols) and (0.9 to 2.9)% of γ-orizanol. This last compound is in fact a complex mixture of triterpene alcohols and phytosterols esterified with ferulic acid.[6,7]

In addition to the hypocholesterolemic activity of these rice oil minor compounds, the isolated ingestion of γ-orizanol has been efficient to decrease early

atherosclerosis[3], to treat nerve imbalance disorders of menopause[8] and inflammatory processes[9]. In relation to vitamin E, with special attention to the tocotrienol fraction, some investigations have shown the efficiency of these compounds in the prevention of cardiovascular disease and some forms of cancer[10-12].

Despite these advantages that make the rice bran oil a functional food, its world production in edible grade is very low (1% of all vegetable oil production) due to difficulties in its processing. The production and refining of vegetable oils consist in its extraction from oilseeds, bran or fruit pulps using hexane petroleum fractions as solvent[13,14], solvent stripping, degumming, bleaching, deacidification and deodorization.[15,16] In comparison with other vegetable oils, crude rice oil tends to contain higher levels of free fatty acids (FFA) induced by intensive enzymatic activity. The high FFA content makes difficult the oil deacidification by the traditional processes, chemical or physical refining.

The chemical refining is based on a saponification reaction of FFA with an alkali solution, it can result in oil losses of (18 to 22) mass% according to Orthoefer[3]; furthermore, the production of soapstock is very large. In relation to physical refining, which is performed at high temperatures and very low pressures, several authors mention disadvantages, such as alterations in oil color, reduction of stability to oxidation and high-energy consumption.[17-19]

It should be further emphasized that both processes induce a significant loss of nutraceutical compounds. Rice bran oil refined by chemical method contains less than 0.2% of oryzanol[3,20] and, in the physical refining, a significant portion of vitamin E is stripped away with the distillate during the deodorization step.[4]

An alternative refining process, performed under more mild conditions (room temperature and atmospheric pressure) is the deacidification by liquid-liquid extraction, which also avoids the formation of waste products. Liquid-liquid extraction for oil refining is based on the difference of solubility of FFA and neutral triacylglycerols in an appropriate solvent. [21] Bhatacharyya et al.[22] and Shah and

Venkatesan[23] studied the deacidification of rice bran and groundnut oils using aqueous 2-propanol as solvent. Kim et al.[24] and Kale et al.[18] tested methanol in the refining of rice bran oil. All studies showed a decreasing of the oil acidic value. Deacidification by liquid-liquid extraction may produce vegetable oils with low acidic levels, and simultaneously minimize the loss of neutral oil and nutraceutical compounds.[24]

Phase equilibrium data are necessary for the design of extraction processes. Batista et al.[25,26] and Gonçalves et al.[27] reported liquid-liquid equilibrium data for systems composed by canola oil + oleic acid + short chain alcohols and corn oil + oleic acid + ethanol + water, respectively. However, equilibrium data for rice oil has not been reported up to date. The aim of this work was to investigate the phase equilibrium of rice bran oil + commercial oleic acid + ethanol + water at 298.2 K. The experimental data were correlated by the NRTL and UNIQUAC equations and the adjusted interaction parameters were used to predict the equilibrium of crude rice bran oil + aqueous ethanol.

2.2. Material

The solvents used in this work were anhydrous ethanol, from Merck, with purity greater than 99.5%, and aqueous solvents with different water contents (2.40±0.02, 6.03±0.01, 6.38±0.02, 8.96±0.05, 10.59±0.04 and 12.41±0.01 mass %), prepared by the addition of deionized water to anhydrous ethanol.

All fatty reagents used in this study, commercial oleic acid (Merck), refined rice bran oil (Tio João, Brazil) and crude rice bran oil (kindly supplied by Helmut Tessmann, Brazil) were analyzed by gas chromatography of fatty acid methyl esters to determine the fatty acid composition, according to the official method (1-62) of the AOCS [28]. Samples were prepared in the form of fatty esters methyl esters according to the methodology developed by Hartman and Lago [29]. A HP5890 gas chromatograph with a flame ionization detector was used under the following

experimental conditions: fuse silica column of cyanopropylsiloxane 0.25 μm, 60 m x 0.32 mm id; hydrogen as the carrier gas at a rate of 2.5 ml/min; injection temperature of 548.2 K; column temperature of 448.2 - 498.2 K (rate of 1.3 K/min); detection temperature of 578.2 K.

The fatty acids methyl esters were identified by comparison with external standards purchased from Nu Check Inc. (Elysian, IL). The quantification was accomplished by internal normalization. The fatty acid compositions of the refined and crude rice bran oils are presented in Table 2.1.

Table 2.1 – Fatty Acid Composition of Refined and Crude Rice Bran Oils

symbol	fatty acid		M^b	refined		crude	
			g·mol^{-1}	mole%	mass%	mole%	mass%
M	Miristic	C14:0[a]	228.38	1.16	0.96		
P	Palmitic	C16:0	256.43	19.57	18.17	21.44	19.91
Po	Palmitoleic	C16:1	254.42	0.66	0.61		
S	Stearic	C18:0	284.49	1.50	1.54	1.79	1.84
O	Oleic	C18:1	282.47	37.66	38.50	39.11	40.01
Li	Linoleic	C18:2	280.45	35.08	35.61	35.78	36.34
Le	Linolenic	C18:3	278.44	2.65	2.67	1.88	1.90
A	Arachidic	C20:0	312.54	1.57	1.78		
Ga	Gadoleic	C20:1	310.52	0.15	0.16		

[a] In C*x:y*. *x*=number of carbons and *y*=number of double bonds; [b] *M*=molar mass.

From this fatty acid composition it was possible to determine the probable triacylglycerol composition of the refined and crude rice bran oil (Table 2.2) by using the algorithm suggested by Antoniosi Filho et al. [30]. In Table 2.2 the main triacylglycerol represents the component of greatest concentration in the isomer set with *x* carbons and *y* double bonds.

Table 2.3 presents the fatty acid composition of commercial oleic acid. This fatty acid was also analyzed by gas chromatograph using the methodology described above. The results showed in Tables 2.2 and 2.3 allows to calculate the average molecular mass of refined rice oil, crude rice oil, free fatty acids in crude rice oil and commercial oleic acid. The values obtained were: 867.78, 866.50, 276.13

and 278.96 g·mol^{-1}, respectively. Crude and refined rice bran oils, free fatty acids in crude rice oil and commercial oleic acid were treated in this work as pseudocompounds with the average molar masses indicated above.

Table 2.2 – Probable Triacylglycerol Composition of Refined and Crude Rice Bran Oils

group	main triacyl glycerol	M[b]	refined		crude	
		g·mol^{-1}	mole%	mass%	mole%	mass%
50:1[a]	POP	833.37	4.66	4.48	5.29	5.09
50:2	PLiP	831.35	4.25	4.07	4.84	4.64
50:3	PLeP	829.35	0.63	0.60		
52:1	POS	861.45	0.87	0.86	0.88	0.88
52:2	POO	859.40	11.73	11.61	11.72	11.63
52:3	POLi	857.39	18.84	18.62	20.01	19.80
52:4	PLiLi	855.37	8.98	8.85	10.18	10.05
52:5	PLiLe	853.37	0.80	0.78	0.96	0.95
54:2	SOO	887.46	1.30	1.33	0.94	0.97
54:3	OOO	885.44	8.25	8.42	7.30	7.46
54:4	OOLi	883.43	17.51	17.83	16.31	16.63
54:5	OLiLi	881.41	15.18	15.42	15.04	15.29
54:6	LiLiLi	879.43	5.49	5.56	5.80	5.89
54:7	LiLiLe	877.38	0.64	0.65	0.72	0.73
56:3	OLiA	913.52	0.59	0.62		
56:4	LiLiA	911.50	0.28	0.30		

[a] In *x*:*y*. *x*=number of carbons (except glicerol carbons) and *y*=number of double bonds;
[b] *M*=molar mass.

Table 2.3 – Fatty Acid Composition of Commercial Oleic Acid

Symbol	Mole %	Mass %
L	1.58	1.13
M	1.09	0.89
P	5.83	5.36
Po	0.13	0.12
S	1.39	1.42
O	77.05	78.02
Li	11.91	11.97
Le	0.49	0.50
A	0.53	0.59

Refined rice bran oil had a residual acidity of 0.07%, expressed as oleic acid. The crude rice oil used in this work presented an acidic value of (9.34±0.01) mass %, expressed as oleic acid. Both oils were analyzed by spectrophotometry, using a UV-VIS dual beam spectrophotometer (Perkin Elmer, model Lambda 40), to determine the presence and concentration of nutraceutical compounds. The concentration of γ-oryzanol was determined at 314.5 nm, as suggested by Seetharamaiah and Prabakar [31], using n-heptane (UV-Fluo, Carlo Erba) as solvent and γ-oryzanol, purity greater than 99%, kindly supplied by Tsuno Rice Fine Chemicals Co., as standard. The quantification of total tocopherols was determined at 520 nm according to the methodology developed by Emmerie-Engel [32]. α-Tocopherol, purity greater than 99% (Sigma), was used as standard and toluene (Em Science) as solvent.

Crude rice bran oil presented (1.72±0.05) mass % of γ-oryzanol and (622.3±4.0) ppm of total tocopherols. γ-Oryzanol concentration in refined rice oil was (0.12±0.01) mass % and tocopherols were not detected in this oil.

2.3. Experimental Procedure

Model fatty systems containing fatty acids and triacylglycerols were prepared by the addition of known quantities of commercial oleic acid to refined rice bran oil. The model fatty systems were mixed with the ethanolic solvents, in the mass ratio oil:solvent 1:1, at (298.2±0.1) K, for determination of liquid-liquid equilibrium data used to adjust NRTL and UNIQUAC parameters. Crude rice bran oil was mixed with aqueous ethanol containing (6.03±0.01) mass % of water or (8.96±0.05) mass % of water, in the mass ratios oil:solvent (1:1, 1:2, 1:3) and (1:1, 1:3), respectively. These data were used to test the prediction capability of the adjusted NRTL and UNIQUAC parameters.

Liquid-liquid equilibrium data were determined using polypropylene centrifuge tubes (50 ml) (Corning Inc.). The components were weighed on an

analytical balance Sartorius model A200S, accurate to 0.0001g. The tubes were vigorously stirred for at least 15 min, centrifuged for 10 min at 4500 *g* (Centrifuge Jouan model BR4i) and left to rest for 2 h in a thermostatic bath at (298.2±0.1) K (Cole Parmer, model 12101-05). This contact time was stated based on a previous study that showed the phase equilibrium was attained after 1 h of rest.

After this treatment, the two phases became clear, with a well-defined interface, and the composition of both phases was measured. The concentration of free fatty acids was determined by titration (official method 2201 of the IUPAC [33]) with an automatic burette (Metrohm, model Dosimat 715). The total solvent concentration was determined by evaporation at 338.2 K in a vacuum oven (Napco model 5831). The water concentration was determined by Karl Fischer titration, according to AOCS method Ca 23-55 [28] with a KF Titrino (Metrohm, model 701). For the refined oil the γ-oryzanol concentration was very low, so that the system was considered as a pseudoquaternary one composed only by triacylglycerols, fatty acids, ethanol and water. In this case, having determined the concentration of fatty acids, solvent and water, the triacylglycerol concentration can be obtained by difference.

For the crude oil the γ-oryzanol concentration in each phase was measured according to the procedure suggested by Seetharamaiah and Prabakar [31]. In this case the system was considered a pseudoquinary one and the triacylglycerol concentration can also be determined by difference.

To have a better insight on the quality of rice bran oil refined by solvent extraction, a further set of experiments were performed to measure only the partition coefficients of the nutraceutical compounds. Such experiments were performed by mixing crude rice bran oil with different aqueous solvents, in the ratio oil:solvent 1:1, at (298.2±0.1) K. The concentration of γ-oryzanol and tocopherol were measured according to the procedure described above.

In this work all measurements were performed at least in triplicate. The uncertainties of the concentrations varied within the following ranges: (0.01 to 0.45) mass% for rice oil, (0.01 to 0.28) mass% for fatty acids, (0.01 to 0.40) mass% for ethanol, (0.01 to 0.04) mass% for water, (0.01 to 0.13) mass% for γ-oryzanol and (0.001 to 0.01) mass% for tocopherols.

2.4. Results

Tables 2.4 and 2.5 present the overall experimental composition of the mixtures, the corresponding tie-lines and the experimental standard deviations for the pseudoternary and pseudoquaternary model systems composed by refined rice bran oil + commercial oleic acid + ethanol and refined rice bran oil + commercial oleic acid + ethanol + water, respectively.

Table 2.4 – Liquid-liquid Equilibrium Data for the System Refined rice bran oil (1) + Commercial oleic acid (2) + Anhydrous Ethanol (3) at 298.2±0.1 K

Overall Composition			Alcohol Phase (II)			Oil Phase (I)		
$100w_1$	$100w_2$	$100w_3$	$100w_1$	$100w_2$	$100w_3$	$100w_1$	$100w_2$	$100w_3$
50.01	0.00	49.99	7.48	0.00	92.52	85.22	0.00	14.78
47.45	2.50	50.06	9.14	2.99	87.87	80.12	2.26	17.62
44.75	5.02	50.23	11.75	5.86	82.38	74.42	4.56	21.01
42.41	7.49	50.10	15.48	8.43	76.09	68.33	6.87	24.80
40.00	10.01	49.99	22.85	10.97	66.18	58.14	9.51	32.34

Table 2.6 shows the overall experimental composition of the mixtures, the corresponding tie-lines and experimental standard deviations for the systems composed by crude rice oil + free fatty acids + ethanolic solution. All concentrations are given as mass percentages.

Table 2.5 – Liquid-liquid Equilibrium Data for the System Refined rice bran oil (1) + Commercial oleic acid (2) + Ethanol (3) + Water (4) at 298.2±0.1 K

$100w_{4S}$[a]	Overall Composition				Alcohol Phase (II)				Oil Phase (I)			
	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_1$	$100w_2$	$100w_3$	$100w_4$
2.40 ±0.02	49.99	0.00	48.81	1.20	4.21	0.00	93.26	2.53	88.56	0.00	11.22	0.22
	47.97	2.01	48.82	1.20	4.92	2.42	90.35	2.31	84.93	2.07	12.70	0.30
	45.70	3.97	49.12	1.21	5.70	4.44	87.46	2.41	82.06	3.72	13.85	0.36
	43.77	6.00	49.02	1.21	6.97	6.82	83.91	2.30	77.91	5.54	16.20	0.36
	41.77	7.98	49.03	1.21	8.48	8.84	80.48	2.20	73.96	7.46	18.18	0.40
6.38 ±0.02	49.90	0.00	46.90	3.20	2.14	0.00	90.85	7.01	91.95	0.00	7.54	0.51
	49.99	2.51	46.82	3.19	2.15	2.61	88.67	6.56	88.36	2.27	9.16	0.21
	49.96	5.00	46.85	3.19	2.88	5.01	86.24	5.88	83.86	5.05	10.45	0.64
	49.80	9.98	47.00	3.20	4.25	10.16	79.88	5.71	75.34	9.90	14.04	0.72
	49.93	14.99	46.88	3.19	7.03	15.49	72.36	5.11	65.58	14.85	18.70	0.88
	49.77	19.90	47.03	3.20	12.89	20.57	62.01	4.53	53.61	19.65	25.25	1.49
10.59 ±0.04	49.88	0.00	44.81	5.31	0.63	0.00	88.42	10.96	93.08	0.00	6.34	0.58
	44.95	5.04	44.71	5.30	1.09	4.53	83.75	10.63	84.70	5.78	8.75	0.78
	39.98	10.01	44.72	5.30	1.94	9.16	79.05	9.85	76.41	11.06	11.60	0.94
	34.92	14.95	44.82	5.31	3.61	14.10	73.36	8.94	67.51	15.89	14.98	1.62
	29.76	20.01	44.73	5.30	6.67	19.42	65.95	7.97	57.39	21.03	19.51	2.07
12.41 ±0.01	49.93	0.00	43.86	6.21	0.16	0.00	88.68	11.16	94.43	0.00	4.54	1.03
	47.31	2.57	43.90	6.22	0.37	2.14	85.49	12.00	89.71	3.24	5.77	1.28
	45.00	5.01	43.79	6.20	0.31	4.06	83.13	12.50	85.68	6.13	6.95	1.23
	39.94	10.04	43.82	6.21	1.13	8.46	78.62	11.78	76.87	12.01	9.74	1.38
	29.95	20.00	43.84	6.21	3.80	18.35	67.32	10.53	58.61	22.31	16.86	2.22

[a]$100w_{4S}$ = water mass percentage in the solvent

Table 2.6 – Liquid-liquid Equilibrium Data for the System Crude rice bran oil (1) + Fatty acids (2) + Ethanol (3) + Water (4) at 298.2±0.1 K

$100w_{4S}$[a]	Overall Composition					Alcohol Phase (II)					Oil Phase (I)				
	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$
6.03 ±0.01	22.25	2.34	70.46	4.52	0.43	1.80	2.39	89.22	6.44	0.16	86.30	2.34	9.44	0.66	1.26
	29.61	3.11	62.69	4.02	0.57	2.08	3.12	88.27	6.33	0.19	84.96	3.08	9.78	0.85	1.34
	44.24	4.64	47.23	3.03	0.86	2.08	4.66	86.44	6.59	0.23	82.11	4.60	11.18	0.70	1.41
8.96 ±0.05	22.28	2.34	68.24	6.71	0.43	0.57	2.31	87.97	8.97	0.18	87.70	2.42	7.66	0.58	1.65
	44.48	4.67	45.51	4.48	0.86	0.61	4.35	85.14	8.86	0.14	84.08	4.86	9.02	0.67	1.37

[a]$100w_{4S}$ = water mass percentage in the solvent

In Figures 2.1 and 2.2 the equilibrium data for the model systems refined rice bran oil + commercial oleic acid + (6.38±0.02) mass % aqueous solvent and refined rice bran oil + commercial oleic acid + (12.41±0.02) mass % aqueous solvent are shown. Figure 2.3 presents the equilibrium data for the system crude rice oil + free fatty acids + (6.03±0.01) mass % aqueous solvent. In these figures ethanol + water were considered as a mixed solvent.

A good alignment can be observed between the experimental data, relative to both overall and phase concentrations. Tie lines based on the experimental data were determined by linear regression of each corresponding set of overall, oil and alcoholic phase concentrations. Correlation coefficients higher than 99.5% were obtained for all tie lines, indicating a low error in the experimental determination of the tie line compositions. The low experimental deviations obtained in the measurement of the phase composition strengthen this comment.

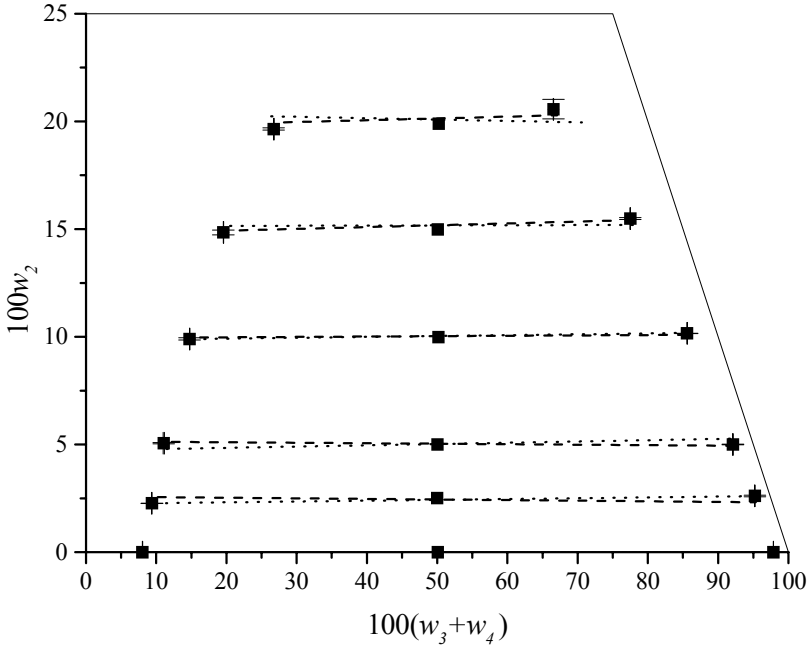

Figure 2.1. System of refined rice bran oil (1) + commercial oleic acid (2) + 6.38±0.02% aqueous solvent [ethanol (3) + water (4)] at 298.2±0.1 K: experimental (■);(- - -) NRTL; (·····) UNIQUAC

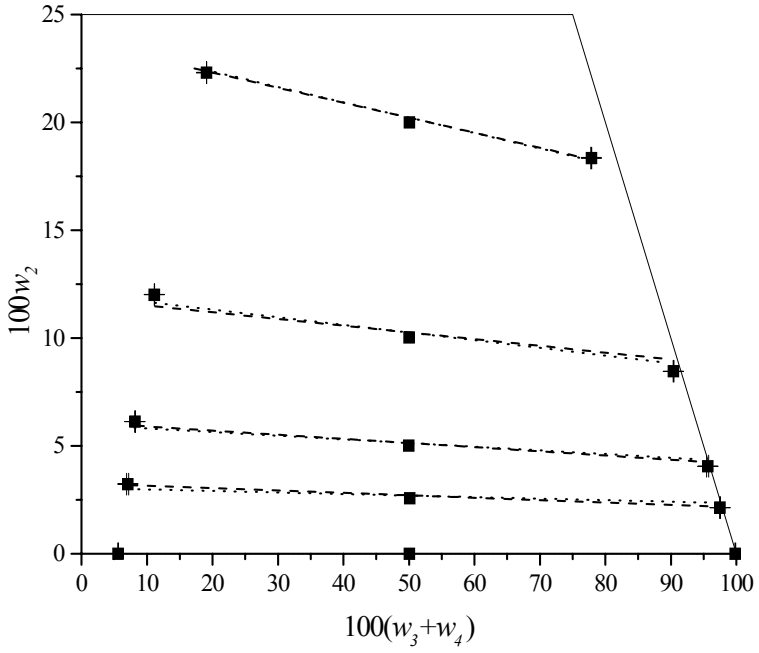

Figure 2.2. System of refined rice bran oil (1) + commercial oleic acid (2) + 12.41±0.01% aqueous solvent [ethanol (3) + water (4)] at 298.2±0.1 K: experimental (■);(- - -) NRTL; (·····) UNIQUAC

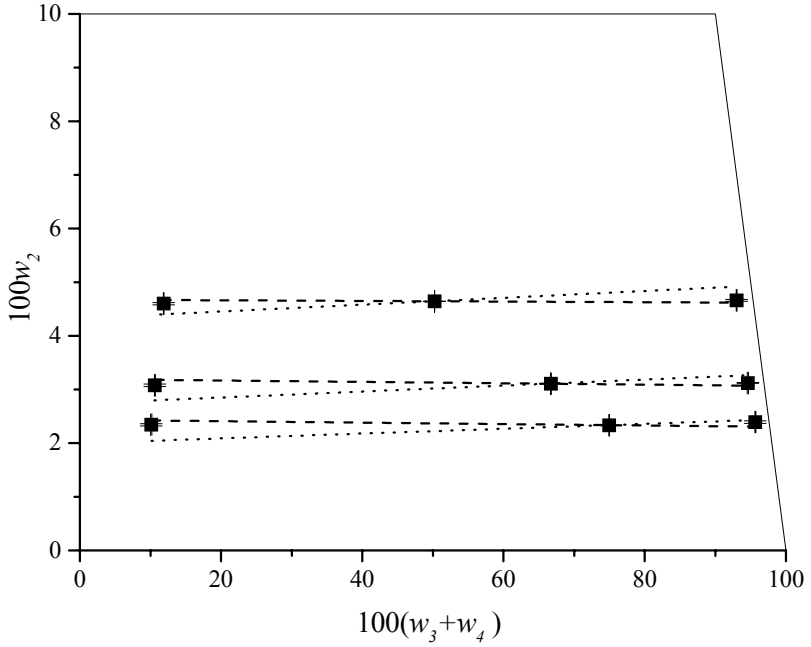

Figure 2.3. Prediction of the liquid-liquid equilibrium for the system of crude rice bran oil (1) + fatty acids (2) + 6.03±0.01% aqueous solvent [ethanol (3) + water (4)] at 298.2±0.1 K: experimental (■); (- - -) NRTL; (·····) UNIQUAC

2.4.1 Modeling

The experimental equilibrium data determined for the model systems were used to adjust the interaction parameters of the NRTL and UNIQUAC models. Mass fraction was used as concentration unit due to the large difference in molecular mass of the components in the system.[25,26] Gonçalves et al. [27] show the activity coefficient equations, expressed in mass fractions, according to the NRTL and UNIQUAC models.

The adjustments were made by treating the model system refined rice bran oil + commercial oleic acid + anhydrous ethanol as a pseudoternary one and the model systems rice bran oil + commercial oleic acid + ethanol + water as pseudoquaternary ones. The systems were considered as composed by a single triacylglycerol having the refined rice bran oil average molecular mass, a representative fatty acid with the molecular mass of the commercial oleic acid, ethanol and water.

The values of r_i' and q_i', volume and area parameters necessary for the UNIQUAC model, are given in Table 2.7. These parameters were calculated via eq 2.1, where x_j is the molar fraction of the fatty component i, $\overline{M}_i$ is the average molecular mass of the pseudocompounds rice oils or fatty acids, C is the number of different components in the pseudocompounds (oils or fatty acids), G is the total number of groups, and R_i and Q_i are Van der Waals parameters taken from Magnussen et al. [34].

$$r_i' = \frac{1}{\overline{M}_i}\sum_j^C x_j \sum_k^G v_k^{(i)} R_k; \qquad q_i' = \frac{1}{\overline{M}_i}\sum_j^C x_j \sum_k^G v_k^{(i)} Q_k \tag{2.1}$$

Table 2.7 - Parameters r_i' e q_i' for Refined rice bran oil, Commercial oleic acid, Ethanol, Water, Free Fatty acids in crude rice bran oil and Crude rice bran oil

Compound	r_i'	q_i'
Refined rice bran oil	0.044055	0.035722
Commercial oleic acid	0.045127	0.037140
Ethanol	0.055905	0.056177
Water	0.051069	0.077713
Crude rice bran oil	0.044069	0.035734
Free Fatty acids in crude rice bran oil	0.045029	0.037058

The interaction parameters estimation was based on the minimization of the objective function of composition (eq 2.2), following the procedure developed by Stragevitch and d'Avila[35].

$$S = \sum_{m}^{D}\sum_{n}^{N}\sum_{i}^{C-1}\left[\left(\frac{w_{inm}^{I,ex} - w_{inm}^{I,calc}}{\sigma_{w_{inm}^{I}}}\right)^2 + \left(\frac{w_{inm}^{II,ex} - w_{inm}^{II,calc}}{\sigma_{w_{inm}^{II}}}\right)^2\right] \qquad (2.2)$$

where D is the total number of groups of data, N is the total number of tie lines, and C is the total number of components or pseudocompounds in the group of data m. w is the mass fraction, the subscripts i, n and m are component, tie line and group number, respectively, and the superscripts I and II stand for oil and alcoholic phases, respectively; *ex* and *calc* refer to experimental and calculated concentrations. $\sigma_{w_{inm}^{I}}$ and $\sigma_{w_{inm}^{II}}$ are the standard deviations observed in the compositions of the two liquid phases.

The adjusted parameters of the NRTL and UNIQUAC models are shown in Tables 2.8 and 2.9, respectively. The interaction parameters between ethanol (3) and water (4) were taken from the previous study on phase equilibrium of the system corn oil + oleic acid + ethanol + water at 298.2±0.1 K.[27]

Table 2.8 - UNIQUAC Parameters for the System Refined rice bran oil (1) + Commercial oleic acid (2) + Ethanol (3) + Water (4) at 298.2±0.1 K

pair ij	A_{ij}/K	A_{ji}/K
12	252.66	-207.24
13	250.71	-56.595
14	7643.9	-134.68
23	67.641	-88.948
24	191.68	157.03
34	337.46	-279.92

Table 2.9 - NRTL Parameters for the System Refined rice bran oil (1) + Commercial oleic acid (2) + Ethanol (3) + Water (4) at 298.2±0.1 K

pair *ij*	A_{ij}/K	A_{ji}/K	α_{ij}
12	-290.55	-165.70	0.49698
13	873.64	1416.8	0.49874
14	-26.977	4624.6	0.16580
23	4800.0	-170.55	0.22957
24	1006.7	4210.6	0.10000
34	-10.984	-173.64	0.15018

The deviations between experimental and calculated compositions in both phases were calculated according to eq 2.3 and are shown in Table 2.10.

$$\Delta w = 100\sqrt{\frac{\sum_{n}^{N}\sum_{i}^{C}\left[\left(w_{i,n}^{I,ex}-w_{i,n}^{I,calc}\right)^2+\left(w_{i,n}^{II,ex}-w_{i,n}^{II,calc}\right)^2\right]}{2NC}} \qquad (2.3)$$

Figures 2.1 and 2.2 show that both thermodynamic models are able to describe with accuracy the phase compositions for the model systems investigated (Table 2.10).

Furthermore, it can be seen that the addition of water expands the region of phase splitting, allowing the refining of highly acidic crude rice bran oils by solvent extraction. *

Table 2.10 – Mean Deviations in Phase Compositions for the model systems and predictions

system	Δw (%)	
	NRTL	**UNIQUAC**
Refined rice oil + Oleic acid + Anhydrous Ethanol	0.43	0.46
Refined rice oil + Oleic acid + Aqueous Ethanol 2.40±0.02%	0.44	0.23
Refined rice oil + Oleic acid + Aqueous Ethanol 6.38±0.02%	0.87	0.90
Refined rice oil + Oleic acid + Aqueous Ethanol 10.59±0.04%	0.53	0.68
Refined rice oil + Oleic acid + Aqueous Ethanol 12.41±0.01%	0.71	0.76
Global Deviation of the correlation	0.68	0.71
Crude rice oil + Free fatty acids + Aqueous Ethanol 6.03±0.01%	0.35	0.45
Crude rice oil + Free fatty acids + Aqueous Ethanol 8.96±0.05%	0.38	0.54
Global Deviation of the prediction	0.37	0.49

Figure 2.4 shows the fatty acid distribution between the phases, indicating that the addition of water reduces the solvent capacity of extracting free fatty acids.

$$k_i = \frac{w_i^{II}}{w_i^{I}} \tag{2.4}$$

$$S = \frac{k_2}{k_1} \tag{2.5}$$

Experimental and estimated selectivities, calculated according to eqs 2.4 and 2.5, are shown in Figure 2.5.

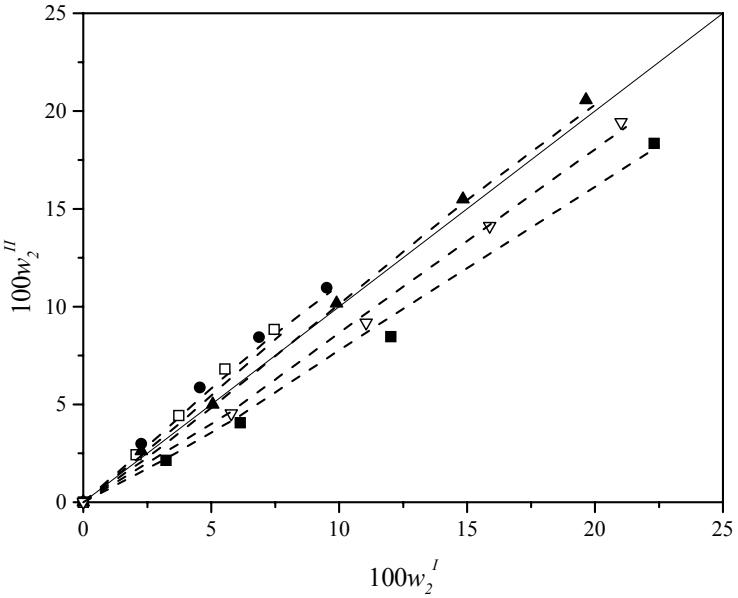

Figure 2.4. Distribution diagram at 298.2±0.1 K for systems of refined rice bran oil (1) + commercial oleic acid (2) + ethanol (3) + water (4): (●) anhydrous ethanol; (□)2.40±0.02% aqueous solvent; (▲)6.38±0.02% aqueous solvent; (▽)10.59±0.04% aqueous solvent; (■)12.41±0.01% aqueous solvent; (- - -) NRTL *

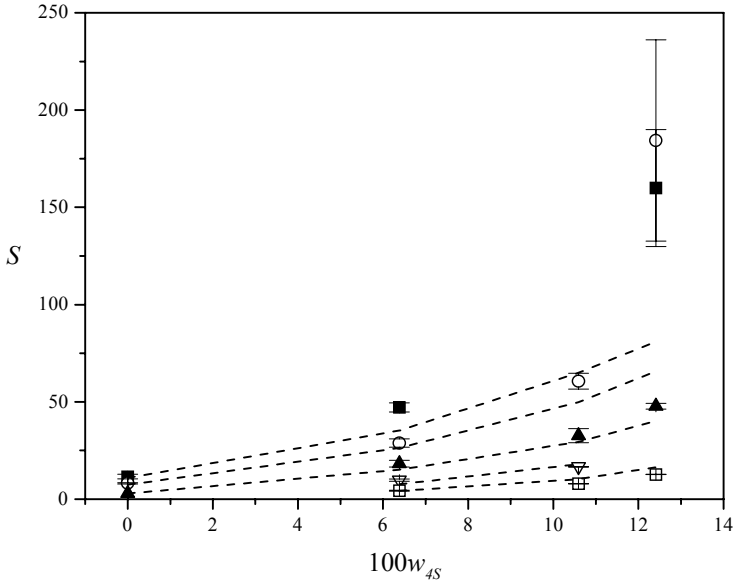

Figure 2.5. Selectivity diagram at 298.2±0.1 K for systems of refined rice bran oil (1) + commercial oleic acid (2) + ethanol (3) + water (4): (- - -) NRTL model; (■) 5 mass % FFA; (○) 10 mass % FFA; (▲) 20 mass % FFA; (▽) 30 mass % FFA; (□) 40 mass % FFA *

* Os resultados referentes ao modelo UNIQUAC estão mostrados no ANEXO A (Figuras A.2 e A.3)

This Figure shows that the addition of water to ethanol increases the solvent selectivity, reducing the loss of neutral oil. Moreover, these results show that NRTL model provides a good description of selectivity, except for the experimental points with 5 and 10 mass % of free fatty acids measured at 12 mass % water content in the solvent. For these systems, the oil concentration in the alcoholic phase is very low and exhibits a relatively high experimental uncertainty, which influences the uncertainties of the oil distribution coefficient and the experimental solvent selectivity. In addition, it should be observed that in this case the model does not describe the oil distribution coefficient and, in consequence, the solvent selectivity with accuracy.

2.4.2 Prediction of Liquid-Liquid Equilibrium

The adjusted parameters for the NRTL and UNIQUAC models were tested in the prediction of liquid-liquid equilibrium for the system crude rice bran oil + ethanol + water at (298.2±0.1) K. Liquid-liquid flash calculations for the estimation of phase compositions were performed on the basis of the overall experimental composition of the mixtures. The r_i' and q_i' values for crude rice bran oil and free fatty acids are given in Table 2.7. Since no interaction parameters involving γ-oryzanol were available, this compound was considered as part of the triacylglycerol fraction in the flash calculations.

The deviations between experimental and estimated compositions in both phases are calculated according to eq 2.3 and are shown in Table 2.10. Figure 2.3 shows the experimental points and the predicted tie lines for the system crude rice oil + free fatty acids + (6.03±0.01) mass % aqueous solvent.

Despite the differences in composition of the refined and crude rice bran oils and the approach incorporating the γ-oryzanol concentration in the triacylglycerol fraction, the parameters adjusted to the model systems allow a good prediction of phase equilibrium for the systems containing crude rice bran oil. This occurs for

both activity coefficient models; nevertheless the UNIQUAC equation overestimated the extraction of free fatty acids for both systems studied, resulting in somewhat higher values for the average percentual deviation (Table 2.10).

It is interesting to note that the prediction global deviations were lower than the corresponding values for the correlations. This occurs because, for the crude oil system, the highest fatty acid concentration in the overall composition was 4.67%. For the model systems this concentration was varied up to 20.0%, measuring tie lines near the plait point. In the case of the crude oil the highest possible fatty acid concentration in the overall mixture was limited by the original acid content in the oil. Correlation global deviations, calculated for tie-lines with up to 5.0% of fatty acids in the overall composition, were 0.33 and 0.40% for the NRTL and UNIQUAC models, respectively.

2.4.3 Partition of nutraceutical compounds

The partition of nutraceutical compounds, γ-oryzanol (5) and total tocopherols (6), naturally present in crude rice bran oil, was studied to evaluate the impact of solvent extraction upon the loss of such compounds. Figure 2.6 presents the distribution coefficients for γ-oryzanol (5) and total tocopherols (6), k_5 and k_6, respectively, calculated according to eq 2.4, for different water contents in solvent (w_{4S}). The equilibrium data for γ-oryzanol (5) and total tocopherols (6) are shown in Table 2.11. It can be observed that the distribution coefficient values were less than unity for both compounds. As the water content increases the distribution coefficients of both compounds decrease, minimizing the loss of nutraceutical components during the refining by liquid-liquid extraction.

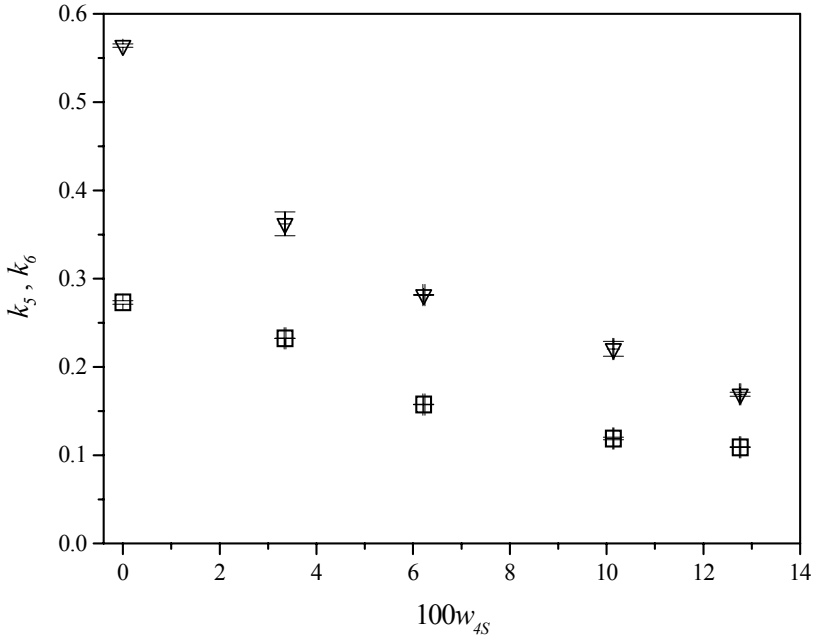

Figure 2.6. Partition of γ-oryzanol (□) and total tocopherols (∇) in several aqueous solvents at 298.2±0.1 K

Table 2.11 – γ-Oryzanol (5) and Total Tocopherol (6) Partition Data for Different Water Contents in the Solvent (w_{4S}) at (298.2±0.1)K

$100w_{4S}$[a]	alcohol phase (II)	oil phase (I)	alcohol phase (II)	oil phase (I)
	$100w_5$	$100w_5$	$100w_6$	$100w_6$
0	0.3510	1.2851	0.0225	0.0398
3.35 ± 0.02	0.3081	1.3246	0.0187	0.0401
6.22 ± 0.02	0.2343	1.4881	0.0132	0.0469
10.14 ± 0.01	0.1772	1.4902	0.0114	0.0516
12.76 ± 0.02	0.1615	1.4807	0.0084	0.0496

[a]$100w_{4S}$ = water mass percentage in the solvent

2.5. Conclusion

Phase equilibrium data for the system refined rice bran oil + commercial oleic acid + ethanol + water were experimentally determined at 298.2 K. The data set were correlated by the NRTL and UNIQUAC models and the adjusted parameters were used in the prediction of liquid-liquid equilibrium for systems

composed by crude rice bran oil + aqueous ethanol. Despite the difference in composition of the crude and refined rice bran oils, both molecular thermodynamic models allow a good prediction of phase equilibrium. The presence of water in the solvent minimizes the loss of neutral oil, making the extraction process more economic. Furthermore, the preliminary studies presented in this work on the minor component partition show that it is possible to refine rice bran oil by liquid-liquid extraction with a minimum loss of nutraceutical compounds

2.6. Literature cited

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2.7. Acknowledgements

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CAPÍTULO 3

Phase Equilibrium for the System Rice Bran Oil + Fatty Acids + Ethanol + Water + γ-Oryzanol + Tocols

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Keywords: Liquid-liquid equilibria; Experimental Data; Deacidification; Rice bran oil; Fatty Acids; Oryzanol; Tocopherols

Abstract

This work reports experimental equilibrium data for fatty systems containing rice bran oil, free fatty acids, ethanol, water, γ-oryzanol and tocols, at 298.2 K. Model fatty systems composed by refined rice bran oil, commercial oleic acid, γ-oryzanol, ethanol and water were used for adjusting NRTL and UNIQUAC interaction parameters between γ-oryzanol and the other pseudocompounds. UNIQUAC interaction parameters between tocols and the other pseudocomponents were determined assuming that the tocols are present at infinite dilution in the liquid-liquid equilibrium system. Despite the complexity of the systems studied, the interaction parameters obtained were capable of correctly predicting the equilibrium for systems containing Brazilian and Thai crude rice bran oils and aqueous ethanol.

3.1. Introduction

Vegetable oils may contain several minor compounds with antioxidant or other beneficial physiological properties [1]. Recently, rice bran oil has received attention because of its unique health benefits [2]. Such benefits may be attributed to its high level of unsaponifiable matter (up to 5 % in mass of crude oil) whose most important components are γ-oryzanol and tocopherols/tocotrienols, hereafter referred to as tocols [3,4].

γ-Oryzanol has been reported to lower serum cholesterol, to decrease early atherosclerosis, and to be appropriate for treatment of inflammatory processes. Initially it was thought to be a single compound but it is known now that γ-oryzanol is a mixture of ten components that consist of ferulic acid and triterpene derived compounds combined by an ester bond. Cycloartenyl ferulate, 24-

methylenecycloartanyl ferulate, and campesteryl ferulate are the three major components and account for 80 % of γ-oryzanol in rice bran oil [3,5].

In the tocols' family, the tocotrienol isomers differ from the tocopherol isomers only in having a side chain with three unsaturated bonds. Rice bran oil is relatively rich in this unsaturated form that possess antioxidant, antitumor and vitamin E activities [1,6,7].

The refining of rice bran oil, whose objective is the removal of impurities, also removes important nutraceutical compounds such as γ-oryzanol and tocols [8,9]. Rice bran oil is generally obtained from bran by extraction using hexane petroleum fractions as solvent [10-12]. Afterwards the crude oil is submitted to a series of purification steps such as solvent stripping, degumming, bleaching, deacidification and deodorization [13]. Deacidification is the most difficult step due to the high level of free fatty acids generally present in the crude rice bran oil. Oil deacidification can be accomplished by chemical or physical refining. In the case of high acidity oils these conventional processing techniques have some disadvantages, including the consumption of large quantities of water and chemicals, the generation of large quantities of waste, as well as the high energy requirement [13-15]. Furthermore, a significant portion of the rice bran oil nutraceutical compounds is lost during the conventional processes [8,9].

Deacidification by solvent extraction has been receiving attention due to its advantages in comparison to the physical and chemical refining. This method, based on the difference of the solubilities of free fatty acids and triacylglycerols in an appropriate solvent [16], can be conducted at low temperature and atmospheric pressure, reducing the energy consumption besides avoiding the formation of waste products.

Kim et al. and Kale et al. [17,18] studied the deacidification of crude rice bran oil by extraction with methanol. Bhatacharyya et al. and Shah and Venkatesan tested aqueous isopropanol as solvent in the deacidification of rice bran and

groundnut oils [19,20]. All studies showed a decreasing of the oil acidic value. Pina and Meirelles [21] studied the performance of a Perforated Rotating Disc Column in the continuous deacidification of corn oil, obtaining good results in relation to the extraction of free fatty acids and loss of neutral oil.

Information on phase equilibrium and physical properties are necessary for designing separation processes involving fatty systems. Zéberg-Mikkelsen and Stenby [22] predicted physical properties of saturated triacylglycerols using group contribution methods. Araújo and Meireles [23] tested the ability of the Peng-Robinson equation of state to predict the vapor-liquid equilibria of binary and ternary fatty systems. Equations of state are also used in the literature for calculating the equilibrium of alkane-vegetable oil mixtures [24,25].

In the case of liquid-liquid equilibrium for fatty systems, the information is relatively scarce in the literature. Liquid-liquid equilibrium data for fatty systems containing canola and corn oils have been reported by Batista et al. and Gonçalves et al., respectively [26,27]. These authors correlated the equilibrium data using NRTL and UNIQUAC models. In additon, Batista et al. [28] predicted the liquid-liquid equilibrium using the UNIFAC and ASOG models. In our previous work equilibrium data for the system rice bran oil + fatty acids + ethanol + water were reported [29]. In the present case, equilibrium data for the systems containing γ-oryzanol and tocols are determined and used for adjusting additional interaction parameters of the NRTL and UNIQUAC models. Despite the complexity of the systems studied, the interaction parameters obtained were capable of correctly predicting the equilibrium for systems containing crude rice bran oils of different sources and aqueous ethanol.

3.2. Experimental Section

3.2.1. Materials

The solvents used in this work were anhydrous ethanol, from Merck, with purity greater than 99.5%, and aqueous solvents with different water contents varying in the range of (3 to 20) mass %, prepared by the addition of deionized water to anhydrous ethanol.

All fatty reagents used in this study, commercial oleic acid (Merck), refined rice bran oil (Tio João, Brazil), Brazilian crude rice bran oil (kindly supplied by Helmut Tessmann, Brazil), and Thai crude rice bran oil (kindly supplied by Thaiedibleoil, Thailand) were analyzed by gas chromatography of fatty acid methyl esters to determine the fatty acid composition, according to the official method (1-62) of the AOCS [30]. The Brazilian crude rice bran oil was submitted to a prior degumming in the refinery (Helmut Tessmann, Brazil) and can be qualified as a semi-processed one. The fatty samples were prepared in the form of fatty acid methyl esters according to the official method (2-66) of the AOCS [31]. A HP5890 gas chromatograph with a flame ionization detector was used under the following experimental conditions: fused silica column of cyanopropylsiloxane 0.25 μm, 60 m x 0.32 mm id.; hydrogen as the carrier gas at a rate of 2.5 ml/min; injection temperature of 548.2 K; column temperature of (448.2 to 498.2) K (rate of 1.3 K/min); detection temperature of 578.2 K.

The fatty acid methyl esters were identified by comparison with external standards purchased from Nu Check Inc. (Elysian, IL). The quantification was accomplished by internal normalization. All the oils used in this work were analyzed by spectrophotometry, using a UV-vis dual beam spectrophotometer (Perkin Elmer, model Lambda 40), to determine the presence and concentration of nutraceutical compounds. The concentration of γ-oryzanol was determined at 314.5 nm, as suggested by Seetharamaiah and Prabakar [32], using heptane (UV-Fluo, Carlo Erba) as solvent and γ-oryzanol, purity greater than 99%, (kindly supplied by

Tsuno Rice Fine Chemicals Co., Japan), as standard. The quantification of tocols (tocopherols and tocotrienols) was determined at 520 nm according to the methodology developed by Emmerie-Engel [33]. α-Tocopherol, purity greater than 99% (Sigma), was used as standard and toluene (Em Science) as solvent.

3.2.2. Experimental Procedure

Six model fatty systems (A to F) containing free fatty acids, triacylglycerols and γ-oryzanol were prepared by the addition of known quantities of commercial oleic acid and γ-oryzanol (from Tsuno Rice Fine Chemicals Co., Japan) to refined rice bran oil. In the systems A, C and E, approximately 1.5 mass % of γ-oryzanol (γ-oryzanol mass percentage in the oil - $100w_5^{oil}$) was incorporated in the refined rice oil and the concentration of free fatty acids was varied. In the case of systems B, D and F fixed quantities of free fatty acids, 10, 20 and 30 mass %, respectively, were added to the refined oil and the concentration of γ-oryzanol was varied up to 2 mass %.

The model fatty systems were mixed with the ethanolic solvents, anhydrous ethanol for A and B systems, aqueous ethanol with 6 mass % of water for C and D systems and aqueous ethanol with 12 mass % of water for E and F systems, in the mass ratio oil:solvent 1:1, at (298.2±0.1) K. The experimental data measured for the model systems were used to adjust NRTL and UNIQUAC interaction parameters between γ-oryzanol and the other components (rice bran oil, commercial oleic acid, ethanol and water). The parameters concerning the interaction between these other components with each other were taken from our prior work [29].

To test the prediction capability of the adjusted NRTL and UNIQUAC parameters, further sets of experiments using model and real fatty systems were performed to measure only the partition coefficients of γ-oryzanol. Three model systems were constructed by the addition of known quantities of commercial oleic acid (10, 25 and 30 mass %) and different contents of γ-oryzanol ($100w_5^{oil}$ = 0 to 2

mass %), to refined rice bran oil. These model fatty systems were mixed with aqueous ethanol with 6 mass % of water, in the mass ratio oil:solvent 1:1, at (298.2±0.1) K. In addition, real systems were constructed to evaluate the distribution of γ-oryzanol, naturally present in crude oil, between the alcoholic and oil phases. These experiments were performed mixing Brazilian crude rice bran oil with different aqueous solvents (water mass percentage in the solvent - $100w_{4S}$ - varying from 0 to 12 mass %), in the mass ratio oil:solvent 1:1, at (298.2±0.1) K. The concentration of γ-oryzanol was measured according to the procedure described above.

Other aim of this work was the determination of interaction parameters between tocols and the other components of the fatty system (rice bran oil, commercial oleic acid, ethanol, water and γ-oryzanol). For this purpose 28 tie-lines were prepared mixing the Brazilian crude rice bran oil with different ethanolic solvents ($100w_{4S}$ varying from 0 to 20 mass %), in different mass ratios oil:solvent, at (298.2±0.1) K. In these tests only the concentration of tocols in both phases were measured according to the Emmerie-Engel methodology [33].

Finally, the sets of parameters obtained were used to predict the liquid-liquid equilibrium of systems composed by a crude rice bran oil from Thailand plus a ethanolic solvent with 7.45 mass % of water. Six tie lines with different oil:solvent mass ratios were prepared and the concentration of all components were measured in both alcoholic and oil phases.

In the case of all experiments the liquid-liquid equilibrium data were determined using polypropylene centrifuge tubes (50 mL) (Corning Inc.). The components were weighed on an analytical balance Sartorius model A200S, accurate to 0.0001 g. The tubes were vigorously stirred for at least 15 min, sonicated for 5 min at 40 kHz (Unique, model T1440), centrifuged for 10 min at 4500 *g* (Centrifuge Jouan, model BR4i) and left to rest for 2 h in a thermostatic bath at (298.2±0.1) K (Cole Parmer, model 12101-05).

After this treatment, the two phases became clear, with a well-defined interface, and the composition of both phases was measured. The concentration of free fatty acids was determined by titration (official method 2201 of the IUPAC [34]) with an automatic burette (Metrohm, model Dosimat 715). The total solvent concentration was determined by evaporation at 338.2 K in a vacuum oven (Napco, model 5831). The water concentration was determined by Karl Fischer titration, according to AOCS method Ca 23-55 [30] with a KF Titrino (Metrohm, model 701). Concentrations of γ-oryzanol and tocols were measured according to the procedures described above. The triacylglycerol concentration was determined by difference.

In this work all measurements were performed at least in triplicate. The uncertainties of the concentrations varied within the following ranges: (0.01 to 0.45) mass % for rice oil, (0.01 to 0.28) mass % for fatty acids, (0.01 to 0.40) mass % for ethanol, (0.01 to 0.04) mass % for water, (0.01 to 0.13) mass % for γ-oryzanol and, (0.0005 to 0.01) mass % for tocols, being the lowest figures obtained for the lowest concentrations.

3.2.3. Modeling Approach

In our prior work [29] the interaction parameters of the NRTL and UNIQUAC models between rice bran oil, commercial oleic acid, ethanol and water were reported. The adjustments were made treating the model system refined rice bran oil + commercial oleic acid + anhydrous ethanol as a pseudoternary one and the model systems refined rice bran oil + commercial oleic acid + ethanol + water as pseudoquaternary ones.

In the present work the experimental data measured for the model systems were used to adjust NRTL and UNIQUAC interaction parameters between γ-oryzanol and the other components of system (rice bran oil, commercial oleic acid, ethanol and water). The parameters concerning the interaction between these other

components with each other were taken from our prior work [29]. In this case, the model systems refined rice bran oil + commercial oleic acid + anhydrous ethanol + γ-oryzanol (systems A and B) were considered as pseudoquaternary ones and the model systems rice bran oil + commercial oleic acid + ethanol + water + γ-oryzanol (systems C to F) as pseudoquinary ones.

For the adjustment process the rice bran oil was treated as a single triacylglycerol with the oil's average molar mass. The same approach was extended to the commercial oleic acid. This approach assumes that the different triacylglycerols present in the rice bran oil behave in a very similar way in the liquid-liquid system under analysis. In this case such components can be adequately replaced by a pseudocompound having the corresponding average physical-chemical properties. The same hypothesis is assumed in relation to the fatty acid mixture, the γ-oryzanol compound and the tocols' family. Such a hypothesis will be tested by the adjustment of parameters to the model systems and the subsequent use of these parameters in the equilibrium prediction for systems containing Brazilian and Thai crude rice bran oils.

The values of r_i' and q_i', volume and area parameters necessary for the UNIQUAC model, were calculated according to eq. 3.1, where x_j is the molar fraction of the component present in each pseudocompound i, M_i is the average molar mass of the pseudocompounds rice oils, fatty acids, γ-oryzanol or tocols, C is the number of different components in the pseudocompounds, G is the total number of groups, $v_k^{(i)}$ is the number of group k in molecule i and R_i and Q_i are Van der Waals parameters taken from Magnussen et al [35].

$$r_i' = \frac{1}{M_i}\sum_j^C x_j \sum_k^G v_k^{(i)} R_k; \qquad q_i' = \frac{1}{M_i}\sum_j^C x_j \sum_k^G v_k^{(i)} Q_k \qquad [3.1]$$

The estimation of interaction parameters between γ-oryzanol and the other components of the fatty system was based on the minimization of the objective

function of composition, OF(*w*), (eq. 3.2), following the procedure developed by Stragevitch and d'Avila [36].

$$OF(w) = \sum_{m}^{D}\sum_{n}^{N}\sum_{i}^{K-1}\left[\left(\frac{w_{inm}^{OP,ex} - w_{inm}^{OP,calc}}{\sigma_{w_{inm}^{OP}}}\right)^2 + \left(\frac{w_{inm}^{AP,ex} - w_{inm}^{AP,calc}}{\sigma_{w_{inm}^{AP}}}\right)^2\right] \qquad [3.2]$$

where *D* is the total number of groups of data, *N* is the total number of tie lines, and *K* is the total number of components or pseudocompounds in the group of data *m*, *w* is the mass fraction, the subscripts *i*, *n* and *m* are pseudocompound, tie line and group number, respectively, and the superscripts *OP* and *AP* stand for oil and alcoholic phases, respectively; *ex* and *calc* refer to experimental and calculated concentrations. $\sigma_{w_{inm}^{OP}}$ and $\sigma_{w_{inm}^{AP}}$ are the standard deviations observed in the compositions of the two liquid phases.

In the above equation the objective function was defined using mass fraction as unity of concentration, according to the unities employed in the experimental measurements. The UNIQUAC and NRTL equations were also appropriately expressed in mass fractions unities (see Appendix A) and this is also the reason to include the pseudocompound average molar mass *Mi* in equation 3.1 above.

UNIQUAC interaction parameters between tocols and the other pseudocomponents of the fatty system (triacylglycerols, free fatty acids, ethanol, water and γ-oryzanol) were also determined. It was assumed that the tocols are present at infinite (∞) dilution in the liquid-liquid equilibrium system.

In this case the pseudocompound distribution coefficient, calculated according to eq. 3.3 below, can be approached by the distribution coefficient at infinite dilution k_i^{∞} . Using the iso-activity criterion this distribution coefficient for tocols, k_6^{∞} , can be calculated by eq. 3.4:

$$k_i = w_i^{AP} / w_i^{OP} \qquad [3.3]$$

$$k_6^{\infty} = \left(\hat{\gamma}_6^{OP}\right)^{\infty} / \left(\hat{\gamma}_6^{AP}\right)^{\infty} \qquad [3.4]$$

As in the present case the iso-activity criterion was expressed in terms of mass fraction, the mass fraction-scale activity coefficient $\hat{\gamma}_i$ should be related to the UNIQUAC activity coefficient γ_i (see Appendix A) by the following equation:

$$\hat{\gamma}_i = \gamma_i \Big/ M_i \left(\sum_{j=1}^{K} w_j / M_j \right) \qquad [3.5]$$

To calculate γ_6^{∞} , the system phase equilibrium was considered established by the knowledge of the interaction parameters between the major compounds of the pseudoquinary fatty system (triacylglycerols, free fatty acids, ethanol, water and γ-oryzanol). These parameters were used to perform liquid-liquid flash calculations for the estimation of phase compositions on the basis of the overall experimental composition of the mixtures. The infinite dilution activity coefficient (γ_i^{∞}) is obtained applying the limit in the UNIQUAC model equation, keeping constant the mass fractions of the others mixture components and making the minor pseudocompound concentration tends to zero. *

For the adjustment of interaction parameters between tocols and other components the estimation was based on the minimization of the distribution coefficient objective function, eq. 3.6 below, following the procedure developed by Pessôa Filho [37]. In eq. 3.6 the additional term is a penalty function suggested by Kang and Sandler [38] and used to preclude interaction parameters with too large absolute values:

$$OF(k_6) = \left(\sum_{n=1}^{N} \left(k_6^{ex} - k_6^{calc} \right)^2 \Big/ N \right)^{1/2} + Q \sum_{l=1}^{L} (p_l^2) / L \qquad [3.6]$$

where n is the tie line index, N is the total number of tie lines, k_6 is the tocols' distribution coefficient, *ex* and *calc* refer to experimental and calculated values, Q is a small value that not alters significantly the function residue, l is the UNIQUAC parameter index, L is the total number of adjustable parameters and p_l is the UNIQUAC parameter.

Further sets of experimental data were obtained with the aim of testing the prediction capability of the models for systems containing rice bran oils submitted to different pretreatments and from different countries. Systems containing refined rice bran oil and crude rice bran oil acquired in Brazil were used to test the parameters related to γ-oryzanol. A further system containing crude rice bran oil, not degummed, kindly supplied by a Thai refinery, was used to evaluate the prediction capability of the adjusted parameters concerning the interaction between all components of the fatty system.

3.3. Results

The fatty acid composition of Thai crude rice bran oil is compared with the fatty acid compositions of Brazilian refined and crude rice bran oils in Table 3.1. From these fatty acid compositions it was possible to determine the probable triacylglycerol composition of the refined and crude rice bran oils by using the procedure suggested by Antoniosi Filho et al [39].

Table 3.1 – Fatty Acid Composition of Brazilian Refined and Crude Rice Bran Oils and Thai Crude Rice Bran Oil

Symbol	Fatty Acid		M[b] g·mol^{-1}	Brazilian				Thai	
				Refined [c]		Crude [c]		Crude	
				Mole %	Mass %	Mole %	Mass %	Mole %	Mass %
M	Miristic	C14:0[a]	228.38	1.16	0.96			0.60	0.49
P	Palmitic	C16:0	256.43	19.57	18.17	21.44	19.91	21.99	20.43
Po	Palmitoleic	C16:1	254.42	0.66	0.61				
S	Stearic	C18:0	284.49	1.50	1.54	1.79	1.84	2.31	2.38
O	Oleic	C18:1	282.47	37.66	38.50	39.11	40.01	39.77	40.69
Li	Linoleic	C18:2	280.45	35.08	35.61	35.78	36.34	32.73	33.24
Le	Linolenic	C18:3	278.44	2.65	2.67	1.88	1.90	1.43	1.44
A	Arachidic	C20:0	312.54	1.57	1.78			0.76	0.86
Ga	Gadoleic	C20:1	310.52	0.15	0.16			0.41	0.47

[a] In C*x:y*. *x*=number of carbons and *y*=number of double bonds;
[b] *M*=molar mass;
[c] data taken from [29].

These results make possible to calculate the average molar masses of free fatty acids in Brazilian and Thai crude rice oils, of refined rice oil, and of Brazilian and Thai crude rice bran oils. The molar masses values obtained as well as volume and area parameters values are presented in Table 3.2. The values for the commercial oleic acid as well as its composition and the fatty acid compositions of the refined and crude Brazilian rice bran oils were taken from [29]. For the nutraceutical compounds the average molar masses were calculated as the weighed media of the major components molar masses present in the crude rice bran oil. In the case of γ-oryzanol the three major components (cycloartenyl ferulate, 24-methylenecycloartanyl ferulate and campesteryl ferulate) were considered, according to results reported by Xu and Godber [5]. Tocols' average molar mass was calculated using data published by Rogers et al. [40]. These authors show that β/γ tocotrienols and β/γ tocopherols are the major isomers present in crude rice bran oil.

Table 3.2 - Average molar masses *M*, Structural parameters r_i' e q_i' and Iodine Values (*IV*) *

Compound	$\underline{M}^b$ g·mol^{-1}	r_i'	q_i'	*IV*
Brazilian refined rice bran oil [a]	867.78	0.044090	0.035751	101.92
Brazilian crude rice bran oil [a]	866.50	0.044084	0.035746	102.32
Thai crude rice bran oil	865.56	0.044114	0.035777	95.73
Free fatty acids in Brazilian crude oil	276.13	0.045028	0.037058	
Free fatty acids in Thai crude rice oil	276.09	0.045009	0.037121	
Commercial oleic acid [a]	278.96	0.045127	0.037140	
γ-Oryzanol	602.10	0.040718	0.031052	
Tocols	412.14	0.043440	0.034340	
Ethanol	46.07	0.055905	0.056177	
Water	18.02	0.051069	0.077713	

[a] data taken from [29];
[b] oils average molar masses calculated using the procedure suggested by Antoniosi Filho et al [39].

It can be seen in Table 3.2 that the small differences of r_i' and q_i' parameters for refined and crude oils and for commercial oleic acid and free fatty acids in crude oils reflect the small differences in composition of the pseudocompounds. In addition, Table 3.2 furnishes the Iodine Values (*IV*) for Brazilian refined and crude rice bran oils and Thai crude rice bran oil. The Iodine Values express the unsaturation level of fatty compounds and were calculated directly from fatty acid compositions (Table 3.1) according to method Cd 1c-85 AOCS [30]. These values show that the Brazilian oils are more unsaturated than Thai crude rice bran oil. It should be noted that the Thai oil has higher levels of palmitic and stearic acids and lower levels of linoleic and linolenic acids. These last two acids are polyunsaturated ones, having two or three double bonds, respectively.

Uma correlação entre massas molares e coeficientes de partição pode ser vista no ANEXO A (Figura A.4)

Refined rice bran oil had a residual acidity of 0.07 mass %, expressed as oleic acid. The Brazilian crude rice oil presented an acidic value of (9.34±0.01) mass %, and the Thai crude rice bran oil presented an acidic value of (7.92±0.05) mass %, both expressed as oleic acid.

Brazilian crude rice bran oil presented (1.72±0.05) mass % of γ-oryzanol and (622.3±4.0) ppm of tocols, and Thai crude rice oil contained (1.87±0.05) mass % of γ-oryzanol and (1026.9±85.3) ppm of tocols. γ-Oryzanol concentration in refined rice oil was (0.12±0.01) mass % and tocols were not detected in this oil.

Tables 3.3, 3.4 and 3.5 present the overall experimental composition of the mixtures and the corresponding tie-lines for the pseudoquaternary and pseudoquinary model systems composed by refined rice bran oil + commercial oleic acid + ethanol + γ-oryzanol (systems A and B) and refined rice bran oil + commercial oleic acid + ethanol + water + γ-oryzanol (systems C to F), respectively.

Table 3.3 - Liquid-liquid Equilibrium Data for the System Refined rice bran oil (1) + Commercial oleic acid (2) + Anhydrous Ethanol (3) + γ-oryzanol (5) at (298.2±0.1) K

System	Overall Composition (OC)				Alcohol Phase (AP)				Oil Phase (OP)			
	$100w_1$	$100w_2$	$100w_3$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_5$
A	49.16	0.00	50.00	0.76	7.95	0.00	91.76	0.29	82.27	0.00	16.41	1.32
	47.25	2.02	50.01	0.73	9.51	2.22	87.95	0.33	78.74	1.78	18.18	1.29
	45.27	4.02	50.02	0.70	11.63	4.38	83.62	0.37	74.30	3.60	20.93	1.18
	43.30	6.03	50.00	0.67	13.39	6.60	79.59	0.42	69.06	5.60	24.26	1.07
	41.37	8.01	49.97	0.64	18.75	8.57	72.22	0.45	62.66	7.61	28.77	0.95
	39.36	10.03	50.01	0.61	26.62	10.40	62.43	0.54	53.31	9.36	36.45	0.87
B	45.00	5.01	49.99	0.00	12.47	5.86	81.67	0.00	74.14	4.56	21.30	0.00
	44.89	5.00	49.91	0.20	12.51	5.84	81.53	0.11	73.67	4.55	21.44	0.35
	44.75	4.98	49.87	0.40	12.60	5.83	81.37	0.21	73.24	4.53	21.67	0.55
	44.71	4.98	49.72	0.59	12.46	5.82	81.42	0.30	72.62	4.53	21.96	0.88
	44.59	4.97	49.66	0.79	12.44	5.81	81.34	0.41	72.01	4.52	22.24	1.23
	44.52	4.96	49.53	0.99	12.58	5.80	81.09	0.54	71.68	4.51	22.28	1.53
	44.44	4.95	49.42	1.19	12.47	5.79	81.11	0.64	71.18	4.50	22.48	1.84
	44.34	4.94	49.32	1.40	12.34	5.77	81.13	0.75	70.81	4.49	22.60	2.10
	44.28	4.93	49.21	1.58	12.35	5.77	81.04	0.85	70.52	4.48	22.65	2.35
	44.21	4.92	49.11	1.76	12.55	5.76	80.66	1.03	69.87	4.48	22.93	2.73
	44.11	4.91	49.03	1.95	12.44	5.81	81.34	0.41	72.01	4.52	22.24	1.23

A – System with around 1.5 mass % of γ-oryzanol in Rice Bran Oil
B – System with around 10 mass % of free fatty acids in Rice Bran Oil

Table 3.4 - Liquid-liquid Equilibrium Data for the System Refined rice bran oil (1) + Commercial oleic acid (2) + aqueous solvent $100w_{4S}$ = 6 mass % [ethanol (3) + water (4)] + γ-oryzanol (5) at (298.2±0.1) K

System	Overall Composition (OC)					Alcohol Phase (AP)					Oil Phase (OP)				
	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$
C	49.22	0.00	47.05	2.97	0.76	2.20	0.00	90.80	6.86	0.14	91.24	0.00	6.59	0.67	1.50
	46.73	2.55	47.03	2.97	0.72	2.33	2.73	88.23	6.54	0.17	87.59	2.35	8.08	0.62	1.36
	44.31	5.00	47.03	2.97	0.68	2.80	5.25	85.38	6.37	0.19	83.63	4.87	9.43	0.83	1.24
	39.38	10.01	47.03	2.97	0.61	4.61	10.16	79.06	5.92	0.25	74.54	9.78	13.56	0.99	1.12
	34.44	15.02	47.03	2.97	0.53	7.29	15.75	71.08	5.57	0.31	65.30	14.46	18.09	1.22	0.93
	29.53	20.00	47.04	2.97	0.46	14.83	20.29	59.75	4.72	0.41	51.51	19.36	26.64	1.79	0.41
D	39.98	10.01	46.86	3.15	0.00	4.76	10.21	79.40	5.63	0.00	74.43	9.84	14.85	0.88	0.00
	39.87	9.98	46.71	3.14	0.31	4.60	10.31	79.30	5.67	0.11	73.97	9.57	15.12	0.84	0.50
	39.46	9.88	46.92	3.15	0.60	4.56	10.18	79.58	5.47	0.21	73.57	9.49	15.08	0.89	0.97
	39.42	9.87	46.68	3.14	0.89	4.32	10.30	79.32	5.75	0.32	72.93	9.48	15.33	0.72	1.54
	39.42	9.87	46.41	3.12	1.17	4.30	10.31	79.29	5.67	0.42	72.29	9.32	15.29	1.07	2.03
	39.41	9.87	46.14	3.10	1.48	4.29	10.22	79.35	5.59	0.54	71.18	9.68	15.71	0.87	2.55
	39.03	9.77	47.04	3.11	1.77	3.81	10.40	79.89	5.19	0.71	71.37	9.19	15.75	0.84	2.85

C – System with around 1.5 mass % of γ-oryzanol in Rice Bran Oil

D – System with around 20 mass % of free fatty acids in Rice Bran Oil

Table 3.5 - Liquid-liquid Equilibrium Data for the System Refined rice bran oil (1) + Commercial oleic acid (2) + aqueous solvent $100w_{4S}$ = 12 mass % [ethanol (3) + water (4)] + γ-oryzanol (5) at (298.2±0.1) K

System	Overall Composition (OC)					Alcohol Phase (AP)					Oil Phase (OP)				
	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$
E	49.24	0.00	43.49	6.45	0.81	0.87	0.00	86.77	12.29	0.07	92.64	0.00	5.27	0.58	1.51
	46.54	2.52	43.69	6.48	0.77	0.84	2.21	84.19	12.69	0.08	88.54	3.03	6.26	0.75	1.42
	44.24	5.01	43.56	6.46	0.73	0.80	4.30	82.70	12.11	0.09	83.88	6.02	7.78	0.99	1.33
	39.30	10.04	43.55	6.46	0.65	1.54	8.56	77.78	12.00	0.12	75.27	11.67	10.93	0.94	1.19
	34.31	14.99	43.65	6.48	0.57	2.65	13.36	71.83	12.00	0.16	66.45	17.13	13.81	1.63	0.99
	29.54	19.99	43.53	6.46	0.49	4.72	18.81	65.66	10.58	0.22	56.88	22.21	18.04	2.06	0.81
F	34.85	14.95	44.12	6.08	0.00	2.95	12.96	73.29	10.80	0.00	67.43	17.09	14.19	1.28	0.00
	34.93	14.99	43.75	6.03	0.30	2.74	13.06	73.16	10.94	0.097	66.83	17.04	14.23	1.36	0.54
	34.38	14.75	44.15	6.09	0.63	2.72	12.94	73.08	11.08	0.19	66.90	16.50	14.04	1.46	1.10
	34.49	14.80	43.74	6.03	0.93	1.92	13.63	73.34	10.83	0.28	66.71	15.79	14.41	1.38	1.71
	34.33	14.73	43.72	6.03	1.18	2.29	13.23	72.99	11.14	0.35	65.65	16.35	14.64	1.32	2.04

E – System with around 1.6 mass % of γ-oryzanol in Rice Bran Oil

F – System with around 30 mass % of free fatty acids in Rice Bran Oil

Table 3.6 presents the parameters of the UNIQUAC and NRTL models adjusted for the interactions between γ-oryzanol and the others pseudocomponents. The deviations between experimental and calculated composition in both phases were calculated according to eq 3.7 and are shown in Table 3.7.

$$\Delta w = \left(\frac{\sum_{n}^{N} \sum_{i}^{K} \left[\left(w_{in}^{OP,ex} - w_{in}^{OP,calc} \right)^2 + \left(w_{in}^{AP,ex} - w_{in}^{AP,calc} \right)^2 \right]}{2NK} \right)^{1/2} \qquad [3.7]$$

Table 3.6 - UNIQUAC and NRTL Parameters for the Refined rice bran oil (1) + Commercial oleic acid (2) + Ethanol (3) + Water (4) + γ-Oryzanol (5) and UNIQUAC Parameters for Crude rice bran oil (1) + Free fatty acids (2) + Ethanol (3) + Water (4) + γ-Oryzanol (5) + Tocols (6), at (298.2±0.1) K

pair ij	Thermodynamic Model				
	UNIQUAC		NRTL		
	A_{ij}/K	A_{ji}/K	A_{ij}/K	A_{ji}/K	α_{ij}
15	1.1884	-176.26	-6963.3	15.569	0.48735
25	2151.6	-134.91	1169.1	315.40	0.70000
35	-501.84	4316.1	-5766.4	889.27	0.33762
45	1250.9	3890.2	6133.4	3169.1	0.39181

	UNIQUAC	
	A_{ij}/K	A_{ji}/K
16	-20.938	-188.60
26	139.22	1151.9
36	654.53	-479.15
46	-1027.1	775.21
56	34.343	-274.83

In Figure 3.1 the equilibrium data for the model system C are shown. In this figure ethanol + water is considered as a mixed solvent and γ-oryzanol was incorporated in the oil fraction. A good alignment can be observed between the experimental data, relative to both overall and phase concentrations.

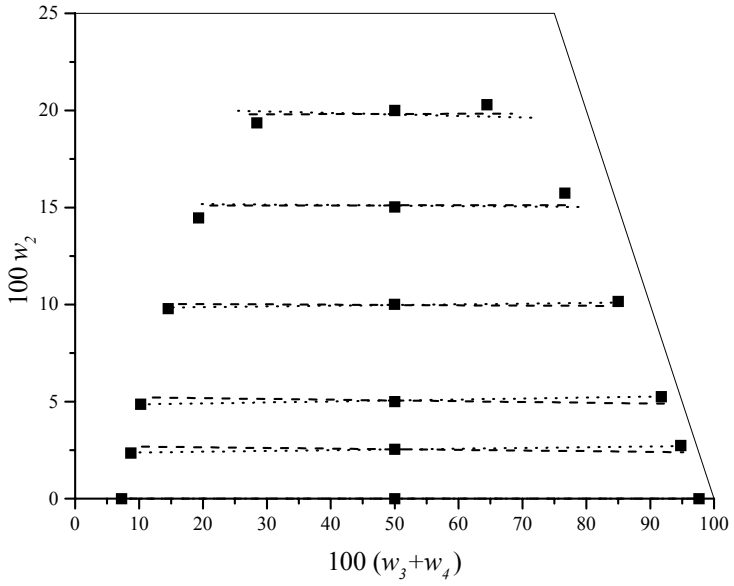

Figure 3.1. System of refined rice bran oil (1) + commercial oleic acid (2) + aqueous solvent $100w_{4S}$ = 6 mass % [ethanol (3) + water (4)] + $100w_5^{oil}$ = 1.5 mass %, at (298.2±0.1) K (C): experimental (■);(- - -) NRTL; (·····) UNIQUAC

Tie lines based on the experimental data were determined by linear regression of each corresponding set of overall, oil and alcoholic phase concentrations. Correlation coefficients higher than 99.3% were obtained for all tie lines, indicating a low error in the experimental determination of the tie line compositions. This figure shows that both thermodynamic models are able to describe with accuracy the phase compositions for the model systems investigated (Table 3.7).

Figure 3.2 shows the partition coefficient of γ-oryzanol (k_5) as a function of acidity level in the oil ($100w_2^{oil}$), for systems A, C and E. It can be observed that a higher free fatty acid content in the oil increases the partition coefficient of γ-oryzanol. This can be attributed to the increase of the oil and solvent mutual solubility at higher free fatty acid concentration. The oil carried out to the alcoholic phase drags a larger part of γ-oryzanol. On the other hand, the addition of water to the solvent decreases γ-oryzanol partition coefficient and consequently reduces the loss of this nutraceutical compound during the solvent extraction.

Table 3.7 – Global Deviations for Correlations and Predictions

Global Deviations	100 Δw [a]		100 Δw_5 [b]		Δk_5 [c]		Δk_6 [d]
	UNIQUAC	NRTL	UNIQUAC	NRTL	UNIQUAC	NRTL	UNIQUAC
Correlation							
Model System	0.86	0.76	0.055	0.049	0.352	0.322	
Real System							0.11
Prediction							
Model System			0.15	1.04			
Real System			0.064	0.66			
Prediction							
Thai crude rice oil System	0.81 [e]	0.56 [e]			0.060 [f]	0.097	0.21 [f]

[a] 100Δw: deviations in phase compositions;
[b] 100Δw_5: deviations in γ-oryzanol composition;
[c] Δk_5: deviations in γ-oryzanol distribution coefficient;
[d] Δk_6: deviations in tocols distribution coefficient;
[e] flash calculations considering 5 components in the system (related to Figure 3.7);
[f] related to Figure 3.8.

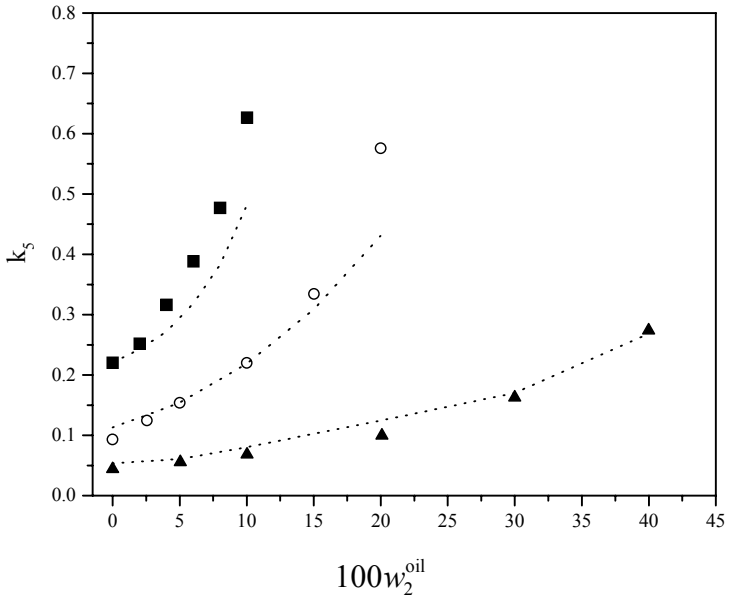

Figure 3.2. Distribution coefficient of γ-oryzanol for model fatty systems with around $100w_5^{oil}$ = 1.5 mass %, at (298.2±0.1) K: (■) $100w_{4S}$ = 0 mass % (A); (○) $100w_{4S}$ = 6 mass % (C); (▲) $100w_{4S}$ = 12 mass % (E); (·····) UNIQUAC *

The UNIQUAC model is able to describe satisfactorily the partition of γ-oryzanol as is shown in Figure 3.2. The slightly higher value of phase composition global deviations (Δw) for the UNIQUAC model, observed in Table 3.7, may be related to the slightly worse description of the triacylglycerol distribution coefficient in case of specific tie-lines in the A and C systems (see Figure 3.3).

The values of Δw_5, global deviation between experimental and estimated compositions of γ-oryzanol in both phases (calculated according to eq. 3.7 where, in this case, K is only γ-oryzanol, i=5), and Δk_5, deviation between experimental and calculated γ-oryzanol distribution coefficient (calculated as shown in eq. 3.8 bellow) are also presented in Table 3.7. It can be observed that these values are slightly higher for the UNIQUAC model than for the NRTL model.

$$\Delta k_i = \left(\sum_{n=1}^{N} \left(k_{in}^{ex} - k_{in}^{calc} \right)^2 \right)^{1/2} \qquad [3.8]$$

* O desempenho do modelo NRTL pode ser visto no ANEXO A (Figura A.5)

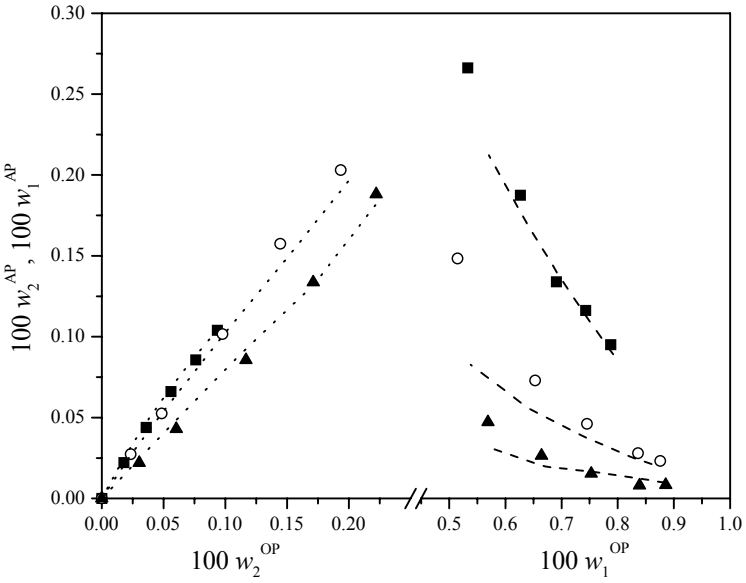

Figure 3.3. Distribution diagram at (298.2±0.1) K for model fatty systems: (■) $100w_{4S}$ = 0 mass % (A); (○) $100w_{4S}$ = 6 mass % (C); (▲) $100w_{4S}$ = 12 mass % (E); UNIQUAC: (- - -) oil (1); (·····) free fatty acids (2)

Experimental and estimated selectivities for fatty acids/γ-oryzanol, calculated according to eq 3.9 below, are presented in Figure 3.4. This figure shows that the solvent selectivity, or the solvent capacity to extract free fatty acids and simultaneously to preserve γ-oryzanol in the oil, is more affected by the oil acidity ($100w_2^{oil}$) than by the water content in the solvent ($100w_{4S}$). This figure also indicates that the UNIQUAC model estimates correctly the fatty acids/γ-oryzanol selectivities, but the NRTL equation does not. In fact, at least for the lower values of fatty acid content in the oil the NRTL model estimates a decrease of the selectivity as the water concentration increases. Despite the slightly higher deviations, the UNIQUAC model estimates a behavior more consistent with the experimental measurements.

$$S_{2/5} = k_2/k_5 \qquad [3.9]$$

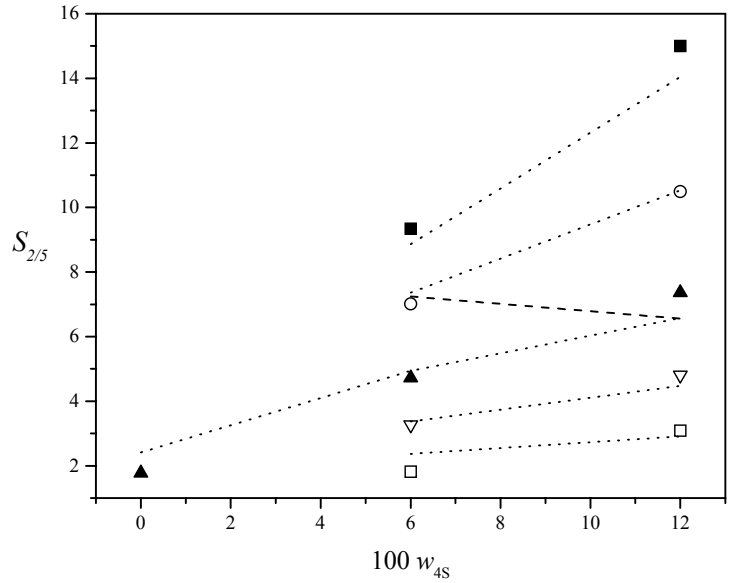

Figure 3.4. Selectivity diagram of fatty acids (2) in relation to γ-oryzanol (5) at (298.2±0.1) K for model fatty systems: (·····) UNIQUAC; (- - -) NRTL; (■)$100w_2^{oil}$ = 5 mass %; (○)$100w_2^{oil}$ = 10 mass %; (▲)$100w_2^{oil}$ = 20 mass %; (▽)$100w_2^{oil}$ = 30 mass %; (□)$100w_2^{oil}$ = 40 mass % *

The adjusted parameters for the NRTL and UNIQUAC models were tested in the prediction of liquid-liquid equilibrium for two kinds of systems: model systems, composed by refined rice bran oil containing (10, 25 or 30) mass % of commercial oleic acid, aqueous ethanol with 6 mass % of water as solvent and different contents of γ-oryzanol, and real systems, composed by crude rice bran oil with different aqueous solvents, in the ratio oil:solvent 1:1, at 298.2±0.1 K. In these cases only the partition coefficients of γ-oryzanol were measured. Table 3.8 shows the overall experimental composition of the mixtures and the corresponding equilibrium data for γ-oryzanol for the model and real systems. Liquid-liquid flash calculations for the estimation of phase compositions were performed on the basis of the overall experimental composition of the mixtures.

* O desempenho do modelo NRTL pode ser visto no ANEXO A (Figura A.6)

The deviations between experimental and estimated compositions of γ-oryzanol in both phases are calculated according to eq. 3.7 and are shown in Table 3.7. Figure 3.5 shows the experimental points and the predicted lines for the model system.

Table 3.8 – Equilibrium Data for γ-oryzanol (5) in the Model and Real Fatty Systems at (298.2±0.1) K

Systems		**Overall Composition**					**Alcohol Phase**	**Oil Phase**
	$100w_2^{oil}$[a]	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_5$	$100w_5$
Model	10	44.93	5.01	47.20	2.86	0.00	0.00	0.00
		43.19	4.82	48.79	2.79	0.42	0.12	0.78
		44.48	4.96	46.92	2.84	0.80	0.23	1.41
		44.47	4.96	46.57	2.82	0.12	0.32	1.97
		44.21	4.93	46.48	2.81	1.57	0.39	2.62
		44.06	4.92	46.30	2.80	1.92	0.56	3.56
	25	37.23	12.41	47.37	2.99	0.00	0.00	0.00
		37.17	12.39	47.05	2.97	0.41	0.20	0.70
		37.19	12.40	46.65	2.94	0.83	0.39	1.38
		36.90	12.30	46.67	2.94	1.18	0.57	1.89
		36.29	12.10	47.08	2.97	1.56	0.88	2.56
		36.79	12.26	46.12	2.91	1.93	0.95	3.09
	30	34.64	15.36	46.94	3.05	0.00	0.00	0.00
		34.49	15.29	46.78	3.04	0.40	0.22	0.60
		34.34	15.23	46.58	3.03	0.81	0.47	1.25
		34.23	15.18	46.39	3.02	1.19	0.69	1.85
		34.09	15.12	46.21	3.01	1.58	0.95	2.52
		33.96	15.06	46.06	3.00	1.93	1.03	2.80
	$100w_{4S}$[b]	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_5$	$100w_5$
Real	0	44.12	4.63	50.39	0.00	0.85	0.35	1.29
	3.35	44.14	4.64	48.68	1.69	0.85	0.23	1.49
	6.22	44.35	4.66	47.01	3.12	0.86	0.31	1.33
	10.14	44.13	4.64	45.27	5.11	0.85	0.18	1.49
	12.76	44.32	4.65	43.76	6.40	0.86	0.16	1.48

[a] $100w_2^{oil}$: free fatty acids mass percentage in the oil;

[b] $100w_{4S}$: water mass percentage in the solvent.

In Figure 3.5 it can be noted the influence of the free fatty acid content on γ-oryzanol distribution. The comparison of Figures 3.2 and 3.5 shows that the content of water in the solvent and the free fatty acids level in the oil influence the γ-oryzanol distribution coefficients in opposite directions, while the increasing of water content diminishes the extraction of oryzanol from oil, the acidity value of oil increases it. In this way, it is possible to achieve the same γ-oryzanol distribution coefficients for oils with different free fatty acid contents only by changing the water concentration in the ethanolic solvent.

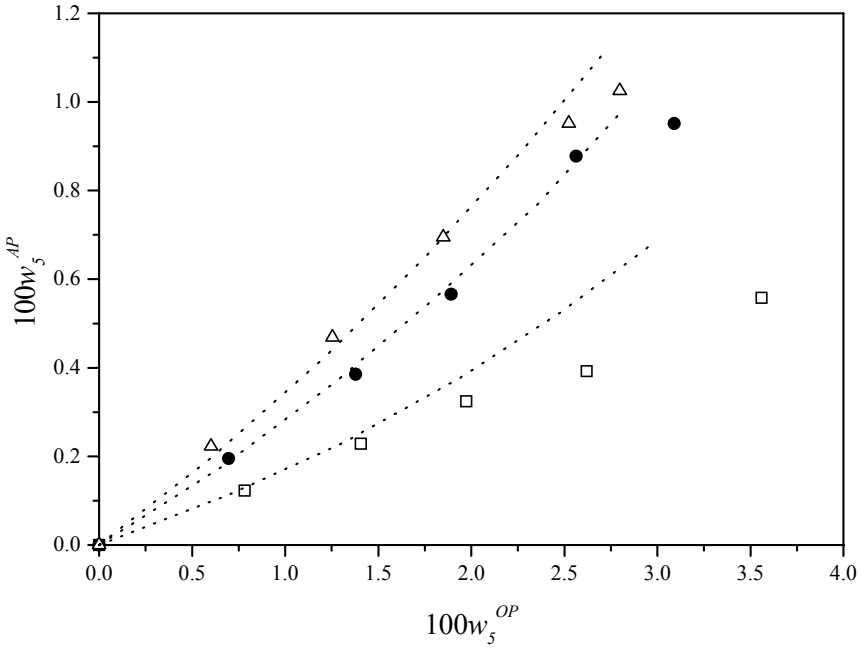

Figure 3.5. Prediction for model systems at (298.2±0.1) K and $100w_{4S}$ = 6 mass %: (□)$100w_2^{oil}$ = 10 mass %; (●)$100w_2^{oil}$ = 25 mass %; (△)$100w_2^{oil}$ = 30 mass %; (·····) UNIQUAC *

In the data corresponding to the partition coefficient of γ-oryzanol in the real systems, presented in Table 3.8, the decreasing of the partition coefficient, as a function of water percentage in the ethanolic solvent, is corroborated.**

* O desempenho do modelo NRTL pode ser visto no ANEXO A (Figura A.7)
** A predição para o sistema real pode ser vista no ANEXO A (Figura A.8)

These results also show that the strategy of constructing model fatty systems for parameter adjustment is valid. In addition, it can be concluded that the small differences in the Brazilian oils' composition do not influence the results of γ-oryzanol partition. Table 3.7 shows that the deviations for the NRTL model are higher than for the UNIQUAC model. These results confirm the best performance of the last thermodynamic equation.

With the aim of obtaining interaction parameters between tocols and the other pseudocomponents, a set of experiments for measuring tocols partition coefficients was performed. Table 3.9 presents the overall experimental composition of the mixtures and the tocols corresponding phase concentrations data for the real systems, composed by Brazilian crude rice oil and ethanolic solvents with different water percentages ($100w_{4S}$). In Figure 6 the tocols' partition coefficient, k_6, are shown as function of the mass ratio crude oil:solvent (O:S). It can be seen that tocols' partition coefficient increases slightly with the increase of the mass ratio crude oil:solvent, but it is strongly influenced by the addition of water to the solvent, a behavior similar to that already reported for the γ-oryzanol partition coefficient. Despite the same behavior for both nutraceutical pseudocompounds, it can be observed that the tocols are transferred to alcoholic phase in a major extension than γ-oryzanol. This can be attributed to structural differences between the molecules. Tocols are less hydrophobic than γ-oryzanol [41]. They are composed by smaller molecules that contain an unsaturated side-chain in the tocotrienol series and a lower number of methyl substitutions than the oryzanol molecules.

The deviation between experimental and calculated tocols' distribution coefficient, Δk_6, was calculated according to eq. 3.8, and presented a value of 0.11. The UNIQUAC model adjusted parameters for the interactions between tocols and the other compounds of the fatty system are presented in Table 3.6.

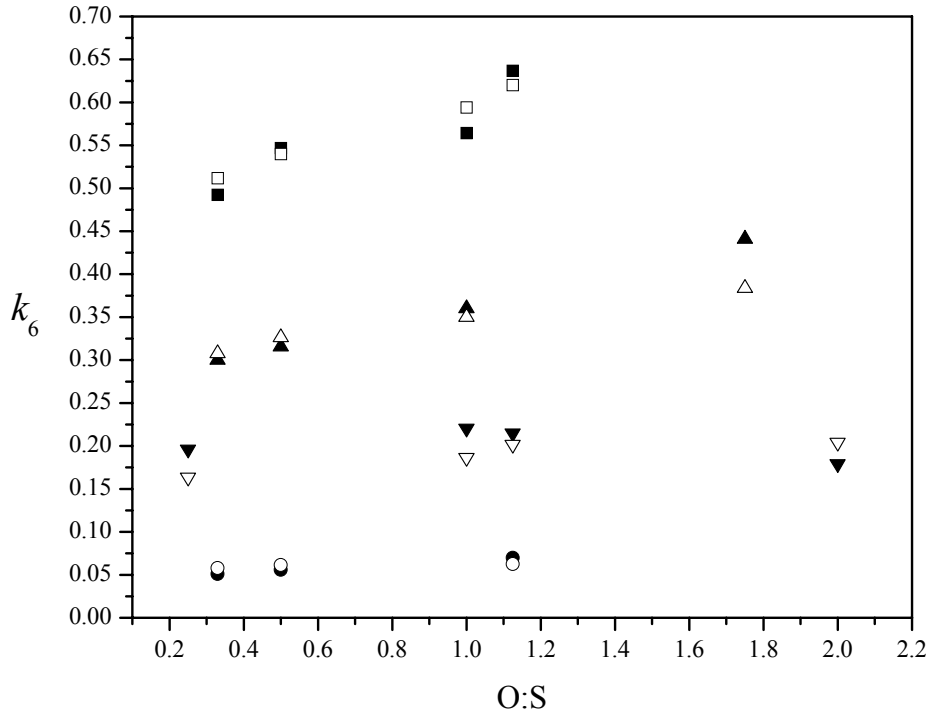

Figure 3.6. Distribution coefficient of tocols for real fatty systems at (298.2±0.1) K: experimental: full symbol; UNIQUAC: empty symbol (■) $100w_{4S}$ = 0 mass %; (▲) $100w_{4S}$ = 4 mass %; (▼) $100w_{4S}$ = 10 mass %; (●) $100w_{4S}$ = 20 mass %

Table 3.9 – Equilibrium Data for Tocols (6) in the Real Fatty Systems at (298.2±0.1) K

$100w_{4S}$[a]	Overall Composition						Alcohol Phase	Oil Phase
	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_6$	$100w_6$	$100w_6$
0	44.20	4.52	50.43	0.00	0.85	0.0311	0.0223	0.0402
	46.54	5.50	47.05	0.00	0.92	0.0392	0.0284	0.0441
	29.27	3.46	66.69	0.00	0.58	0.0245	0.0184	0.0330
	22.04	2.60	74.93	0.00	0.43	0.0191	0.0140	0.0293
3	44.73	4.57	48.16	1.67	0.86	0.0310	0.0185	0.0400
	29.71	3.04	64.44	2.23	0.57	0.0253	0.0158	0.0459
	22.29	2.28	72.49	2.51	0.43	0.0190	0.0124	0.0382
4	55.91	6.61	34.77	1.62	1.10	0.0471	0.0218	0.0504
	43.94	5.19	47.78	2.22	0.86	0.0313	0.0144	0.0391
	29.26	3.46	63.75	2.96	0.58	0.0250	0.0112	0.0364
	21.97	2.60	71.67	3.33	0.43	0.0191	0.0109	0.0372
6	44.53	4.55	46.95	3.11	0.86	0.0312	0.0133	0.0471
	29.16	3.45	62.98	3.85	0.57	0.0254	0.0086	0.0362
	21.91	2.59	73.77	4.32	0.43	0.0187	0.0095	0.0460
10	44.54	4.55	44.97	5.07	0.86	0.0313	0.0114	0.0521
	57.92	6.84	30.69	3.41	1.14	0.0491	0.0102	0.0576
	17.62	2.08	71.96	8.00	0.35	0.0149	0.0069	0.0356
	46.32	5.47	42.57	4.73	0.91	0.0390	0.0088	0.0444
12	44.60	4.56	43.61	6.38	0.86	0.0312	0.0081	0.0504
	29.14	3.44	62.34	8.53	0.57	0.0249	0.0059	0.0451
	22.00	2.60	65.40	9.57	0.43	0.0193	0.0069	0.0420
14	55.93	6.61	31.22	5.13	1.10	0.0472	0.0074	0.0482
	43.94	5.19	42.94	7.06	0.86	0.0310	0.0069	0.0540
	29.32	3.47	57.23	9.41	0.58	0.0250	0.0058	0.0494
	21.97	2.60	64.41	10.59	0.43	0.0185	0.0034	0.0304
20	46.54	5.50	37.39	9.61	0.92	0.0392	0.0039	0.0502
	29.29	3.46	53.03	13.62	0.58	0.0250	0.0029	0.0470
	21.97	2.60	59.66	15.32	0.43	0.0191	0.0024	0.0404

[a] $100w_{4S}$: water mass percentage in the solvent.

Table 3.10 presents the equilibrium data for the systems containing Thai crude rice bran oil plus ethanolic solvent with 7.45 mass % of water. These experimental data were used to evaluate the prediction capability of the adjusted parameters concerning the interaction between all components of the fatty system

(parameters published in our prior work [29] and, parameters reported in the present work (Table 3.6)).

Experimental data and predicted tie lines using the UNIQUAC and NRTL models are shown in Figure 3.7. In this figure ethanol + water is considered as a mixed solvent. Since no interaction parameters involving tocols were available for the NRTL model, this family of compounds was incorporated in the oil fraction and only the parameters between oil, free fatty acids, ethanol, water and oryzanol were considered in the flash calculations. It can be seen that the UNIQUAC equation overestimated the extraction of free fatty acids. For this reason the mass fraction average deviation is comparatively higher in the case of the UNIQUAC model (Table 3.7).

The distribution coefficients of the nutraceutical compounds of Thai crude rice bran oil, are shown in Figure 3.8. It can be observed that the distribution coefficient, for both functional compounds, is not much influenced by oil:solvent mass ratio. Furthermore, the tocols' distribution coefficient values are higher than the corresponding values for oryzanol. The UNIQUAC model predicts this behavior, although it underestimates the extraction of both pseudocompounds. The average deviations between experimental and calculated distribution coefficients for γ-oryzanol and tocols are shown in Table 3.7.

It should be considered that Thai oil has a lower level of polyunsaturated fatty compounds (see Iodine Values in Table 3.2). Despite these differences in composition of rice bran oils from different sources, both thermodynamic models and the approach based on the pseudocompounds showed similar results for correlating as well as predicting purposes. The main exception to this behavior was the incorrect estimation of the γ-oryzanol partition by the NRTL model.

The correlated parameters enable the modeling and simulation of liquid-liquid extractors for vegetable oils deacidification, making also possible the estimation of nutraceutical compounds losses during the extraction.

Table 3.10 – Liquid-liquid Equilibrium Data for the System Thai crude rice oil (1) + Fatty acids (2) + Ethanol (3) + Water (4) + Oryzanol (5) + Tocols (6) at (298.2±0.1) K

Overall Composition (OC)						Alcohol Phase (AP)						Oil Phase (OP)					
$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_6$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_6$	$100w_1$	$100w_2$	$100w_3$	$100w_4$	$100w_5$	$100w_6$
59.99	5.27	30.94	2.49	1.24	0.0680	3.87	4.77	84.34	6.74	0.27	0.0232	80.55	5.52	11.79	0.56	1.51	0.0734
45.05	3.96	46.29	3.73	0.93	0.0512	1.33	3.60	87.32	7.47	0.27	0.0134	82.48	4.32	11.00	0.60	1.54	0.0556
36.04	3.17	55.53	4.47	0.75	0.0413	1.08	2.86	89.54	6.27	0.23	0.0168	83.98	3.75	10.39	0.35	1.45	0.0689
30.03	2.64	61.71	4.97	0.62	0.0341	1.30	2.47	88.90	7.10	0.22	0.0132	85.54	3.20	9.37	0.36	1.47	0.0574
22.56	1.98	69.39	5.59	0.47	0.0260	1.12	1.90	89.87	6.91	0.19	0.0104	86.58	2.56	8.87	0.55	1.40	0.0469
17.93	1.58	74.13	5.97	0.37	0.0204	1.56	1.51	90.34	6.41	0.17	0.0101	87.17	2.14	8.97	0.39	1.29	0.0444

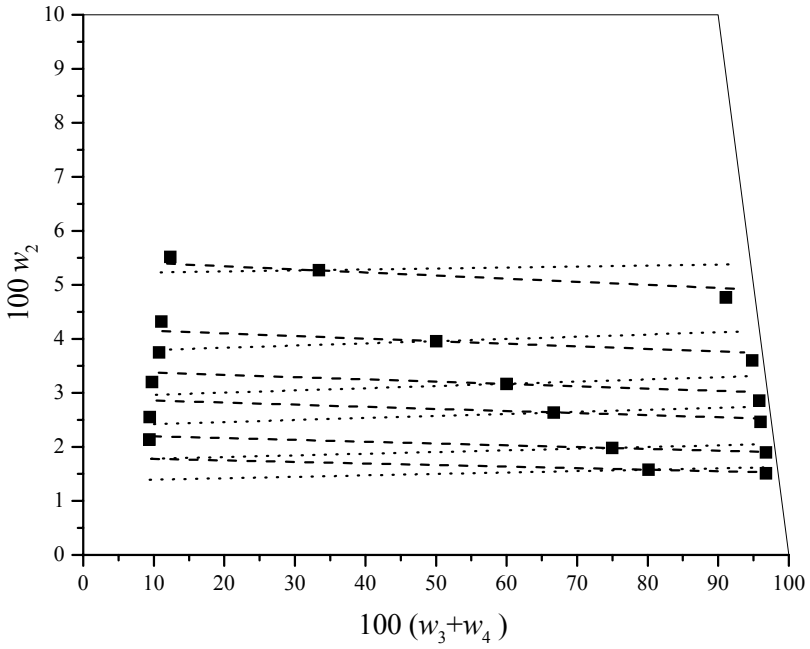

Figure 3.7. Liquid-liquid equilibrium prediction for the system of Thai crude rice bran oil (1) + Fatty acids (2) + aqueous solvent $100w_{4S}$ =7.45 mass % [ethanol (3) + water (4)] + Oryzanol (5) + Tocols (6) at (298.2±0.1) K: experimental (■); (- - -) NRTL; (·····) UNIQUAC

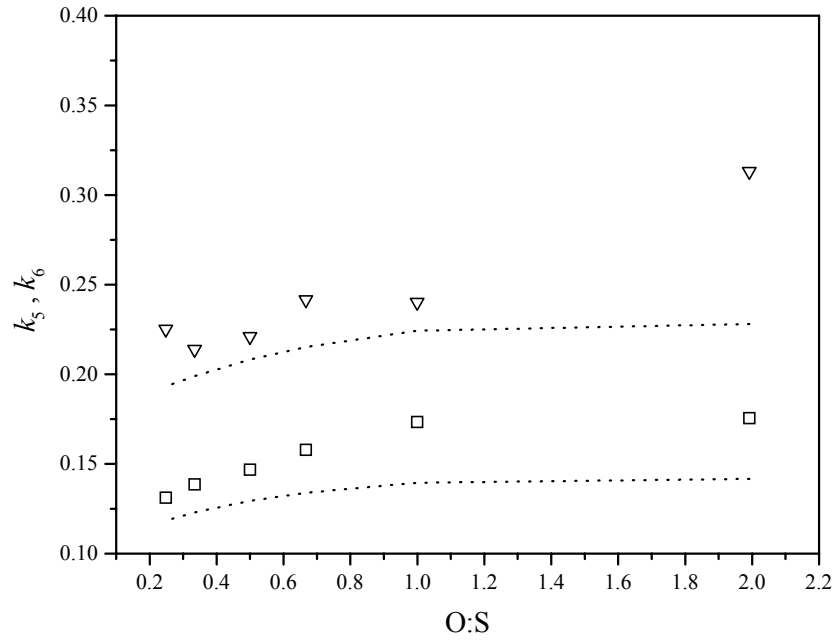

Figure 3.8. Prediction of γ-oryzanol (□) and tocols (∇) distribution coefficient at (298.2±0.1) K: (·····) UNIQUAC

3.4. Conclusions

The adjusted parameters for the NRTL and UNIQUAC models described with accuracy the phase compositions for the model systems investigated. The results obtained in the prediction of liquid-liquid equilibrium for real systems composed by Brazilian crude rice bran oil showed that the strategy of constructing model fatty systems for parameter adjustment was valid. The NRTL model presented a worse performance in the estimation of the γ-oryzanol partition than the UNIQUAC model. The interaction parameters between tocols and the others components of the system were adjusted for this last thermodynamic model using the approach of infinite dilution. The parameters obtained in this work with the parameters published in a previous paper were tested in the prediction of phase equilibrium of a system composed by crude rice bran oil from Thailand. Despite the differences in composition of rice bran oils from different sources both thermodynamic models showed similar results for correlating as well as predicting purposes, although the UNIQUAC equation overestimates the extraction of free fatty acids and underestimates the extraction of nutraceutical compounds. These results confirm that the approach considering the fatty systems composed by pseudocompounds can be used in this kind of system. The correlated parameters makes possible the estimation of nutraceutical compound losses during the deacidification of rice bran oil by liquid-liquid extraction.

3.5. List of Symbols

A_{ij}, A_{ji}	NRTL or UNIQUAC interaction parameters
C	total number of different components in the pseudocompounds
D	total number of data groups
G	total number of groups
L	total number of adjustable UNIQUAC parameters
k_i	distribution coefficient of compound i

K	total number of components or pseudocompounds in the data group m
M_i	average molar mass of the pseudocompound i
N	total number of the tie lines
$OF(w)$	objective function of composition
$OF(k)$	objective function of distribution coefficient
p_l	UNIQUAC parameters in the penalty term of eq. 6
q_i'	area parameter of component i
Q	small value constant
Q_k	Van der Waals area of group k
r_i'	volume parameter of component i
R_k	Van der Waals volume of group k
$S_{i/j}$	selectivity of compound i in relation to compound j
T	temperature (K)
$v_k^{(i)}$	number of group k in molecule i
x_i	molar fraction of compound or pseudocompound i
w_i	mass fraction of compound or pseudocompound i
Δw	phase composition global deviation
Δk_i	distribution coefficient global deviation

Greek symbol

α_{ij}	NRTL interaction parameter
γ_i	molar fraction-scale activity coefficient of compound i
$\hat{\gamma}_i$	mass fraction-scale activity coefficient of compound i
γ_i^{∞}	infinite dilution activity coefficient of compound i
τ_{ji}, τ_{ij}	NRTL or UNIQUAC interaction parameters
$\sigma_{w_{inm}^{OP}}$ e $\sigma_{w_{inm}^{AP}}$	standard deviations observed in the compositions of the two liquid phases

Subscripts

i, j	component or pseudocomponent
k	group
l	UNIQUAC parameter index
m	group number
n	tie line
s	solvent

Superscripts

AP	alcoholic phase
OP	oil phase
ex	experimental value
calc	calculated value

3.6. Appendix A

Activity coefficient (γ_i) for the NRTL model using mass fractions as unity of concentration:

$$\ln\gamma_i = \frac{\sum_{j=1}^{K}\frac{\tau_{ji}G_{ji}w_j}{\overline{M}_j}}{\sum_{j=1}^{K}\frac{G_{ji}w_j}{\overline{M}_j}} + \sum_{j=1}^{K}\left[\frac{w_j G_{ji}}{\overline{M}_j\sum_{\lambda=1}^{n}\frac{G_{\lambda j}w_\lambda}{\overline{M}_\lambda}}\left(\tau_{ij} - \frac{\sum_{\lambda=1}^{K}\frac{\tau_{\lambda j}G_{\lambda j}w_\lambda}{\overline{M}_\lambda}}{\sum_{\lambda=1}^{K}\frac{G_{\lambda j}w_\lambda}{\overline{M}_\lambda}}\right)\right] \quad [3.10]$$

where

$$G_{ij} = exp\left(-\alpha_{ij}\tau_{ij}\right) \quad [3.11]$$

$$\tau_{ij} = A_{ij}/T \quad [3.12]$$

$$\alpha_{ij} = \alpha_{ji} \quad [3.13]$$

Activity coefficient (γ_i) for the UNIQUAC model using mass fractions as unity of concentration:

$$\ln\gamma_i = \ln\gamma_i^{Comb} + \ln\gamma_i^{Res} \quad [3.14]$$

$$\ln\gamma_i^{Comb} = \frac{\ln\Psi_i'}{\ln\left(w_i/\zeta\,\overline{M}_i\right)} + 1 - \frac{\zeta\,\overline{M}_i\Psi_i'}{w_i} + \frac{z}{2}\overline{M}_i\,q_i'\ln\frac{\theta_i'}{\Psi_i'} - \frac{z}{2}\overline{M}_i\,q_i'\left(1-\frac{\Psi_i'}{\theta_i'}\right) \quad [3.15]$$

where $\zeta = \sum_{j=1}^{K}\frac{w_j}{\overline{M}_j}$ [3.16]

$$\theta_i' = \frac{q_i'w_i}{\sum_{j=1}^{K}q_j'w_j}; \qquad \Psi_i' = \frac{r_i'w_i}{\sum_{j=1}^{K}r_j'w_j} \quad [3.17]$$

and

$$\ln\gamma_i^{Res} = \overline{M}_i\,q_i'\left[1-\ln\left(\sum_{j=1}^{K}\theta_j'\tau_{ji}\right)-\sum_{j=1}^{K}\left(\theta_i'\tau_{ij}\Big/\sum_{k=1}^{K}\theta_k'\tau_{kj}\right)\right] \quad [3.18]$$

where $\tau_{ij} = \exp\left(-\frac{A_{ij}}{T}\right)$ [3.19]

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CAPÍTULO 4

Optimization of the Rice Bran Oil Deacidification Process by Liquid-liquid Extraction

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Keywords: Liquid-liquid extraction; Deacidification; Rice bran oil; Oryzanol; Tocopherols; Response surface analysis; UNIQUAC

Abstract

The present paper reports the process variable influence on the losses/transfer of fatty compounds during the deacidification process of rice bran oil by liquid-liquid extraction. The influence of process variables were analyzed using the response surface methodology, having the aim to maximize the transfer of free fatty acids and minimize the losses of neutral oil plus minor compounds. In addition, the UNIQUAC equation was used to predict the transfer/losses of fatty and nutraceutical compounds to the alcohol phase. Both methodologies, the mathematical model from non-linear multiple regression and the UNIQUAC model, presented good agreement with the experimental data.

4.1. Introduction

Rice bran oil (RBO) is extensively consumed as edible oil in Asian countries such as Japan, China, Korea, Taiwan, Thailand and Pakistan. The utilization of this oil is increasing in the western countries due to its potential as nutraceutical food (Orthoefer, 1996). RBO presents unique health benefits that may be attributed to its high level of unsaponifiable matter, whose most important components are γ-oryzanol, a complex mixture of ferulate esters with sterols and triterpene alcohols (Kim, Godber, King & Prinyawiwatkul, 2001), and tocopherols/tocotrienols, hereafter referred as tocols, a family of isomers that presents vitamin E activity (Kim *et al*., 2001; Shin, Godber, Martin & Wells, 1997).

Important physiological effects in the human body are attributed to γ-oryzanol, such as hypocholesterolemic activity, decreasing of early atherosclerosis and treatment of inflammatory processes. In relation to the tocols family, the tocotrienol isomers are the majoritary form in the rice bran oil and they present

antioxidant and antitumor activities (Kim *et al*., 2001; Xu & Godber, 1999; Deckere & Korver, 1996; Eitenmiller, 1997; Qureshi, Bradlow, Salser & Brace, 1997).

The low production of RBO in western countries may be attributed to difficulties in its processing. The production and refining of vegetable oils consist of several steps such as: its extraction from solid matrix by pressing or using organic solvents (Resa, González, Fanega, Ortiz de Landaluce & Lanz, 2002; Liu, Shi, Diosady & Rubin, 1995); solvent stripping; degumming (Indira, Hemavathy, Khatoon, Krishna & Bhattacharya, 2000); bleaching; deacidification and deodorization (Leibovitz & Ruckenstein, 1983; Cvengros, 1995). In comparison with other vegetable oils, RBO tends to contain higher levels of free fatty acids (FFA) induced by intensive enzymatic activity that difficults its deacidification by conventional processes, chemical or physical methods (Orthoefer, 1996).

These traditional techniques of vegetable oil deacidification present several disadvantages, such as high losses of neutral oil (hereafter referred as TAGs, from triacylglycerols), large production of soapstock and high energy consumption (Leibovitz & Ruckenstein, 1983; Cvengros, 1995). Furthermore, according to Orthoefer (1996), a significant portion of the nutraceutical RBO compounds is lost during these processes.

The deacidification of edible oils by solvent extraction has been receiving attention due to some clear advantages: this refining technique can be performed under mild conditions of temperature and pressure and it avoids the formation of waste products. Deacidification by solvent extraction is based on the difference of solubility of FFA and neutral triacylglycerols in an appropriate solvent (Thomopoulos, 1971) and several results reported in the literature indicate the decrease of the oil acidic value using this technique (Bhattacharyya, Majumdar & Bhattacharyya, 1987; Shah & Venkatesan, 1989; Kale, Katikaneni & Cheryan, 1999; Pina & Meirelles, 2000). Some works reported the losses of neutral oil and nutraceutical compounds during the process (Kim, Kim, Cheigh & Yoon, 1985;

Turkay and Civelekoglu, 1991a,b). In other works the liquid-liquid equilibrium for systems composed by TAGs, FFA and short chain alcohols were determined, correlated and predicted (Batista, Monnerat, Kato, Stragevitch & Meirelles, 1999; Batista, Monnerat, Stragevitch, Pina, Gonçalves & Meirelles, 1999; Gonçalves, Batista & Meirelles, 2002; Rodrigues, Antoniassi & Meirelles, 2003). In our previous article the distribution of γ-oryzanol and tocols between oil and alcoholic phases were measured and correlated by thermodynamic models (Rodrigues, Pessôa Filho & Meirelles, 2004).

The present work reports the process variable influence on the losses of γ-oryzanol, tocols and neutral oil, and on the transferred amount of FFA during the deacidification process of RBO by liquid-liquid extraction. The influence of process variables such as acidity content in the oil, water content in the ethanolic solvent and oil:solvent ratio, were analyzed using the response surface methodology. The main goal is to maximize the transfer of FFAs and minimize the losses of neutral oil and nutraceutical compounds. For comparison purposes, the UNIQUAC thermodynamic model was also used to predict the transfer/losses of fatty and nutraceutical compounds to the alcohol phase. Both methodologies, the mathematical model from non-linear multiple regression and the UNIQUAC model, presented good agreement with the experimental data.

4.2. Materials and Methods

4.2.1. Materials

The solvents used in this work were anhydrous ethanol, from Merck, with purity greater than 99.5%, and aqueous solvents with different water contents, varying in the range of (2 to 20) mass %, prepared by the addition of deionized water to anhydrous ethanol.

All fatty reagents used in this study, commercial oleic acid (Merck), refined rice bran oil (Tio João, Brazil) (RBOr), degummed rice bran oil (kindly supplied by

Helmut Tessmann, Brazil) (RBOd-HT), and degummed rice bran oil (kindly supplied by Josapar, Brazil) (RBOd-JP) were analyzed by gas chromatography of fatty acid methyl esters to determine the fatty acid composition, according to the official method (1-62) of the AOCS (1988). The fatty samples were prepared in the form of fatty acid methyl esters according to the official method (2-66) of the AOCS (1998). A HP5890 gas chromatograph with a flame ionization detector was used under the following experimental conditions: fused silica column of cyanopropylsiloxane 0.25 μm, 60 m x 0.32 mm id.; hydrogen as the carrier gas at a rate of 2.5 ml/min; injection temperature of 275.1 °C; column temperature of (175.1 to 225.1) °C (rate of 1.3 °C/min); detection temperature of 305.1 °C.

The fatty acid methyl esters were identified by comparison with external standards purchased from Nu Check Inc. (Elysian, IL). The quantification was accomplished by area normalization. All the oils used in this work were analyzed by spectrophotometry, using a UV-vis dual beam spectrophotometer (Perkin Elmer, model Lambda 40), to determine the presence and concentration of nutraceutical compounds. The concentration of γ-oryzanol was determined at 314.5 nm, as suggested by Seetharamaiah & Prabakar (1986), using heptane (UV-Fluo, Carlo Erba) as solvent and γ-oryzanol (Tsuno Rice Fine Chemicals Co., Japan), purity greater than 99%, as standard. The quantification of tocols (tocopherols and tocotrienols) was performed at 520 nm according to the methodology developed by Emmerie-Engel (Parrish, 1980); α-Tocopherol, purity greater than 99% (Sigma), was used as standard and toluene (Em Science) as solvent.

4.2.2. Methods

Model systems containing free fatty acids, triacylglycerols and γ-oryzanol were prepared by the addition of known quantities of commercial oleic acid and approximately of 1.5 mass % of γ-oryzanol to refined rice bran oil. The model fatty systems were mixed with the ethanolic solvents in the mass ratio oil:solvent 1:1, at

(25.0±0.1) ^{o}C. The experimental data measured for the model systems were planned in order to investigate the effect of oil acidity and water content in the solvent on the of losses of γ-oryzanol, and of neutral oil and on the FFA transfer.

The losses of tocols were investigated using real systems prepared by mixing the RBOd-HT with different ethanolic solvents (water content in solvent, $100w_{4S}$, varying from 2 to 20 mass %), in different mass ratios oil:solvent, at (25.0±0.1) ^{o}C. In these tests only the concentration of tocols in both phases were measured according to the Emmerie-Engel methodology (Parrish, 1980). These experimental data were planned in order to investigate the effect of the mass ratios oil:solvent and of the water content in solvent on the losses of tocols.

Liquid-liquid equilibrium data were determined using polypropylene centrifuge tubes (50 mL) (Corning Inc.). The components were weighed on an analytical balance (Sartorius model A200S), accurate to 0.0001 g. The tubes were vigorously stirred for at least 15 min, sonicated for 5 min at 40 kHz (Unique, model T1440), centrifuged for 10 min at 4500 *g* (Centrifuge Jouan, model BR4i) and left to rest for 2 h in a thermostatic bath at (25.0±0.1) °C (Cole Parmer, model 12101-05).

After this treatment, the two phases became clear, with a well-defined interface, and the composition of both phases was measured. The concentration of free fatty acids was determined by titration (official method 2201 of the IUPAC, 1979) with an automatic burette (Metrohm, model Dosimat 715). The total solvent concentration was determined by evaporation at 65.0 °C in a vacuum oven (Napco, model 5831). The water concentration was determined by Karl Fischer titration, according to AOCS method Ca 23-55 (1988), with a KF Titrino (Metrohm, model 701). Concentrations of γ-oryzanol and tocols were measured according to the procedure described above. The triacylglycerol concentration was determined by difference. In this work all measurements were performed at least in triplicate.

In order to have a better insight about the factors influencing the loss of nutraceutical compounds during the liquid-liquid extraction, the γ-oryzanol

solubility in ethanol with 6 mass % of water was measured at 25 °C. γ-Oryzanol was added to a known amount of aqueous ethanol until saturation was observed by the formation of a solid phase whose permanent presence was observed along the whole experimental period. The equilibrium cell was kept at the desired temperature by a thermostatic bath for 24 hours. Afterwards samples of the liquid phase were taken and the concentration of γ-oryzanol was analysed according to method described above (Seetharamaiah & Prabakar, 1986).

4.2.2.1. Response Surface Methodology

The response surface methodology was used to investigate the effect of some process variables on the losses of γ-oryzanol, neutral oil, tocols and on the FFA transfer, during a equilibrium stage of the liquid-liquid extraction deacidification process. Two experimental sets were performed, the first one for investigating losses of γ-oryzanol and neutral oil and the FFA transfer, and the second one for investigating the losses of tocols.

The FFA transfer, TAG loss, γ-oryzanol loss and tocols loss were calculated by eq 4.1.

$$\%(Transfer/Loss) = 100\,\frac{m^{AP} w_i^{AP}}{m^{Oil} w_i^{Oil}} \qquad [4.1]$$

where m is mass, w is mass fraction, AP is alcohol phase, Oil is oil and i is FFA, TAG, γ-oryzanol or tocol. m^{AP} was calculated by mass balance.

Both experimental sets were planned to obtain a quadratic model, consisting of 2^2 trials plus a star configuration with four repetitions in central point (Box, Hunter & Hunter, 1978; Khuri & Cornell, 1987). Surfaces were then built using the quadratic model for the statistically significant variables. The software Statistica (Statsoft, v. 5.0) was used to analyze the results by non-linear multiple regression.

4.2.2.2. FFA transfer and Losses of TAGs, γ-Oryzanol and Tocols

To test the prediction capacity of the models developed by the response surface methodology new sets of experiments with degummed oil were performed. These experiments were performed at 25.0 °C contacting RBOd-HT with solvents (anhydrous ethanol, aqueous ethanol with 6 and 9 mass % of water) and RBOd-JP with aqueous ethanol with 7.45 mass % of water, in different oil to solvent mass ratios. In both cases FFA transfer and losses of TAGs and γ-oryzanol were measured, but for the oil from Josapar (RBOd-JP) the losses of tocols were also determined. In order to compare the results obtained using the response surface methodology with a thermodynamic approach based on activity coefficients, the adjusted parameters of the UNIQUAC model, determined in previous works (Rodrigues *et al.*, 2003, 2004) were used in the prediction of transfer/losses of FFA, TAGs, γ-oryzanol and tocols. In this last case a liquid-liquid flash is used for the calculation of phase compositions based on the overall experimental composition of the mixture, following the procedure developed by Stragevitch & d'Avila (1997). The mean quadratic deviations were calculated according to eq 4.2.

$$\Delta(Transfer) = 100\sqrt{\sum_{i=1}^{n}[t_i^{ex} - t_i^{calc}]^2 \Big/ n} \qquad [4.2]$$

where t is transfer/loss in percentage, n is the number of data, *ex* and *calc* are the experimental and calculated data by UNIQUAC and RSM models, respectively.

The experimental and predicted percentages of FFA transfer, TAG loss, γ-oryzanol and tocols losses were calculated by equation 4.1.

4.3. Results and Discussion

The fatty acid composition of the degummed rice bran oil from Josapar is presented in Table 4.1. From this fatty acid composition it was possible to determine the probable triacylglycerol composition of the RBOd-JP (Table 4.2) by

using the algorithm suggested by Antoniosi Filho, Mendes & Lanças (1995). In Table 4.2 the main triacylglycerol represents the component of greatest concentration in the isomer set with *x* carbons and *y* double bonds.

Table 4.1 – Fatty Acid Composition of Degummed Rice Bran Oil from Josapar (RBOd-JP)

Symbol	Fatty Acid		*M* [b] g·mol^{-1}	Mole %	Mass %
M	Miristic	C14:0[a]	228.38	0.36	0.30
P	Palmitic	C16:0	256.43	19.62	18.18
S	Stearic	C18:0	284.49	1.89	1.94
O	Oleic	C18:1	282.47	40.07	40.90
Li	Linoleic	C18:2	280.45	35.23	35.70
Le	Linolenic	C18:3	278.44	1.71	1.73
A	Arachidic	C20:0	312.54	0.65	0.73
Ga	Gadoleic	C20:1	310.52	0.47	0.53

[a] In C*x:y*. *x*=number of carbons and *y*=number of double bonds; [b] *M*=molar mass.

Table 4.2 – Probable Triacylglycerol Composition of RBOd-JP

Group	Main Triacyl glycerol	*M* [b] g·mol^{-1}	Mole %	Mass %
50:1[a]	POP	833.37	4.56	4.38
50:2	PLiP	831.35	4.21	4.03
50:3	MOLi	828.01	0.55	0.51
52:1	POS	861.45	0.94	0.93
52:2	POO	859.40	11.36	11.25
52:3	POLi	857.39	18.66	18.43
52:4	PLiLi	855.37	9.09	8.96
52:5	PLiLe	853.37	0.79	0.78
54:2	SOO	887.46	1.58	1.61
54:3	OOO	885.44	8.19	8.36
54:4	OOLi	883.43	17.14	17.44
54:5	OLiLi	881.41	15.16	15.39
54:6	LiLiLi	879.43	5.58	5.65
54:7	LiLiLe	877.38	0.64	0.65
56:3	OLiA	913.52	0.86	0.91
56:4	OLiGa	911.50	0.69	0.73

[a] In *x:y*. *x*=number of carbons (except glicerol carbons) and *y*=number of double bonds; [b] *M*=molar mass.

Table 4.3 presents a summary of the composition of the fatty material used in the present work. The compositions of the commercial oleic acid, RBOr and RBOd-HT were taken from Rodrigues *et al.* (2003). It can be observed, based on the triacylglycerol compositions, that the oils present a great similarity concerning their chemical composition. All kinds of oils present around 90 % of triacyglycerols with molar mass between 850 and 890 $g.mol^{-1}$. This great similarity became possible the treatment of these fatty systems as composed by pseudocompounds. This approach was used by the authors with success in previous works (Rodrigues *et al.*, 2003, 2004) where model fatty systems composed by refined rice bran oil plus commercial fatty acids were used to adjust UNIQUAC interaction parameters between the system components. The prediction capability of these parameters was evaluated in the phase equilibrium description of real systems, composed by rice bran oils not refined from different sources. The calculated results were in good agreement with the experimental measurements (Rodrigues *et al.*, 2003, 2004).

Table 4.3 – Properties of Fatty Compounds

Characteristic		RBOr [a]	RBOd-HT [a]	RBOd-JP	Oleic Acid [a]
FA [b] level (mass %)	saturated	22.45	21.75	21.15	9.39
	monounsaturated	39.27	40.01	41.43	78.14
	diunsaturated	35.61	36.34	35.70	11.97
	triunsaturated	2.67	1.90	1.72	0.50
Average Molar Mass ($g \cdot mol^{-1}$)		866.50	867.78	868.19	278.96
Iodine Value [c]		101.92	102.32	101.93	93.24

[a] data taken from Rodrigues *et al.* (2003); [b] fatty acid;[c] calculated from fatty acid composition according to method Cd 1c-85 AOCS (1998).

Refined rice bran oil had a residual acidity of 0.07 mass %, expressed as oleic acid. RBOd-HT presented an acidic value of (9.34±0.01) mass %, and RBOd-JP presented an acidic value of (11.32±0.05) mass %, both expressed as oleic acid.

RBOd-HT presented (1.72±0.05) mass % of γ-oryzanol and (622.3±4.0) ppm of tocols, and RBOd-JP contained (1.73±0.08) mass % of γ-oryzanol and (801.7±38.9)

ppm of tocols. γ-Oryzanol concentration in refined rice oil was (0.12±0.01) mass % and tocols were not detected in this oil.

Table 4.4 presents all combinations of the studied variables in the statistical analysis and the correspondent responses for both experimental designs studied.

Table 4.4 – Experimental Designs: 2^2 + star configuration + central points *

Experimental design with model Systems							Experimental design with real Systems				
Coded Variables		Real Variables		Responses			Coded Variables		Real Variables		Responses
FFA	Water	FFA	Water	FFA Transfer	TAG Loss	Oryzanol Loss	O:S Ratio	Water	O:S Ratio	Water	Tocols' Loss
-1	-1	5	6	48.09	2.20	8.74	-1	-1	1.75	4.10	10.07
1	-1	15	6	50.06	3.57	13.42	1	-1	0.5	4.10	30.92
-1	1	5	12	37.05	0.95	4.54	-1	1	1.75	14.10	4.95
1	1	15	12	39.94	1.15	8.43	1	1	0.5	14.10	15.08
0	0	10	9	47.72	1.73	10.03	0	0	1.125	10	11.45
0	0	10	9	46.20	1.93	9.70	0	0	1.125	10	9.26
0	0	10	9	46.84	1.91	9.27	0	0	1.125	10	13.75
0	0	10	9	48.04	1.92	9.70	0	0	1.125	10	11.27
-1.414	0	2.91	9	53.33	1.04	6.31	-1.414	0	2	10	6.15
1.414	0	17.07	9	46.59	2.62	12.52	1.414	0	0.25	10	38.61
0	-1.414	10	4.76	51.15	4.11	12.50	0	-1.414	1.125	2	29.68
0	1.414	10	13.24	38.36	0.60	3.56	0	1.414	1.125	20	4.03

The statistical analysis of the experimental results allowed to formulate models, eqs 4.3 to 4.6, representing the percentage of FFA transfer, loss of TAGs, loss of γ-oryzanol and loss of tocols, respectively, as a function of statistically significant variables. Table 4.5 shows the analysis of variance (ANOVA) for these responses, at 95.0% of confidence. The four responses presented high correlation coefficients, in addition the F-test shows that the models are reliable since the calculated F values are at least 4 times greater than the obtained values (Box *et al*., 1978).

$$\%(FFA-Transfer) = 47.59 - 4.91(Water^*) - 2.21(Water^*)^2 \quad [4.3]$$

$$\%(TAG-Loss) = 1.83 + 0.48(FFA^*) - 1.08(Water^*) + 0.22(Water^*)^2 - 0.29(FFA^*)(Water^*) \quad [4.4]$$

$$\%(Oryzanol-Loss) = 9.58 + 2.17(FFA^*) - 2.73(Water^*) - 0.78(Water^*)^2 \quad [4.5]$$

$$\%(Tocols-Loss) = 12.73 + 9.61(O:S^*) + 4.06(O:S^*)^2 - 7.15(Water^*) \quad [4.6]$$

where: $Water^*$, FFA^* and $O:S^*$ are coded variables

Table 4.5 – Analysis of Variance (ANOVA)

Source of Variation	Transfer of FFA				Loss of TAGs				Loss of γ-Oryzanol				Loss of Tocols			
	SS[a]	MS[b]	DF[c]	F value[d]	SS	MS	DF	F value[e]	SS	MS	DF	F value[f]	SS	MS	DF	F value[g]
Regression	225.21	112.60	2	17.13	11.79	2.95	4	60.54	101.32	33.77	3	132.66	1258.2	419.4	3	22.23
Residual	59.15	6.57	9		0.34	0.05	7		2.04	0.25	8		150.93	18.87	8	
Total	284.36		11		12.13		11		103.36		11		1409.1		11	
Correlation coefficient	0.89				0.99				0.99				0.94			

[a] Sum of squares; [b] Mean square; [c] Degrees of freedom;

[d] F calc = $F_{0.95;\,2;\,9} = 4.26$; [e] F calc = $F_{0.95;\,4;\,7} = 4.12$; [f] F calc = $F_{0.95;\,3;\,8} = 4.07$; [g] F calc = $F_{0.95;\,3;\,8} = 4.07$

With these models it is possible to generate surfaces that represent the influence of different acidity values in the oil and water contents in the ethanolic solvent on the losses of γ-oryzanol and neutral oil and on the FFA transfer (Figures 4.1, 4.2 and 4.3, respectively).

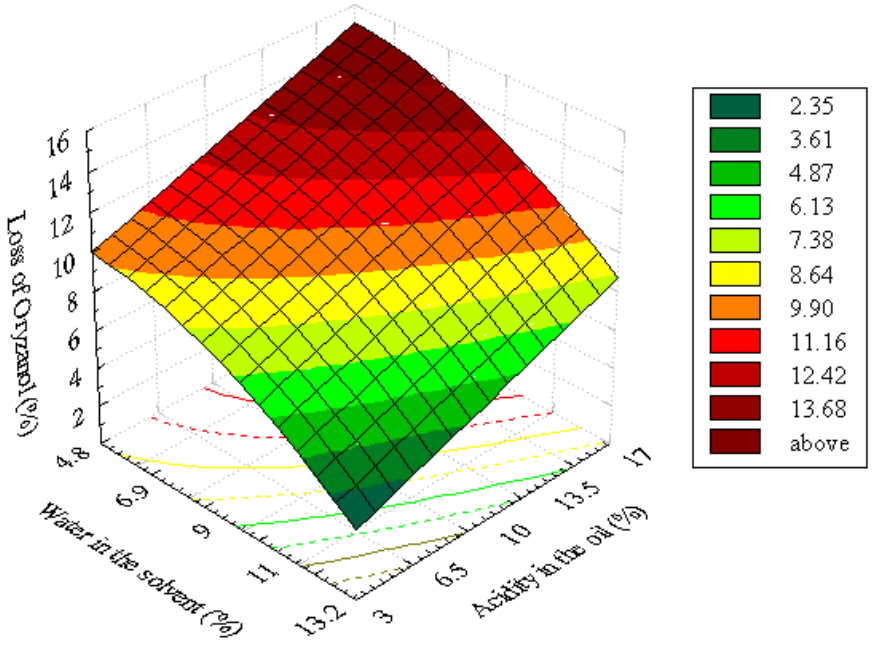

Figure 4.1. Response surface of Loss of γ-oryzanol expressed as a function of FFA in the oil and water in the solvent

In Figure 4.1 it can be observed that increasing the FFA content in the oil increases the loss of γ-oryzanol. The addition of water to the solvent reduces the solvent capacity of extracting this minor compound. In this way the losses of this nutraceutical compound can be minimized for large quantities of water in the solvent and low levels of FFA in the oil.

Figure 4.2 shows that the loss of TAGs can be reduced with the increase of water content in the solvent. In this case, the acidity of the oil has only a slight influence and specially when the content of water in the solvent is low. In Figure 4.3 it can be seen that the content of water is the principal effect on the transfer of FFA. At high values of water in the solvent, the deacidification is affected because the solvent has a lower capacity of extracting free fatty acids, nevertheless the solvent becomes more selective with the water increase, reducing the loss of TAGs (Figure 4.2) and γ-oryzanol (Figure 4.1).

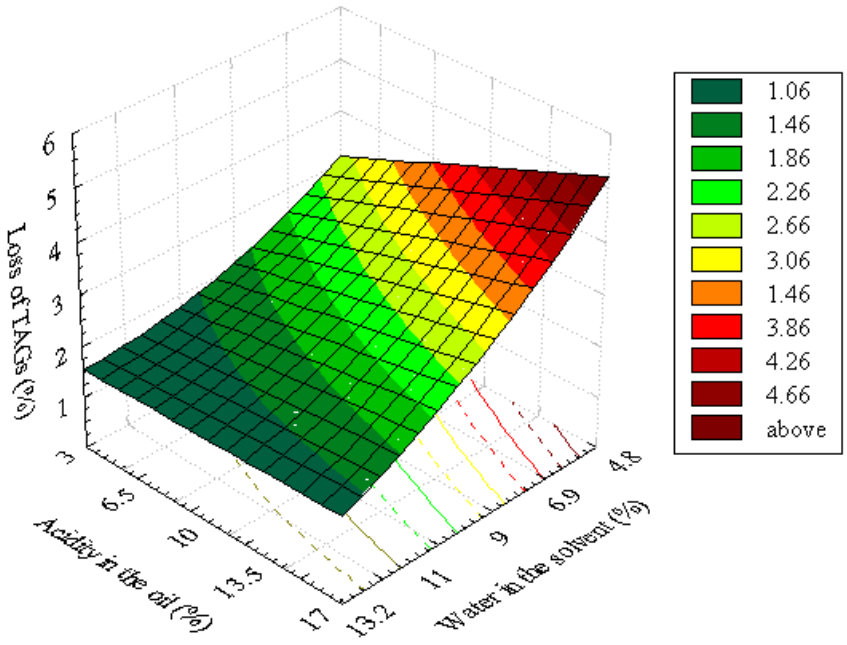

Figure 4.2. Response surface of Loss of TAGs expressed as a function of FFA in the oil and water in the solvent

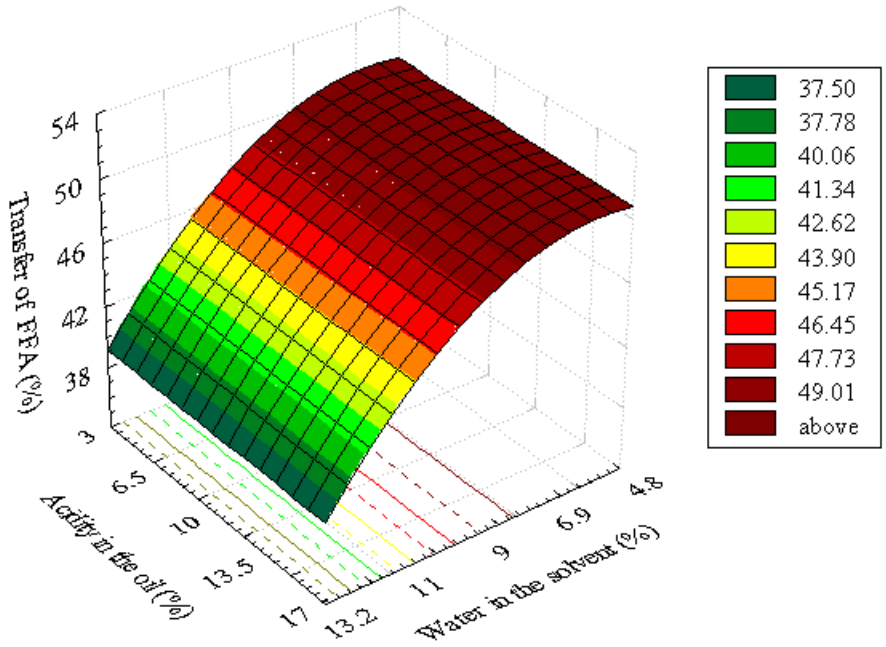

Figure 4.3. Response surface of Transfer of FFA expressed as a function of FFA in the oil and water in the solvent

An optimized objective function (OF) can be proposed as the division between the response that should be maximized by the responses that should be minimized. In this way, FO was calculated according to eq 4.7 below.

$$OF = \frac{(FFA - Transfer)}{(TAG - Loss)(Oryzanol - Loss)} \qquad [4.7]$$

Figure 4.4 shows OF as function of the studied variables for the first experimental design. It can be observed that to maximize the FFA transfer and to minimize TAG loss, simultaneously preserving the γ-oryzanol in the oil, is necessary to process oils with low contents of FFA (until 10 mass %), using ethanolic solvents with at least 12 mass % of water. In this case the objective function should be higher than 12 (in arbitrary units).

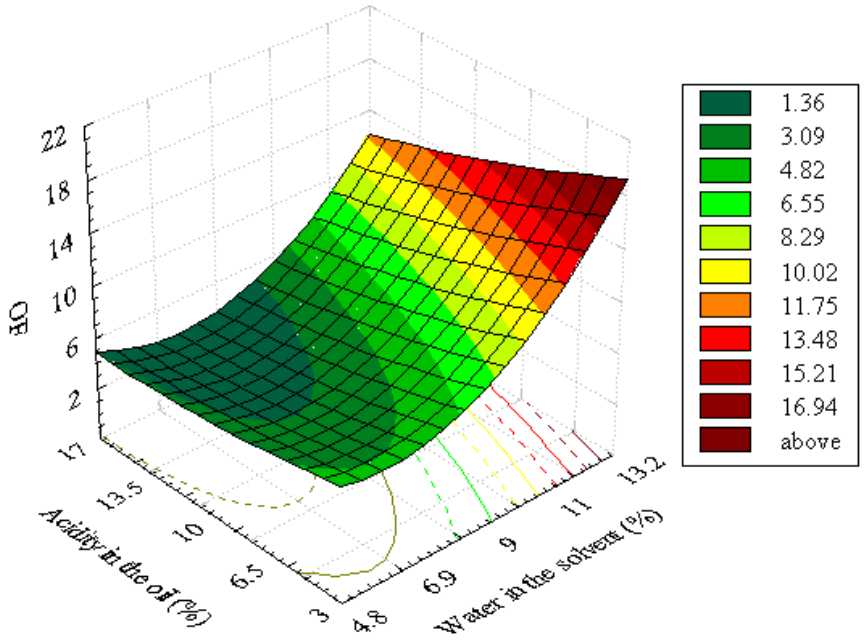

Figure 4.4. Response surface of Optimized Function (FO) expressed as a function of FFA in the oil and water in the solvent

In liquid-liquid extraction, the losses of neutral oil and nutraceutical compounds are mainly determined by the compounds solubility in the solvent. According to the equilibrium data, the oil solubility in ethanol with 6 mass % of water varies from 2.14 to 2.88 mass % as the fatty acid content changes from 0 to 10 mass % (Rodrigues *et. al.*, 2003). In the case of γ-oryzanol the solubility in aqueous ethanol with 6 mass % of water was experimentally determined at 25°C. The value of 0.12 mass % was obtained and it is in accordance with the equilibrium data where the solubility varies from 0.14 to 0.25 mass % as the fatty acid content changes from 0 to 10 mass % (Rodrigues *et. al.*, 2004). On the basis of such results the higher concentration of free fatty acids in the ethanolic solvent increases the γ-oryzanol solubility. It can be estimated that in a counter current continuous process of rice bran oil deacidification, keeping the mass ratio oil to solvent at approximately 1:1, the maximal loss of neutral oil and the γ-oryzanol in the extract stream must be lower than 3 mass % and 0.3 mass %, respectively.

On the other hand, the total transfer of FFA can be controlled by the number of the successive stagewise contacts with the solvent. In this way it is interesting to note that the denominator in equation 4.7 will be controlled by each pseudocompound solubility while the numerator is controlled by the number of the contact stages in the deacidification column.

Figure 4.5 shows the surface that represents the influence of different oil:solvent mass ratios and water contents in the ethanolic solvent on the losses of tocols, representing the results given by eq. 4.6.

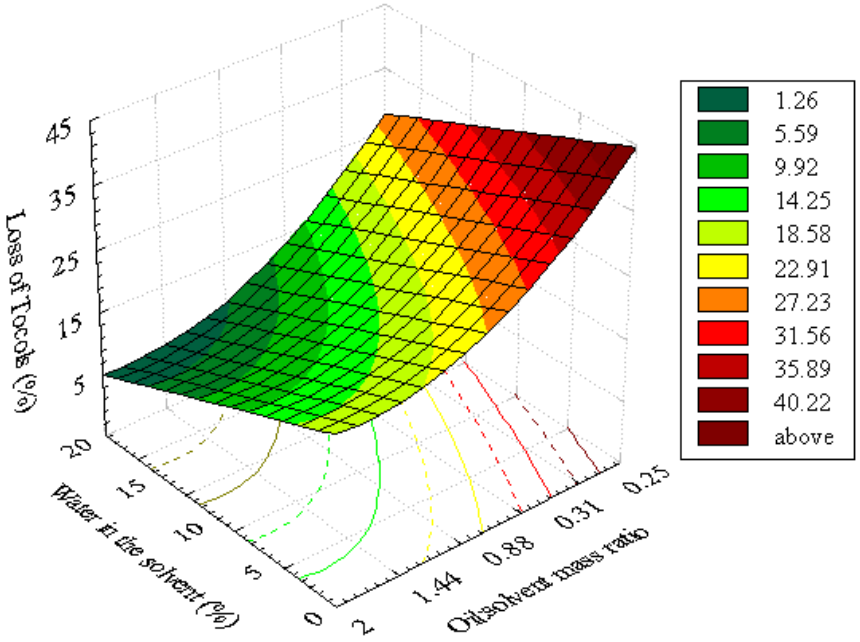

Figure 4.5. Response surface of Loss of Tocols expressed as a function of mass ratio oil:solvent and water in the solvent

In Figure 4.5 it can be observed that the effect of oil:solvent mass ratio on tocols losses is larger than the effect of water content in the solvent. The tocols losses increase when the mass ratio oil:solvent is low. In a way similar to the behavior observed for γ-oryzanol, the addition of water to the solvent reduces the solvent capacity of extracting tocols.

Table 4.6 presents experimental and calculated data obtained with degummed oils and ethanolic solvents, in different oil to solvent mass ratios. In this case the calculated data were obtained by UNIQUAC model using the interaction parameters taken from Rodrigues *et al.* (2003, 2004).

Table 4. 6 –Percentage of FFA transfer, loss of TAGs, γ-Oryzanol and Tocols

Oil	Solvent	Transfer/ Loss	Ratio Crude oil : Solvent					
			1:1		1:2		1:3	
			ex	calc[a]	ex	calc[a]	ex	calc[a]
RBOd-HT	Anhydrous ethanol	FFA	50.62	54.30	73.38	77.50	81.21	83.79
		TAGs	21.73	21.88	37.02	39.29	48.13	50.45
		γ-Oryzanol	13.90	13.90	21.05	24.96	28.26	33.57
	Aqueous ethanol with 6 mass % of water	FFA	47.54	50.56	67.00	70.42	77.33	78.89
		TAGs	1.93	3.15	4.69	6.07	7.08	8.65
		γ-Oryzanol	12.66	13.37	22.26	24.14	28.17	32.05
	Aqueous ethanol with 9 mass % of water	FFA	43.02	46.55	63.29	66.41	73.10	75.34
		TAGs	0.64	1.73	1.14	5.04	1.90	6.25
		γ-Oryzanol	7.52	7.84	22.05	23.84	31.00	32.37
RBOd-JP	Aqueous ethanol with 7.45 mass % of water	FFA	45.45	48.97	66.81	69.91	77.82	78.45
		TAGs	3.34	2.77	5.79	5.33	7.41	7.49
		γ-Oryzanol	12.77	12.40	23.84	22.28	31.86	29.14
		Tocols	15.22	19.05	21.60	33.11	32.41	41.97

[a] calculated data by the UNIQUAC model

It can be seen that the transfer of FFA and losses of TAGs, γ-oryzanol and tocols to the alcohol phase increase with the decrease of degummed oil to solvent ratio. In addition, the use of anhydrous ethanol as solvent causes a transfer/loss of compounds higher than the results obtained with aqueous ethanol, keeping the same ratio oil:solvent.

Figure 4.6 presents the experimental results and predicted values obtained by the UNIQUAC model. It can be seen that the UNIQUAC model furnishes a good prediction of the experimental behavior (Rodrigues *et al*., 2003, 2004). Figure 4.7 shows the percentages of transfer/losses of FFA, TAGs, γ-oryzanol and tocols for RBOd-JP as function of the oil:solvent mass ratio and the corresponding values predicted by the UNIQUAC model. The model predicts very well the observed behavior, although it overestimates the transfer of tocols.

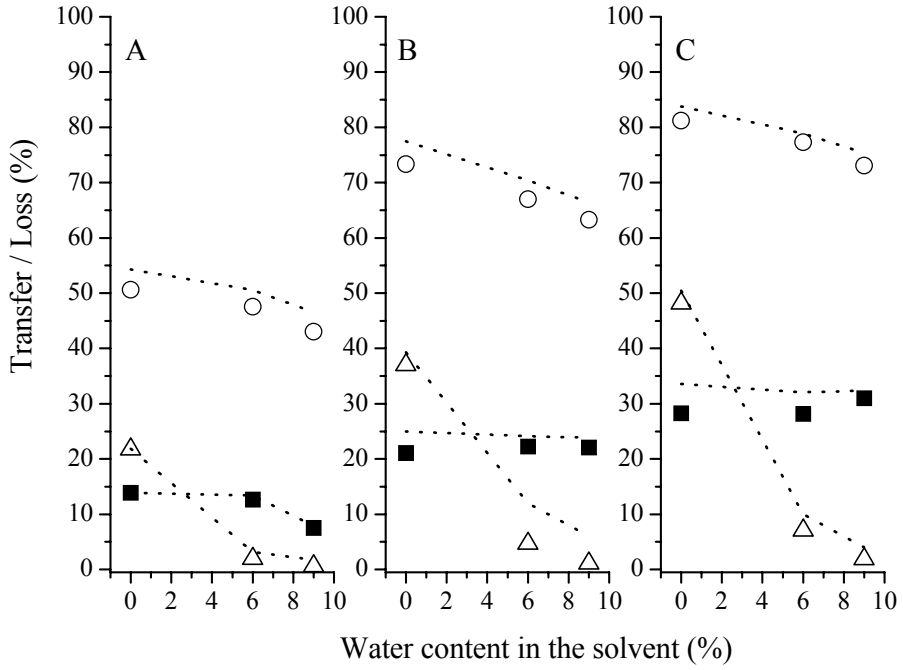

Figure 4.6. Percentage of transfer/loss for RBOd-HT using different solvents and oil:solvent mass ratios: loss of TAGs (Δ); loss of γ-oryzanol (■); transfer of FFA (○); UNIQUAC model (·····); (A) O:S 1:1; (B) O:S 1:2; (C) O:S 1:3

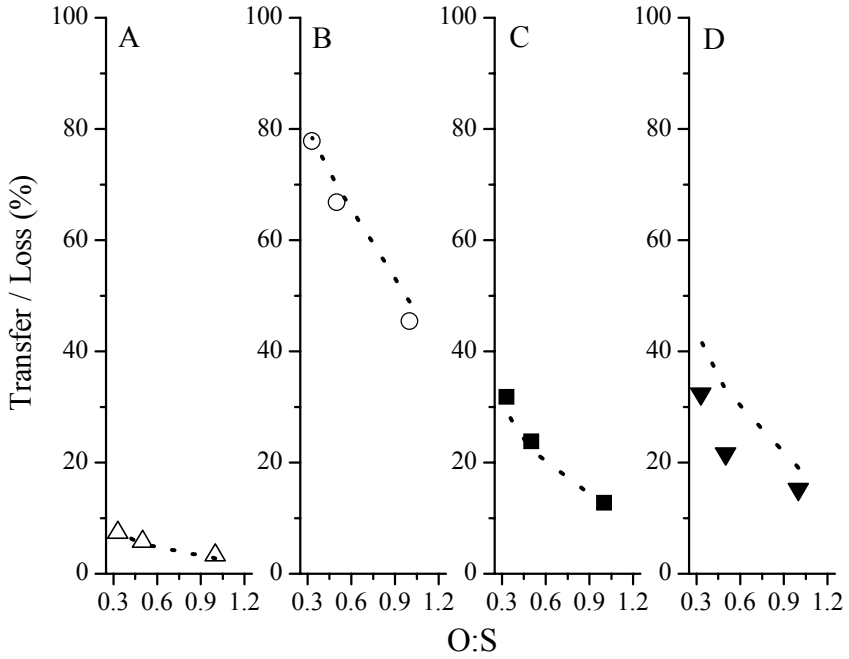

Figure 4.7. Percentage of transfer/loss for RBOd-JP as function of O:S mass ratio: (A) loss of TAGs (Δ); (B) transfer of FFA (○); (C) loss of γ-oryzanol (■); (D) loss of tocols (▼); UNIQUAC model (·····)

Table 4.7 presents the mean quadratic deviations, calculated by equation 4.2, for the two approaches used in the present work, the UNIQUAC model and the response surface methodology (RSM). In the case of the UNIQUAC model the deviations are not higher than 5%, except for the tocols losses, a result that confirms the behavior already shown in Figure 4.7. Concerning the RSM approach it can be observed that the responses transfer of FFAs and losses of tocols presented the higher deviations, which are according to the variance analysis results showed in Table 4.5. In that table it can be noted the low values of F test for these both responses.

Table 4.7 –Mean Quadratic Deviations in the Prediction of Transfer/Losses

Transfer/ Losses	Global Deviations (%)			
	RBOd-HT		RBOd-JP	
	UNIQUAC	RSM [a, b]	UNIQUAC	RSM [a, b]
FFA	3.12	3.77	2.73	4.09
TAGs	2.39	1.16	0.43	0.72
γ-Oryzanol	2.74	1.61	1.82	1.39
Tocols	-	-	8.91	2.19

[a] response surface methodology; [b] mass ratio oil:solvent 1:1

In general the results show that the total deacidification of rice bran oil by liquid-liquid extraction is possible. The losses of neutral oil (TAGs) and nutraceutical compounds can be controlled by the choice of solvent. In addition it can be suggested that the conditions corresponding to oil solvent mass ratios around of 1:1, water content in the solvent of at least 12 mass % and low level of acidity in the degummed oil permit the oil deacidification with acceptable levels of neutral oil (TAGs) and nutraceutical compounds losses.

4.4. Conclusions

The experimental results of the oil deacidification by liquid-liquid extraction in one equilibrium stage and the corresponding values generated by the response surface methodology allow the selection of process conditions that maximize the transfer of FFAs and minimize the losses of neutral oil and nutraceutical compounds. In addition, the UNIQUAC model was able to predict correctly the phase equilibrium behavior of the fatty systems studied in the present work.

4.5. Acknowledgements

The authors wish to acknowledge FAPESP (Fundação de Amparo à Pesquisa do Estado de São Paulo – 99/12033-1 and 01/10137-6) and CNPq (Conselho Nacional de Desenvolvimento Científico e Tecnológico – 521011/95-7), for the financial support.

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CAPÍTULO 5

Performance of a Perforated Rotating Disc Contactor in the Deacidification of Rice Bran Oil

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Keywords: Rice bran oil, deacidification, liquid-liquid extraction, PRDC, mass transfer coefficients.

Abstract

The deacidification of rice bran oil by continuous liquid-liquid extraction, using as solvent ethanol containing approximately 6 mass % of water, was investigated in a rotating disc column (PRDC). The influence of the presence of acylglycerols and minor compounds in the free fatty acids mass transfer was observed. The experimental results obtained proved that the total deacidification by solvent extraction is feasible. The losses of neutral oil and nutraceutical compounds were significantly lower than the results reported in the literature for alkali or physical refining of rice bran oil. The simulation of extraction process was also performed presenting good agreement with experimental data.

5.1. Introduction

Rice bran oil (RBO) has been used only to a small extent of its potential as edible oil because of problems with the stability and storage of rice bran. Since 1930 the use of RBO as edible oil has increased, particularly in Japan, where it is used as salad and frying oil (Orthoefer, 1996). Crude rice bran oil is not suitable for human consumption and must be refined. Refining of crude RBO involves dewaxing, degumming, deacidification, bleaching to improve color, and steam deodorization.

In a general way the dewaxing step consist in a cooling and filtration to separate the wax from oil. Degumming prior the deacidification is a crucial preliminary step because the stability of the oil is affected by traces of metals and phosphorus (Indira *et al*., 2000). These compounds are eliminated from oil by addition of water, with or without additives such as citric and phosphoric acids. Bleaching is adopted to lighten the color and improve the stability of the oil and is

performed using activated carbon or bleaching earth. Deodorization of the deacidified, bleached RBO involves steam stripping to remove the objectionable odors resulting from peroxides, aldehydes, and ketones from oil degradation as well as the characteristic rice oil odors and flavors (Juliano, 1985).

In relation to deacidification the methods most used are alkali refining or physical refining. The neutralization of FFA in rice oil by alkali refining uses either batch or continuous processes, 15-20% sodium hydroxide with a 0.5-2% excess, based on the FFA content. Excessive losses of neutral oil may occur due to saponification and entrainment by the soapstock. A crude oil with 5 mass % of FFA has losses ranging 12 to 40% (Orthoefer, 1996). Alkali refining requires centrifuges for neutralization and washing, implying higher investments and effluent treatment costs (Leibovitz and Ruckenstein, 1983).

Physical or steam refining is practiced by various refineries in Japan and United States, and it combines deacidification with deodorization. This method is performed at high temperatures and low pressures involving high-energy inputs and costly machinery. Physical refining is more efficient for higher FFA contents in rice oils giving better yields of neutralized oils than alkali refining although, the quality of the final product depends on the quality of the crude oil (Juliano, 1985; Cvengros, 1995).

These traditional refining methods were shown to affect the content of unsaponifiable matter in the rice bran oil (Deckere and Korver, 1996). This fraction of crude RBO presents as major compounds γ-oryzanol and vitamin E, and it was suggested as being responsible for various physiological functions such as cholesterol reduction, prevention of cardiovascular disease and some forms of cancer (Rukmini, 1988; Qureshi *et al*., 2000). Alkali refining results in a near order of magnitude decrease in total oryzanol of 1.6% to 0.2% (mass % in the oil) while for physical refining most of 50% of the oryzanol is lost. In relation to vitamin E

(tocopherols plus tocotrienols) content it is drastically reduced during the deodorization step (McCaskill and Zhang, 1999; Krishna *et al*., 2001).

Various refining methods able to deacidify RBO preserving the oryzanol and vitamin E in the oil have been tried. The use of liquid-liquid extraction technique in the deacidification step can be a feasible alternative to attain this aim (Kim *et al*., 1985). Liquid-liquid or solvent extraction can be carried out at room temperature and atmospheric pressure, reducing the energy consumption for oil refining without losses of nutraceutical compounds. Some researchers report the decreasing of the rice bran oil acidic value (Bhatacharyya *et al*., 1987; Shah and Venkatesan, 1989; Kim *et al*., 1985; Kale *et al*., 1999) using this approach with several solvents such as isopropanol and methanol.

To make possible the use of liquid-liquid extraction as a substitute of deacidification by neutralization or steam distillation, the study of performance of equipment, hydrodynamic behavior and mass transfer phenomena, are required. Pina and Meirelles (2000) studied the dispersed phase holdup performance of a Perforated Rotating Disc Column in the continuous deacidification of systems composed by corn oil/oleic acid/aqueous ethanol, obtaining good results in relation to the extraction of free fatty acids and loss of neutral oil. In addition to hydrodynamic and mass transfer aspects studies about the quality of oil deacidified by liquid extraction are necessaries. With this purpose, this paper presents a investigation about mass transfer in a rotating disc column for the rice bran oil deacidification process. The results obtained proved that the total deacidification by solvent extraction is feasible, with low level of neutral oil and nutraceutical compounds losses.

5.2. Experimental Apparatus

The perforated rotating disc contactor (PRDC) was made of glass, jacketed, presenting 50 mm internal diameter, 1300 mm column height and 1000 extraction zone height.

Perforated discs (total of 33, with 47 mm diameter, drilled with holes of 3 mm diameter presenting 20 % of flow free area) were placed at constant spacing distances (25 mm) in the column. They were mounted on a central shaft, which was rotated at different velocities (50 to 300 rpm).

A schematic drawing of the PRDC used in this work is shown in Pina and Meirelles (2000). The apparatus was maintained at (298 ± 1) K and local atmospheric pressure (727 mmHg).

The equipment was feed with the continuous phase (ethanol food grade containing aproximately 6% of water) through the bottom of the column and its flow rate was maintained at the desired constant value. The rotor was started and the rotating speed was measured by a digital tachometer 1726 (Ametek, Largo, FL). The dispersed oil phase was feed to the top of the column with its flow rate at the desired value. Both phases were pumped into the column by peristaltic pumps (Cole Parmer, Chicago, IL).

For the mass transfer experiments the operational variables were rotor speed (50-300 rpm), phase ratio (V_d/V_c = 0.70 to 1.19 – where V_d and V_c are the superficial velocity of the dispersed and continuous phases, respectively. These values are very close to a mass ratio oil to solvent *O:S*=1), and fatty acid content in the oil feed stream (5.5 to 11.7 mass %). Inside the column, steady state was achieved after aproximately 120 min of continuous operation. This was verified by the constant position of the interface level at the bottom of the equipment.

After achieved the steady state five samples of outlet phases, extract and raffinate, were taken (every 15 minutes) and analyzed to determine the alcohol, water, and acid concentrations of each phase. These analyses were performed at least in triplicate. In addition the mass flow rates of extracted and raffinated phases were monitorated by the totalization of mass on time during the experiment *.

5.3. Materials and Methods

To evaluated the influence of the fatty raw material kind in the volumetric mass transfer values three kinds of rice bran oils were used, refined oil (RBOr) with addition of acidity, crude (RBOc) and degummed (RBOd) oils

Refined rice bran oil (Blue Ville) was purchased from Santalúcia, Brazil, and crude and degummed rice bran oils were kindly supplied by Helmut Tessmann, Brazil. The fatty acid source used in the experiments with refined oil was a commercial oleic acid (Merck, Germany). The solvent used in all experimental runs was neutral aqueous ethanol, food grade, purchased from Usina Ester, Brazil.

All fatty reagents used in this work were analyzed by gas chromatography of fatty acid methyl esters according to official method 1-62 of the American Oil Chemists' Society (AOCS) (1988) to determine the fatty acid composition. The fatty samples were prepared in the form of fatty acid methyl esters according to the official method (2-66) of the AOCS (1998). A HP5890 gas chromatograph with a flame ionization detector was used under the following experimental conditions: fuse silica column of cyanopropylsiloxane 0.25 μm, 60 m x 0.32 mm id; hydrogen as the carrier gas at a rate of 2.5 ml/min; injection temperature of 548.2 K; column temperature of 448.2 - 498.2 K (rate of 1.3 K/min); detection temperature of 578.2 K. The FAME were identified by comparison with external standards purchased from Nu Check Inc. (Elysian, IL). The quantification was accomplished by internal normalization.

* Um exemplo da evolução das concentrações com o tempo de operação está mostrado na Figura D.1 (Anexo D)

In addition, the fatty samples were characterized in terms of concentration of phosphorus (colorimetric method AOCS Ca 12-55); mono-, di-, triacylglycerols and polymerized triacylglycerols by gel permeation HPLC (AOCS Cd 22-91; HPLC system Perkin Elmer model 250, refractive index detector Sicon Analytic, columns Jordi Gel DVB 300 mm x 7.8 mm id, 0.01 and 0.05 μm, mobile phase tetrahydrofurane, sample solution 1% (w/v) in tetrahydrofurane); peroxide value (method AOCS Cd 8b-90); iodine and saponification values calculated from fatty acid composition (AOCS methods Cd 1c-85 and Cd 3a-94, respectively), determination of total tocopherols (tocopherols and tocotrienols) by HPLC (AOCS official method Ce 8-89; HPLC system Perkin Elmer model 250, fluorescence detector Shimadzu RF-10 AXL with excitation wavelength at 290 nm and emission wavelength at 330 nm, column Merck Li Chrosorb Si 60, 5 μm, 250 mm x 4 mm id, mobile phase isopropanol in hexane 1:99 (v/v)); concentration of γ-oryzanol determined by spectrophotometry, using a UV-vis dual beam spectrophotometer (Perkin Elmer, model Lambda 40) at 314.5 nm, as suggested by Seetharamaiah and Prabakar (1986), using heptane (UV-Fluo, Carlo Erba) as solvent and γ-oryzanol, purity greater than 99% (kindly supplied by Tsuno Rice Fine Chemicals Co., Japan) as standard; and Lovibond color read in a Lovibond Tintometer model E in a 5.25" cell and expressed in units of yellow (Y), red (R), and blue (B).

For all experiments the concentration of free fatty acids was determined by titration (official method 2201 of the IUPAC, 1979) with an automatic burette (Metrohm, model Dosimat 715). The total solvent concentration was determined by evaporation at 338.15 K in a vacuum oven (Napco, model 5831). The water concentration was determined by Karl Fischer titration, according to AOCS method Ca 23-55 (1988), with a KF Titrino (Metrohm, model 701). The acylglycerol concentration was determined by difference. In this work all measurements were performed at least in triplicate.

Physical properties of the liquids were measured at 298.15 K. The density measurements were performed using a DMA 58 Density Meter (Anton Paar, Austria) and the viscosity data were obtained by an AMV 200 Viscometer (Anton Paar).

Table 5.1 summarizes the experimental runs accomplished.

Table 5.1– Experiments Description

Oil	Acidity in the Feed (mass %)	Rotor Speed (rpm)	Phase ratio V_d/V_c	Experiment number
RBOr	5.53	150	0.70	1
		200		2
		250		3
		300		4
	6.42	50	1.18	5
		150		6
	11.67	200	1.19	7
RBOc	7.66	200	0.82	8
RBOd	8.52	150	0.81	9

Other aim of this work was to evaluate the impact of the deacidification by liquid-liquid extraction on the quality of deacidified oil. For this reason the raffinate stream obtained from experiment 9 (see Table 5.1) was reprocessed four times in a scheme similar to a crosscurrent extraction as showed in Figure 5.1.

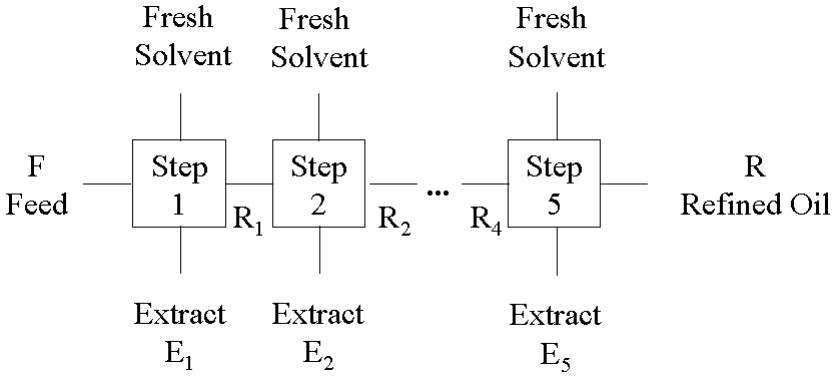

Figure 5.1. Schematic representation of the crosscurrent extraction.

In this scheme the raffinate of the first step (experiment 9) was successively contacted with fresh solvent obtaining a single final raffinate while the extracts of the sucessive steps may be combined to provide the composite extract. Each step of processing is a countercurrent contact while the global process is a crosscurrent configuration. In all steps of processing the rotor speed was maintained at 150 rpm and the ratio of continuous and dispersed phases V_d/V_c at 0.81. For these experiments the outlet phases were submitted to quantification of acidity, ethanol, water, triacylglycerols, partial acylglycerols, phosphorus, total tocopherols and γ-oryzanol levels as described above. Five steps were necessary in order to obtain the final acidity required by the Codex Alimentarius (1993) for refined vegetable oils.

5.4. Process Simulation

The degummed rice bran oil deacidification process was simulated using the algorithm suggested by Naphtali and Sandholm (1971), according to the version presented by Fredenslund *et al*. (1977) and adapted by Batista *et. al.* (2002) for simulating stagewise liquid-liquid extraction. The whole solvent extraction process was simulated considering the following configuration: each extraction step was considered as a countercurrent one and the set of 5 steps was organized in a crosscurrent way because fresh solvent was used in each subsequent step.

To compare the experimental and calculated values of column outlet phases concentrations the number of ideal stages required in each step was calculated based on the operational and equilibrium concentrations of free fatty acids in the raffinate stream. This approach is valid when the operating and equilibrium lines are both straight over a given concentration range, and is given in equation 5.1 below:.

$$N = \frac{\log\left[\left(w_{R,2}{}' - w_{R,2}^{*}{}'\right)\Big/\left(w_{F,2}{}' - w_{E,2}^{*}{}'\right)\right]}{\log\left[\left(w_{E,2}^{*}{}' - w_{R,2}^{*}{}'\right)\Big/\left(w_{F,2}{}' - w_{R,2}{}'\right)\right]} \qquad (5.1)$$

where N is the number of stages, $w_{F,2}^{'}$ $w_{E,2}^{'}$ and $w_{R,2}^{'}$ are the concentration of fatty acids in the feed, extract and raffinate streams, respectively; the superscripts ' and * denote, acid free-basis and equilibrium concentration, respectively (McCabe and Smith, 1976). This approach is necessary in order to transform the differential contact prevailing in the PRDC into the stagewise contact calculated in the computational algorithm (Batista *et al*., 2002).

Phase equilibrium was predicted using parameters of UNIQUAC adjusted by Rodrigues and coworkers (2003,2004a) in previous papers for systems composed by rice bran oil, free fatty acids, γ-oryzanol, tocols, ethanol and water.

Considering that industrial equipments are usually operated in a countercurrent way, for comparison purposes the whole extraction process was also simulated in this version.

The mean quadratic deviations for crosscurrent and countercurrent simulations were calculated according to eq 5.2.

$$\Delta w(\%) = 100\sqrt{\sum_{j=1}^{K}\sum_{i=1}^{n}\left(w_{i,j}^{ex} - w_{i,j}^{calc}\right)^2 \Big/ K} \qquad (5.2)$$

where w is concentration of pseudocompound i in the refined oil (mass %, in solvent free-basis), K is the number of extraction steps, *ex* and *calc* are the experimental and calculated concentrations, respectively.

5.5. Results and Discussion

The fatty acid compositions of the refined oil (RBOr), crude oil (RBOc), degummed oil (RBOd), degummed oil deacidified by liquid-liquid extraction (RBOd-LE), and commercial oleic acid, are presented in Table 5.2. It can be

observed that the oils present a great similarity concerning their chemical composition.

The results shown in Table 5.2 allows to calculate the average molar mass (M_{AV}) of free fatty acids in crude and degummed rice oils and in commercial oleic acid.

Densities and viscosities of the liquids at 298.15 K are given in Table 5.3.

Table 5.2 – Fatty Acid Composition of Refined, Crude, Degummed, and Deacidified by Liquid Extraction Rice Bran Oils and Commercial Oleic Acid

Symbol	Fatty Acid		$\underline{M}$ g·mol^{-1}	RBOr		RBOc		RBOd		RBOd-LE		Oleic Acid [a]	
				Mol %	Mass %	Mol %	Mass %	Mol %	Mass %	Mol %	Mass %	Mol %	Mass %
L	Lauric	C12:0	200.32	-	-	-	-	-	-			1.58	1.13
M	Miristic	C14:0	228.38	0.32	0.26	0.34	0.28	0.39	0.32	0.33	0.27	1.09	0.89
P	Palmitic	C16:0	256.43	0.20	0.18	0.21	0.19	0.22	0.20	0.20	0.18	5.83	5.36
Po	Palmitoleic	C16:1	254.42	18.82	17.43	20.93	19.42	21.32	19.79	20.78	19.28	0.13	0.12
S	Stearic	C18:0	284.49	1.74	1.79	1.99	2.05	2.07	2.13	1.98	2.04	1.39	1.42
O	Oleic	C18:1	282.47	40.66	41.48	39.73	40.60	39.29	40.17	39.46	40.32	77.05	78.02
Li	Linoleic	C18:2	280.45	36.19	36.65	33.76	34.26	33.73	34.24	34.16	34.66	11.91	11.97
Le	Linolenic	C18:3	278.44	1.02	1.03	1.90	1.91	1.88	1.90	1.91	1.93	0.49	0.50
A	Arachidic	C20:0	312.54	0.57	0.64	0.65	0.74	0.62	0.70	0.66	0.75	0.53	0.59
Ga	Gadoleic	C20:1	310.52	0.48	0.54	0.49	0.55	0.49	0.55	0.51	0.57	-	-
M_{AV} of Free Fatty Acids (g·mol^{-1})				-		276.39		276.25		276.44		278.96	

[a] data taken from Rodrigues et al. (2003).

Table 5.3 – Physical Properties of the Oils, Solvent and the Phases at 298.2 K

		Density (kg/m^3)	Viscosity (mPa.s)
Inlet Streams	RBOr	915.19	65.59
	RBOc	918.86	112.75
	RBOd	917.32	107.87
	Aqueous Ethanol	809.78	1.72
Outlet Streams	**RBOr** [a]		
	Extracted	813.38	1.71
	raffinate	901.80	38.34
	RBOc		
	Extracted	815.64	2.38
	raffinate	891.92	68.14
	RBOd		
	Extracted	814.39	1.89
	raffinate	895.27	72.69

[a] relative to experiment 1.

In order to evaluate the deacidification experiments the volumetric overall mass transfer coefficients for the fatty acids were estimated based on the units of concentration of dispersed phase. The calculation of an overall coefficient applicable to entire column requires that the film coefficients for each phase be approximately constant and that the equilibrium curve be a straight line (Treybal, 1980). Both approximations are valid in the present case. It should be further considered that the effect of axial mixing and the influence of the mass transfer of other compounds were not taken into account, so that the calculated values should be better considered as apparent overall coefficients.

Despite this fact, the obtained results can undoubtedly help in the design of liquid-liquid extractors for edible oil deacidification.

The equilibrium curve was obtained from equilibrium data determined according to the procedure described by Rodrigues *et al.* (2003) for the rice bran oil / oleic acid / ethanol / water system. The equilibrium curve is a straight line with a correlation coefficient of 0.9998 for a fatty acid concentration range of 0 to 25 mass %. This line is given by equation:

$$w'_{E,2} = 0.9462 w'_{R,2} \qquad (5.3)\ *$$

where $w'_{E,2}$ and $w'_{R,2}$ are the concentration of fatty acids in the extract and raffinate phases, respectively. These concentrations were the mass fractions on a fatty acid-free basis, calculated using the following equation:

$$w'_2 = \frac{w_2}{1 - w_2} \qquad (5.4)$$

where w_2 is the mass fraction of fatty acids in each phase **

The overall mass transfer coefficients can be calculated using equation 5.5:

$$R'(w'_{F,2} - w'_{R,2}) = K_R a V \Delta w'_{R,2M} \qquad (5.5)$$

where R' is the dispersed phase mass flow on a fatty acid free-basis in kg/s; $w'_{F,2}$ and $w'_{R,2}$ are the fatty acid concentrations in the feed and raffinate streams, respectively; K_R is the overall mass transfer coefficient in Kg fatty acids/[m^2.s.(kg fatty acids/ kg oil phase)]; a is the mass transfer area per unit of extraction zone volume in m^2/m^3; V is the volume of extraction zone in m^3; and $\Delta w'_{R,2M}$ is the logarithmic average of the concentration differences at the ends of the column based on the units of the concentration of the oil phase. $\Delta w'_{R,2M}$ was calculated by taking into account the equilibrium concentrations given by equation 5.3 (Treybal, 1980).

* Um exemplo de curva de equilíbrio e linha de operação pode ser visto na Fig. D.2 (Anexo D)
** Os dados das composições das correntes do extrator estão mostrados na Tab. D.1 (Anexo D)

Note that by dividing the K values by the density of the oil phase, given in Table 5.3, we obtain the overall mass transfer coefficient in m/s, which is an entity frequently used in the literature.

In the evaluation of mass transfer coefficients the systems were treated as composed by pseudocomponents as a fatty acid equivalent to commercial oleic acid, in the case of systems with refined rice oil, or a fatty acid equivalent to free fatty acids presents in crude or degummed rice oil, and a triacylglycerol equivalent to rice bran oil, ethanol and water. This approach was successfully used by Batista et al. (1999), Gonçalves et al. (2002) and Rodrigues et al. (2003,2004a). These last authors used this approach for modeling the phase equilibrium of systems composed by rice bran oil, free fatty acids, ethanol, and water. The equivalent fatty acid was the main pseudocompound that moved away from the oil to the alcoholic phase.

The volumetric mass transfer coefficients are given in Table 5.4. Their values increased as the rotor speed and the oil phase flow increased, as a consequence of the higher turbulence level in the apparatus.

Table 5.4 – Volumetric Mass Transfer Coefficients

Experiment number	Phase ratio V_d/V_c	$K_r a$. 10 [kg fatty acids / m³s (kg fatty acids /kg oil phase)]
1	0.70	4.60
2		4.69
3		4.75
4		5.04
5	1.18	2.64
6		4.67
7	1.19	5.15
8	0.82	3.93
9	0.81	3.52

The values of volumetric mass transfer coefficients of this work are in accordance with the values obtained by Pina and Meirelles (2000) for deacidification of corn oil. The experiments 8 and 9 accomplished with crude and degummed rice oils, respectively, presented the lowest values of volumetric mass transfer coefficients. These results can be a consequence of the presence of high contents of phosphorus in the degummed and crude oils (55.19 and 121.33 ppm, respectively, while for refined rice oil the level of phosphorus quantified was lower than 1 ppm). These compounds cause the formation of a emulsion between oil and solvent that difficults the phase separation at the bottom of the column as shown by the high content of solvent in the raffinate stream (22.56 mass % for experiment 8 and 13.19 mass % for experiment 9). For all the others experiments the content of solvent in the raffinate stream is not higher than 11 mass %, a value that is in accordance with the solubility of aqueous ethanol in oil which varies from 8 to 11 mass % as the fatty acid content changes from 0 to 10 mass % (Rodrigues *et. al.*, 2003).

Figure 5.2 shows a linear dependence of the solvent content in the raffinate stream in relation to the phosphorus content in the oil. In addition, it can be seen that the content of solvent in the raffinate stream is not influenced by the discs rotating speed. In the other hand the solvent content increases with the increasing of the continuous phase velocity and the acidity in the oil.

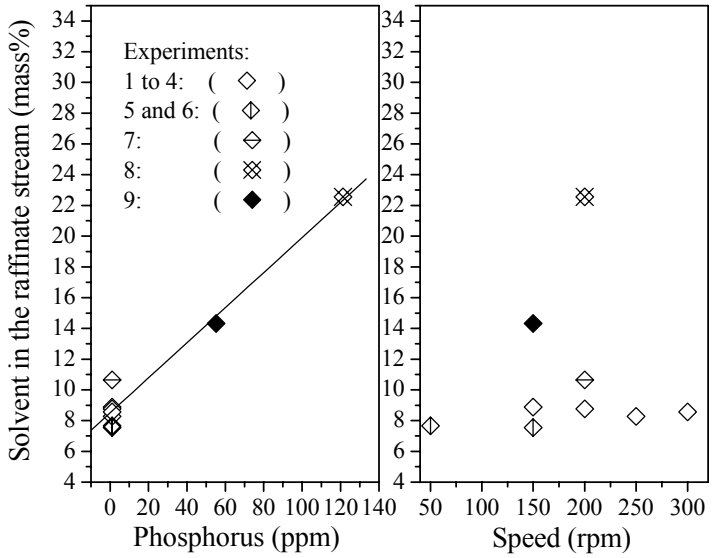

Figure 5.2. Solvent content in the raffinate stream as function of phosphorus content in the oil and of discs rotating speed

Furthermore the presence of phospholipids can cause a backflow of continuous phase, possibly due to increasing of the dispersed phase viscosity (see Table 5.3); in other words, the continuous phase is dragged by dispersed phase diminishing the mass transfer efficiency. The backflow is only one of many effects that compose the axial mixing. This phenomenon reduces the extraction efficiency, and its value, given by the coefficient of axial mixing, increases as the increase of the rotor speed. In this work the influence of the axial mixing in the mass transfer calculations were not taken into account. To include such effect in mass transfer calculations, it would be necessary to accomplish special experiments for measuring the coefficient of axial mixing for each phase as reported in detail by Coimbra *et al*. (1995).

From a technological point of view, it is also important to discuss the results of the mass transfer experiments in relation to the total amount of extracted fatty acids (T_2) and the loss of neutral oil in the extract stream (L_1). The values for T_2 and L_1 were calculated by performing mass balances for both types of fatty compounds. In the case of fatty acids, the following equation can be derived:

$$T_2 = \frac{\left[\left(F \times w_{F,2}\right) - \left(R \times w_{R,2}\right) \times 100\right]}{F \times w_{F,2}} \tag{5.6}$$

where $w_{F,2}$ and $w_{R,2}$ are the mass fractions of fatty acids in the feed and raffinate, respectively. In the same way, F and R are the total mass flow rates of the feed and raffinate.

Figure 5.3 shows the values of the total amount of extracted fatty acids (T_2) and the loss of neutral oil in the extract stream (L_1) for the experiments accomplished. The values of T_2 varied in the range of 70 to 90 %. The values increased with increasing rotor speed. The values for L_1 showed a slightly increasing with increasing of discs speed and varied in the range of 0.80 to 3.13 % (Figure 5.2).

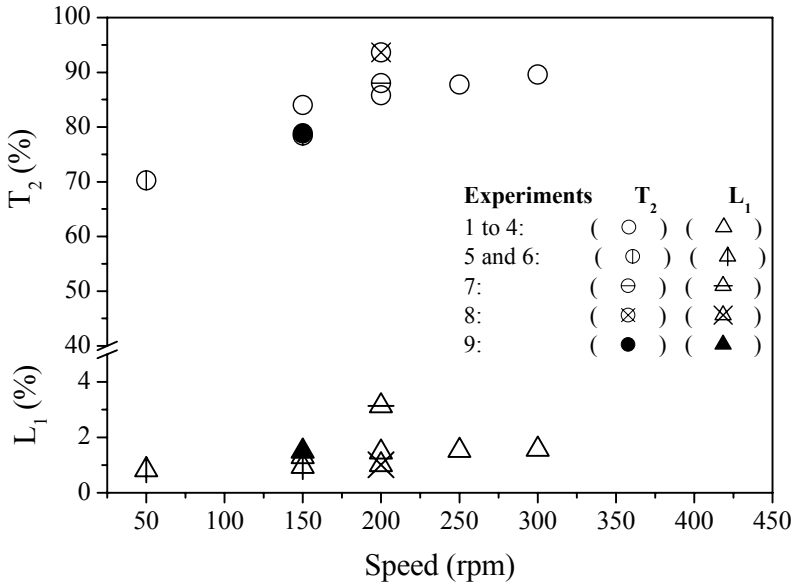

Figure 5.2. Transfer of free fatty acids and loss of neutral oil as function of discs rotating speed

It should be emphasized that these values for loss of neutral oil are significantly lower than the results reported in the literature for alkali or physical refining of rice bran oil (Orthoefer, 1996). In liquid-liquid extraction, the loss of neutral oil is largely determined by the oil solubility in the solvent. According to

the equilibrium data, the oil solubility in aqueous ethanol varies from 2.14 to 3.10 mass % as the fatty acid content changes from 0 to 12 mass %. In this case, using an oil/solvent flow ratio of 0.8, the loss of neutral oil due to its solubility in the extract stream must be approximately 3.9 %.

Since all experimental values for the loss of neutral oil were lower than or in the range of the oil solubility in the solvent, it can be concluded that the mechanical entrainment of oil phase droplets by yhe extract phase was negligible.

Table 5.5 shows the physical-chemical properties of refined, degummed and crude rice bran oils and of the degummed oil submitted to deacidification by liquid-liquid extraction (RBOd-LE). Five consecutive extraction steps with fresh solvent were necessary to obtain a rice bran oil with a low level of free fatty acids (0.16 mass % in solvent free-basis, Table 5.5).

Table 5.5– Properties of Rice Bran Oils

Characteristic	RBOr	RBOc	RBOd	RBOd-LE
Acidity Level (mass %)	5.53 [a]	7.66	8.52	0.16
Phosphorus (ppm)	< 1	121.33	55.19	43.24
DAG+MAG (mass %)	12.68	13.01	13.85	9.82
PV [b]	2.28	na [c]	14.09	12.11
Tocols (ppm)	393.60	970.30	793.93	584.70
γ-Oryzanol (mass %)	0.82	1.95	1.89	1.39
IV [d]	101.84	100.38	98.41	99.01
SV [e]	193.42	192.69	193.84	193.72

[a] commercial oleic acid ; [b] peroxide value; [c] not available; [d] iodine value;
[e] saponification value.

Figure 5.4 shows the transfers of phosphorus, diacylglycerols and monoacylglycerols during the liquid-liquid extraction process for RBOd, RBOr and RBOc. In the case of the last two oils, this evaluation was based on the level of MAG, DAG or phosphorus measured in the extract phase of one refining step (experiments 1 and 8, respectively). In the case of RBOd the extract phases of the first, second and fifth refining steps were used for the evaluation. The transfer levels were calculated according to equation 5.7.

$$T_i = \frac{\lfloor E \times w_{fatty,i} \times w_{E,fatty} \times 100 \rfloor}{F \times w_{F,i}} \tag{5.7}$$

where i is MAG, DAG or phosphorus, $w_{F,i}$ is the mass fraction of MAG, DAG or phosphorus in the feed, $w_{E,fatty}$ is the mass fraction of fatty material in the extract stream and, $w_{fatty,i}$ is the mass fraction of MAG, DAG or phosphorus in the fatty material present in the extract stream. In the same way, F and E are the total mass flow rates of the feed and extract.

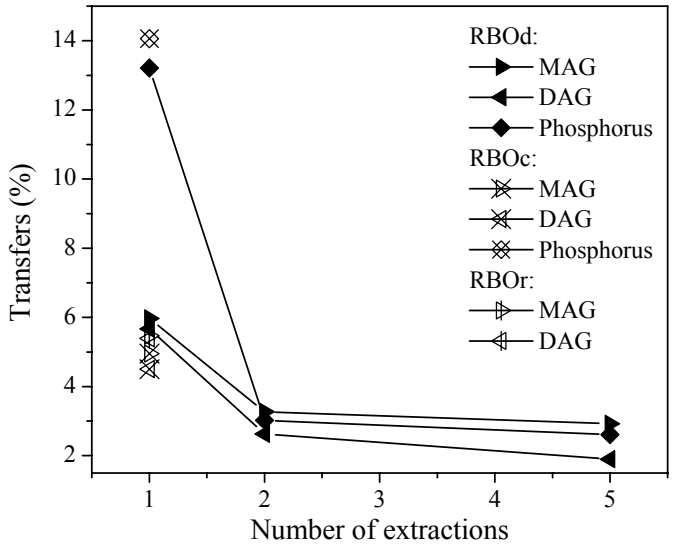

Figure 5.4. Transfer of MAG, DAG and phosphorus

It can be seen in Figure 5.4 that the contents of MAG, DAG and phosphorus on the oils were reduced during the liquid-liquid extraction process. The transfer of mono (MAG) and diacylglycerols (DAG) were very similar for RBOr, RBOc and RBOd, considering the first extract stream. In the same way, the phosphorus transfer from degummed or crude oils to the alcoholic phase was around 14 %. It can be infered that raw material characteristics do not significantly influence the extension of phosphorus, MAG and DAG transfers. On the other hand, in Figure 5.4 it can be seen that the transfers in the second and fifth refining steps were significantly lower than in the first one. Considering that in the last extraction steps

the oil fed into the column has a lower level of free fatty acids, so that less acidity is extracted to the solvent phase, it is possible that the solubility limit of the partial acylglycerols in the ethanolic solvent was attained at the given free fatty acids concentration. In a previous work Rodrigues and coworkers (2004b) showed that in liquid-liquid extraction, the losses of neutral oil and nutraceutical compounds are mainly determined by the compounds solubility in the solvent.

The impact of liquid-liquid extraction on the peroxide value (PV) of the deacidified oil (RBOd-LE) was negligible, as presented in Table 5.5. The Lovibond Color decreased from 70(Y)-14(R)-6(B) (RBOd) to 25(Y)-14(R)-8(B) (RBOd-LE). In spite of the reduction of the yellow units the deacidified oil presents a dark color showing that a bleaching step (before or after the deacidification step) and a deodorization step are necessaries to obtain a oil with a suitable color (16(Y)-3.5(R) for RBOr) and with a lower level of peroxide value (see RBOr in Table 5.5). In addition, the gel permeation HPLC analysis showed that polymeric acylglycerols were not detected in any samples studied.

Table 5.5 shows the levels of total tocopherols present in the rice bran oils. It can be seen that the loss of total tocopherols in the liquid-liquid extraction deacidification step was about 26%. Figure 5.5 shows the individual tocopherols and tocotrienols present in the rice oils, determined by HPLC.

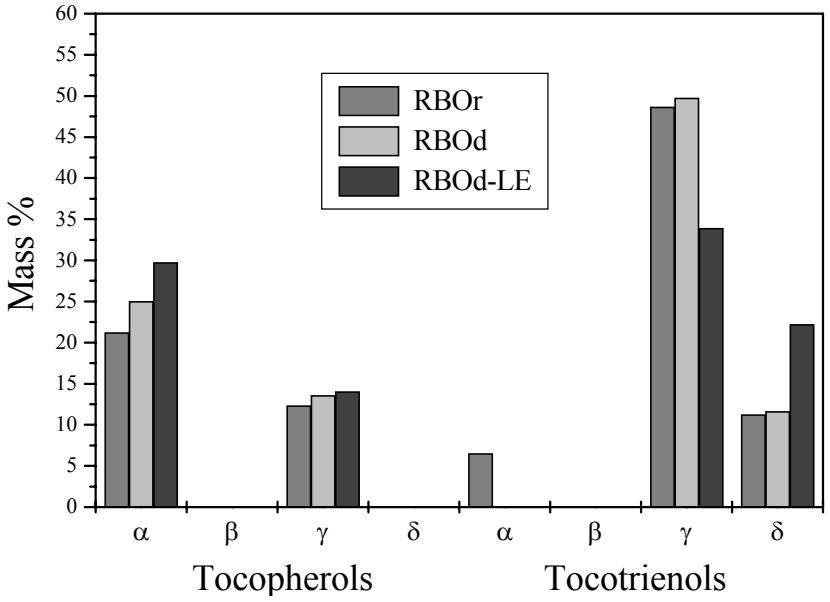

Figure 5.5. Tocopherols and Tocotrienols Composition of rice bran oils

γ-Tocotrienol was the major component, which constitued more than 49% in RBOr, 50% in RBOd, and 34% in RBOd-LE. It can be observed in Figure 5.5 that the relative proportion of individuals tocopherols and tocotrienols in RBOd-LE suffered a slighty change during the deacidification step. The concentration of the γ-tocotrienol form decreases while of the concentrations of the δ-tocotrienol and α-tocopherol forms increases. This can be due to the higher solubility of mono and dimethyltocols (β, γ and δ forms) in polar solvents, such as ethanol, than trimethyltocols (α forms). In addition, tocotrienols are more soluble than tocopherols isomers (Abidi *et al.*, 2002).

The behavior of γ-oryzanol during the deacidification step was also investigated. Table 5.5 shows that the level of this nutraceutical compound was reduced in 25% when the RBOd was submitted to deacidification by liquid-liquid extraction.

Using the fatty acid concentrations obtained after each experimental extraction step equation 5.1 gives the following numbers of ideal stages: step 1 – N=1.64; step 2 – N=1.09; step 3 – N=1.07; step 4 – N=3.40; step 5 – N=2.97. In the

case of the three first extraction steps the PRDC has a number of ideal stages in the range 1 to 1.5, a relatively lower extraction efficiency due probably to the mass transfer competition of other fatty compounds, mainly acylglycerols, that were also extracted to the solvent phase. In the last two extraction steps the nuber of ideal stages increase to a value around 3, a result that might indicate a higher extraction efficiency. Although in the last steps the transfer of acylglycerols is relatively low, the same is true for the fatty acids, since their concentration in the oil fed to the column in steps 4 and 5 is also low.

For these reasons one can not be sure if the higher number of ideal stages was caused by a low mass transfer competition between the different fatty components or is due to the higher uncertainty in the estimation of the number of ideal stages.

In fact the standard deviation of N, calculated by error propagation applied to eq. 5.1, results in a low value for the first step (ΔN=0.02) and a higher one for the last (ΔN=0.85), because the uncertainty in the experimental concentrations (0.01 mass %) have a higher influence on the low concentrations obtained in these last steps *. One should also keep in mind that eq. 5.1 is rigorously valid for a one-component mass transfer system.

Despite all these difficulties, the simulation results shown in Figure 5.5 for the crosscurrent way are very similar to the experimental ones, with a global deviation of 0.99% (calculated by eq. 5.2).

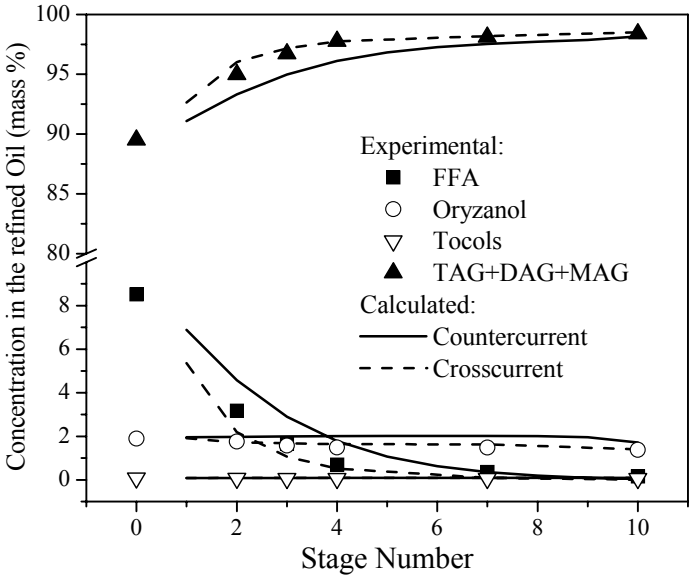

Figure 5.5. Evolution of concentrations in the deacidified rice bran oil by liquid-liquid extraction

The crosscurrent simulation was performed approximating the fractional number of ideal stages given above to the closest integer numbers. In the case of countercurrent unique extraction equipment the simulation was performed using the whole number of ideal stages (N=10) and the obtained deviation is slightly higher (1.54%). The simulation of this last configuration (countercurrent) was done for comparison purposes being the most used in industrial processing that involves liquid-liquid extraction.

* A Tabela D.2 (Anexo D) apresenta uma análise de sensibilidade e os desvios padrões para N

Figure 5.6 shows the transfers of neutral oil (TAG+DAG+MAG) and of γ-oryzanol after each experimental extraction step. The transfer levels were calculated according to equation 5.7.

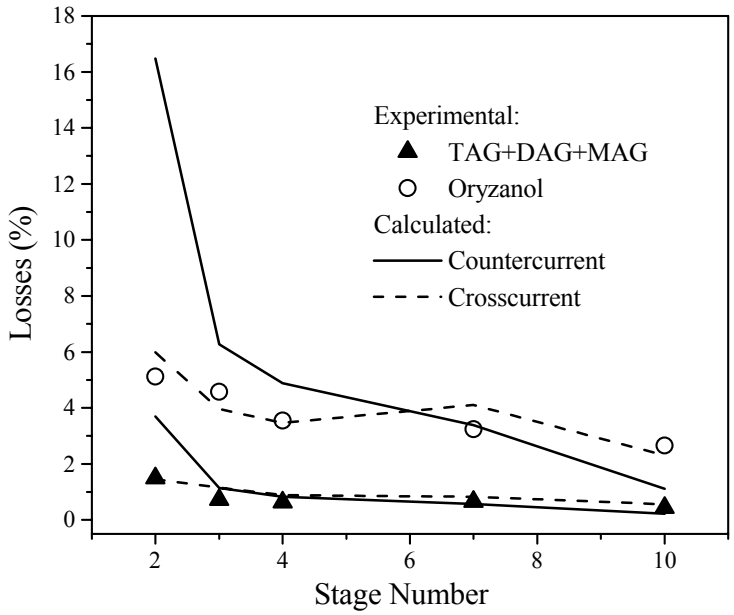

Figure 5.6. Evolution of neutral oil and of γ-oryzanol losses

The experimental global losses of neutral oil and of γ-oryzanol were 3.92 % and 19.14%. The simulation of transfers performed for crosscurrent and countercurrent configurations furnished to neutral oil loss 4.89 and 3.69 %, respectively. The losses obtained for γ-oryzanol were 19.82% for crosscurrent and 16.48% countercurrent way. The results shown in Figures 5.5 and 5.6 indicate that the losses of nutraceutical and other fatty compounds are dependent on the fatty acid level in the extract phase, because this level influences their solubility in the solvent, as already observed for MAG, DAG, and phosphorus (Figure 5.4).

In relation to iodine and saponification values the results given in Table 5.5 show that these quality indices did not sufer significant changes after liquid-liquid extraction. This information corroborates that the deacidification process by liquid-liquid extraction does not affect the fatty acid composition of the oil and preserves a good level of nutraceutical compounds.

5.6. Conclusions

The experimental results of the present work show the feasibility of the total deacidification of edible oils by solvent extraction. The losses of neutral oil and nutraceutical compounds were significantly lower than the results reported in the literature for alkali or physical refining of rice bran oil. The simulation of extraction process presented good agreement with experimental data obtained.

5.7. Acknowledgements

The authors wish to acknowledge FAPESP (Fundação de Amparo à Pesquisa do Estado de São Paulo – 99/12033-1 and 01/10137-6) and CNPq (Conselho Nacional de Desenvolvimento Científico e Tecnológico – 521011/95-7), for the financial support.

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Conclusões Gerais

Conclusões Gerais

✓ O equilíbrio líquido-líquido de sistemas graxos compostos por óleo de farelo de arroz + ácidos graxos livres + etanol + água + γ-orizanol + tocoferóis totais foi extensivamente estudado a 25 °C. Os dados experimentais obtidos apresentaram baixos desvios experimentais atestando a adequação da metodologia experimental adotada para obtenção das composições das fases alcoólica e oleosa. Os valores dos coeficientes de distribuição e seletividade, que são grandezas calculadas à partir dos dados de concentração, apresentaram uma baixa dispersão, o que reforça a boa qualidade dos dados experimentais;

✓ Os dados experimentais de equilíbrio de fases foram correlacionados pelos modelos moleculares NRTL and UNIQUAC. Foram ajustados 30 parâmetros de interação entre binários da equação NRTL (para 5 componentes do sistema graxo), e 30 parâmetros de interação entre binários da equação UNIQUAC (para 6 componentes), obtendo-se baixos desvios médios nas composições das fases (menores que 1%);

✓ Os parâmetros de interação ajustados foram testados na predição do equilíbrio de fases de diversos sistemas graxos compostos por óleo de farelo de arroz de diversas procedências (brasileiros e tailandês) e submetidos a diferentes pré-tratamentos industriais (óleos refinados, degomados e brutos). Pode-se observar que a estratégia de se ajustar os parâmetros de interação com base nos dados obtidos com sistemas graxos modelo funcionou na predição do equilíbrio de fases, para ambos modelos termodinâmicos, apesar das diferenças sutis nas composições dos óleos. Os bons resultados obtidos nas etapas de correlação e predição dos dados experimentais de equilíbrio líquido-líquido atestam que o procedimento de se considerar os sistemas compostos por pseudocomponentes foi válido. Em linhas gerais o modelo UNIQUAC apresentou um melhor desempenho preditivo do equilíbrio dos sistemas estudados;

- ✓ Observou-se que a presença de água no solvente minimiza a perda de óleo neutro, aumenta a região de extração, possibilitando o processamento de óleos com alta acidez e, minimiza a perda dos antioxidantes naturais. A presença de água possibilita ao solvente uma melhor diferenciação entre as moléculas de ácidos graxos livres, óleo neutro e nutracêuticos no momento da extração;
- ✓ A metodologia de superfície de resposta permitiu uma fácil visualização de como as principais variáveis do processo, acidez no óleo de alimentação, teor de água no solvente e razão óleo:solvente, influenciam as perdas de óleo neutro e dos compostos nutracêuticos, e a transferência dos ácidos graxos livres para a fase extrato. Os modelos gerados pela análise de superfície de resposta apresentaram bom desempenho na predição das perdas/transferências dos componentes do sistema, embora apresentem limitação em termos de extrapolação, o que não acontece com o modelo termodinâmico UNIQUAC;
- ✓ Realizou-se a desacidificação total do óleo de farelo de arroz degomado em equipamento contínuo do tipo PRDC. Esta desacidificação foi possível após 5 reprocessamentos na coluna existente no LASEFI (coluna com 1 metro de região de extração), partindo de um óleo com 8,5% de acidez livre em massa. Os cinco reprocessamentos equivaleram a cerca de 10 estágios teóricos;
- ✓ No estudo do processo de transferência de massa observou-se que o coeficiente volumétrico de transferência de massa aumentou com o aumento da velocidade de rotação dos discos perfurados, como conseqüência do maior nível de turbulência no equipamento. O valor dos coeficientes volumétricos de TM para os óleos bruto e degomado foram menores que os obtidos para os sistemas com OFA refinado devido, possivelmente, ao alto conteúdo de fósforo nestes óleos o qual, também, dificultou a separação de fases na base da coluna. Esta observação confirma a necessidade de uma degomagem efetiva do óleo antes da desacidificação por extração líquido-líquido;

- ✓ Observou-se que a transferência dos ácidos graxos livres e a perda de óleo neutro aumentam com o aumento da velocidade de rotação. As perdas de óleo neutro aumentam, também, com o aumento da acidez inicial do óleo a ser processado;
- ✓ A procedência da matéria-prima, óleo degomado, bruto ou refinado, não influenciou as transferências de mono e diacilgliceróis e de fósforo. Observou-se que o processo de extração pouco influenciou o índice de peróxido e reduziu apenas a componente amarela da cor, mostrando que uma etapa posterior de branqueamento é necessária;
- ✓ As perdas dos compostos nutracêuticos, orizanol e tocoferóis totais, foram concordantes com as perdas preditas pela metodologia de superfície de resposta. Os parâmetros de interação do modelo UNIQUAC permitiram a simulação do processo de desacidificação com baixo desvio em relação aos dados experimentais (menor que 1,5%);
- ✓ O objetivo da tese de doutorado foi plenamente atingido obtendo-se um óleo de farelo de arroz com baixo nível de ácidos graxos livres (menor que 0,30% em massa, conforme o recomendado pela legislação), com altos teores de compostos nutracêuticos (cerca de 580 ppm de tocoferóis totais e 1,3 % de γ-orizanol), teores que podem ser considerados elevados em comparação aos níveis residuais nos processos de refino tradicionais, refinos químico e físico. Ademais, o processo por extração líquido-líquido possibilitou a desacidificação a temperatura ambiente com baixa perda de óleo neutro (menor que 4%).

Sugestões para Trabalhos Futuros

Sugestões para Trabalhos Futuros

↳ Correlacionar os dados de equilíbrio utilizando outras versões dos modelos moleculares e função objetivo dependente do coeficiente de distribuição;

↳ Testar outros equipamentos de extração (colunas de recheio, por exemplo);

↳ Realizar modificações no equipamento PRDC, aumentando a região de separação de fases ou utilizando aceleradores de coalescência de gotas nestas regiões;

↳ Realizar estudo de viabilidade econômica do processo de desacidificação por extração líquido-líquido;

↳ Realizar estudo de formas de tratamento das correntes de saída do extrator, correntes de extrato e refinado.

Anexos

Anexo A

A.1. Figuras referentes a dados experimentais de equilíbrio líquido-líquido apresentados no Capítulo 2.

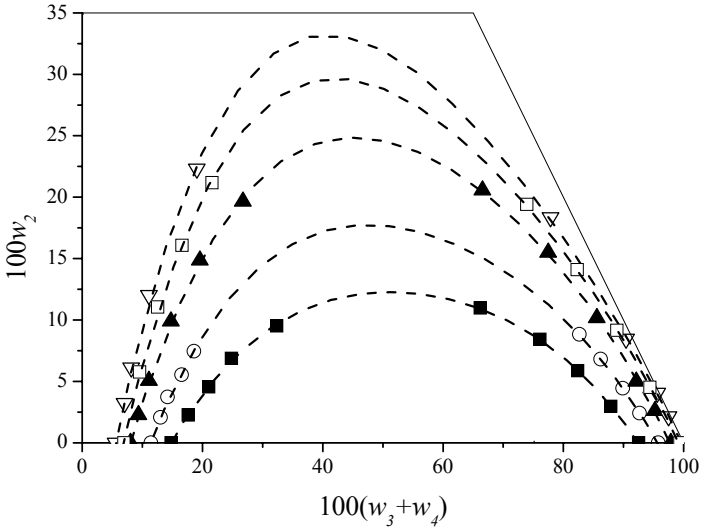

Figura A.1. Diagrama de Distribuição a 298.2±0.1 K para sistemas OFA refinado (1) + ácido oléico comercial (2) + etanol (3) + água (4):; (■) etanol anidro; (○) etanol com 2.40% de água ; (▲) etanol com 6.38% de água; (□) etanol com 10.59% de água; (▽) etanol com 12.41% de água; (- - -) ajuste polinomial de 4° grau (Ref: Tabelas 2.4 e 2.5)

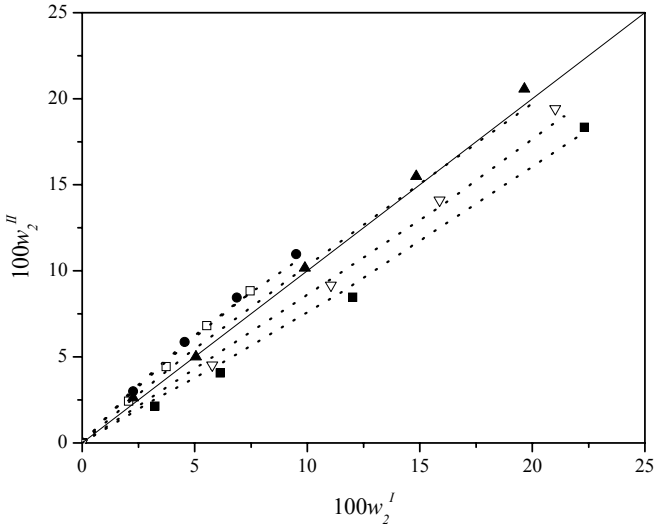

Figura A.2. Diagrama de Distribuição a 298.2±0.1 K para sistemas OFA refinado (1) + ácido oléico comercial (2) + etanol (3) + água (4): (●) etanol anidro; (□) 2.40±0.02% água; (▲) 6.38±0.02% água; (▽)10.59±0.04% água; (■)12.41±0.01% água; (····) UNIQUAC (Ref: Figura 2.4)

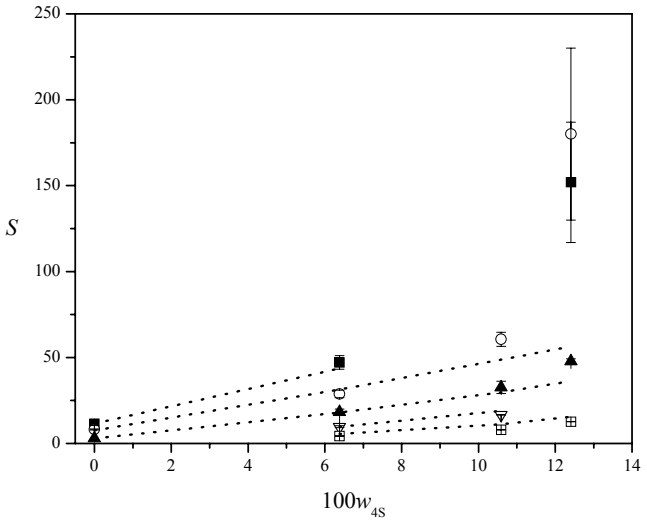

Figura A.3. Diagrama de Seletividade a 298.2±0.1 K para sistemas OFA refinado (1) + ácido oléico comercial (2) + etanol (3) + água (4): (···) UNIQUAC; (■) 5 % AGL; (○) 10 % AGL; (▲) 20 % AGL; (▽) 30 % AGL; (□) 40 % AGL (Ref: Figura 2.5)

A.2. Figuras referentes a dados experimentais de equilíbrio líquido-líquido apresentados no Capítulo 3.

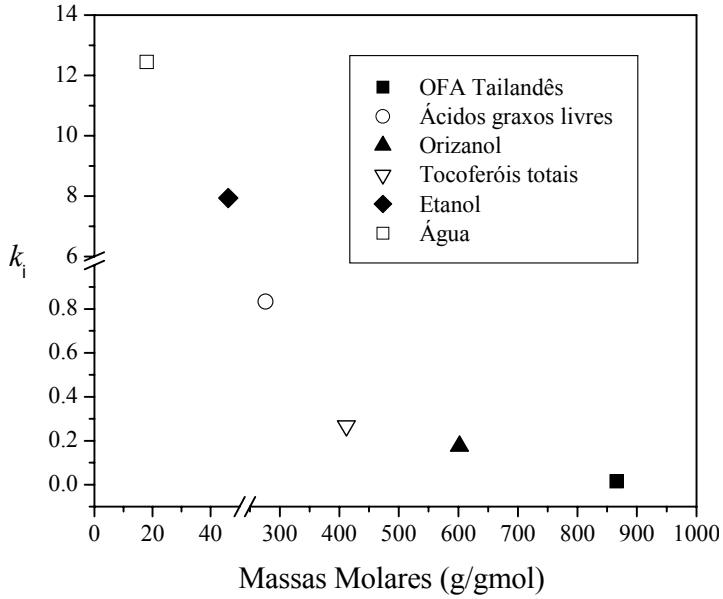

Figura A.4. Coeficientes de partição dos componentes do sistema graxo em função das massas molares (Ref: Tabelas 3.2 e 3.10 – 2ª tie- line)

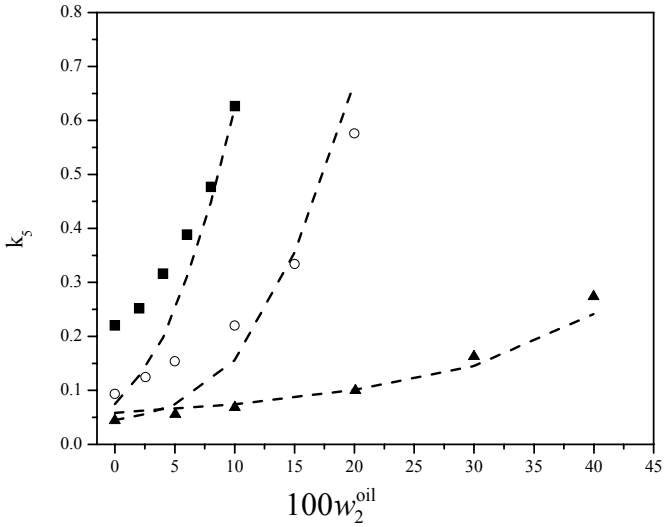

Figura A.5. Coeficiente de Distribuição do γ-orizanol para sistemas graxos modelo com cerca de $100w_5^{oil}$ = 1.5 %, at (298.2±0.1) K: (■) $100w_{4S}$ = 0 % (A); (○) $100w_{4S}$ = 6 % (C); (▲) $100w_{4S}$ = 12 % (E); (- - -) modelo NRTL (Ref: Figura 3.2)

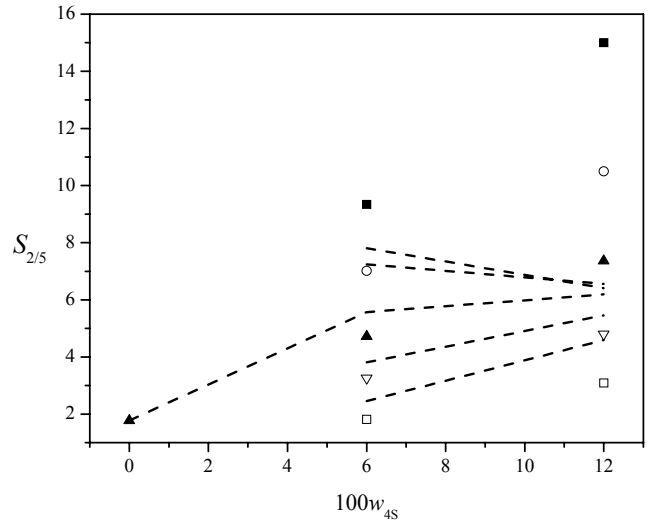

Figure A.6. Diagrama de Seletividade de ácidos graxos (2) em relação a γ-orizanol (5) a (298.2±0.1) K para sistemas graxos modelo: (- - -) NRTL; (■)$100w_2^{oil}$ = 5 % AGL; (○)$100w_2^{oil}$ = 10 % AGL; (▲)$100w_2^{oil}$ = 20 % AGL; (∇)$100w_2^{oil}$ = 30 % AGL; (□)$100w_2^{oil}$ = 40 % AGL (Ref: Figura 3.4)

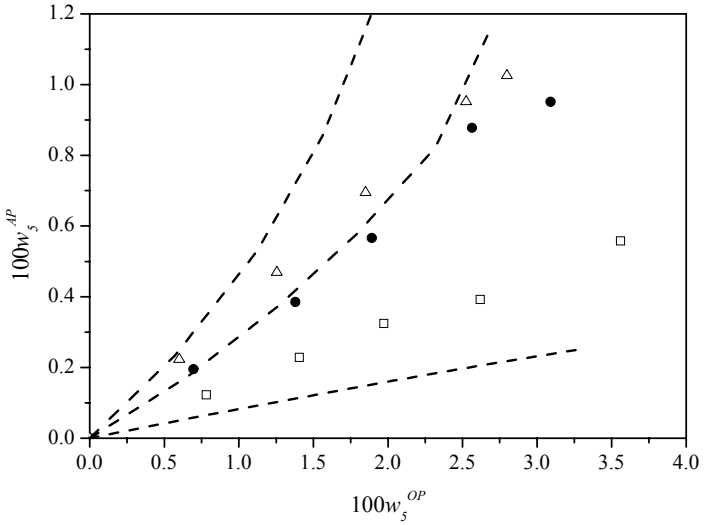

Figura A.7. Predição para sistemas modelo a (298.2±0.1) K and $100w_{4S}$ = 6 %: (□)$100w_2^{oil}$ = 10 % AGL; (●)$100w_2^{oil}$ = 25 % AGL; (△)$100w_2^{oil}$ = 30 % AGL; (- - -) NRTL (Ref: Figura 3.5)

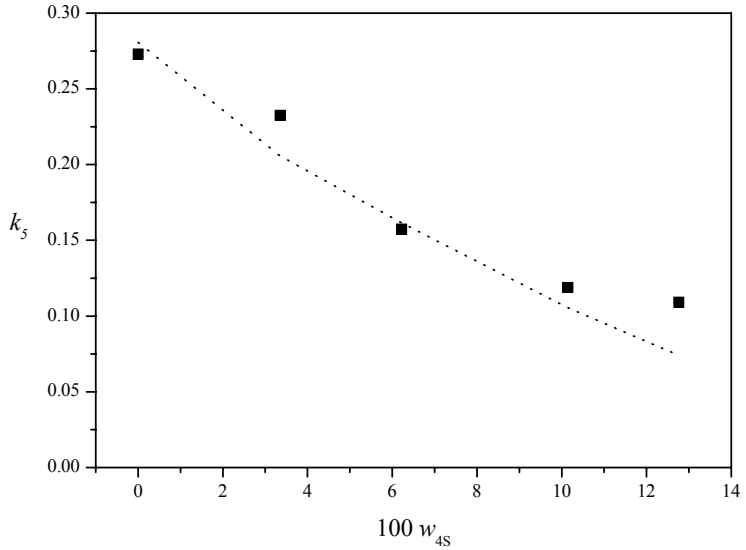

Figure A.8. Predição para sistemas reais a (298.2±0.1) K; (· · ·) modelo UNIQUAC (Ref: Tabela 3.8)

Anexo B

B.1. Coeficiente de atividade a diluição infinita

O coeficiente de atividade a diluição infinita é obtido tomando-se o limite da expressão que relaciona o coeficiente de atividade e a energia livre de Gibbs excedente (equação B.1).

$$ln\gamma_i = \frac{1}{RT}\left(\frac{\partial \underline{G}^E}{\partial n_i}\right)_{T,P,n_{j\neq i}} \tag{B.1}$$

Aplica-se o limite na expressão acima mantendo constante a quantidade de matéria dos outros componentes da mistura e fazendo a quantidade de matéria do composto minoritário tender a zero. Nota-se que, embora a atividade desse composto também se aproxime de zero, o coeficiente de atividade se aproxima de um valor definido não necessariamente nulo.

Aplicando-se o limite na expressão do modelo UNIQUAC, em frações mássicas, tem-se:

$$ln\gamma_i = ln\gamma_i^C + ln\gamma_i^R \tag{B.2}$$

$$lim_{w_i\to 0}(ln\gamma_i) = (ln\gamma_i)^{\infty} = lim_{w_i\to 0} ln\gamma_i^C + lim_{w_i\to 0} ln\gamma_i^R \tag{B.3}$$

$$lim_{w_i\to 0} ln\gamma_i^C = 1 + \frac{z}{2}\overline{M}_i q_i^{'} ln\left(\frac{q_i^{'}\sum_{j=1}^{n} r_j^{'} w_j}{r_i^{'}\sum_{j=1}^{n} q_j^{'} w_j}\right) - \frac{z}{2}\overline{M}_i q_i^{'}\left(1 - \left(\frac{r_i^{'}\sum_{j=1}^{n} q_j^{'} w_j}{q_i^{'}\sum_{j=1}^{n} r_j^{'} w_j}\right)\right) \tag{B.4}$$

$$lim_{w_i\to 0} ln\gamma_i^R = \overline{M}_i q_i^{'}\left[1 - ln\left(\sum_{j=1}^{n}\theta_j^{'}\tau_{ji}\right) - \sum_{j}^{n}\left(\theta_j^{'}\tau_{ij} \Big/ \sum_{k=1}^{n}\theta_k^{'}\tau_{kj}\right)\right] \tag{B.5}$$

$$\text{sendo: } \theta_j^{'} = \frac{q_j^{'} w_j}{\sum_{k=1}^{n} q_k^{'} w_k} \tag{B.6}$$

onde:

γ_i^C: é a contribuição combinatorial ou entrópica;

γ_i^R: é a contribuição residual ou entálpica;

θ_i': fração de área do componente *i*;

w_i: fração mássica do componente *i*;

q_i': parâmetro de área do componente *i*;

r_i': parâmetro de volume do componente *i*;

$\overline{M}_i$: massa molar do componente *i*;

τ_{ij}, τ_{ji}: parâmetros de interação entre os componentes *i* e *j*

Os parâmetros τ_{ij} e τ_{ji} são definidos como:

$$\tau_{ij} = exp - \left(\frac{u_{ij} - u_{jj}}{RT} \right) = exp - \left(\frac{A_{ij}}{T} \right) \quad \text{(B.7)}$$

$$\tau_{ji} = exp - \left(\frac{u_{ji} - u_{ii}}{RT} \right) = exp - \left(\frac{A_{ji}}{T} \right) \quad \text{(B.8)}$$

onde:

$u_{ij} - u_{jj}$: diferença das energias de interação entre duas moléculas distintas *i* e *j* e duas moléculas da mesma espécie *jj*;

R: constante dos gases;

T: temperatura absoluta;

A_{ij}, A_{ji}: parâmetros de interação ajustáveis aos dados experimentais.

Pode-se observar na equação B.4 que a contribuição da parte combinatorial corresponde a componente entrópica do coeficiente de partição e este termo é idêntico ao componente entrópico do sistema livre do componente minoritário. O termo entrópico não é influenciado pelas interações entre os vários componentes do sistema, mas somente pela probabilidade de sua distribuição espacial.

A equação B.5, que mostra a componente entálpica, inclui somente os parâmetros de interação entre a molécula minoritária e os demais componentes do sistema, isto é, constitui o termo responsável pelo cálculo das interações energéticas favoráveis (ou desfavoráveis) entre a molécula minoritária e os demais compostos do sistema graxo.

A condição de equilíbrio para a molécula minoritária distribuída em duas fases pode ser escrita como:

$$a_i^{\ I} = a_i^{\ II} \Rightarrow w_i^{\ I}\gamma_i^{I} = w_i^{\ II}\gamma_i^{II} \qquad \text{(B.9)}$$

onde a_i: atividade do composto i na fase I ou II

Definindo-se o coeficiente de partição como a razão entre as concentrações:

$$k_i = \frac{w_i^{\ II}}{w_i^{\ I}} \qquad \text{(B.10)}$$

Substituindo-se o coeficiente de atividade na equação (B.9) pelo coeficiente de atividade a diluição infinita, obtém-se a expressão que calcula a partição ou o coeficiente de distribuição da molécula minoritária i a diluição infinita:

$$k_i^{\infty} = \frac{(\gamma_i^{\ II})^{\infty}}{(\gamma_i^{\ I})^{\infty}} \qquad \text{(B.11)}$$

Anexo C

C.1. Dados de equilíbrio líquido-líquido (Capítulo 4).

Os dados de equilíbrio líquido-líquido apresentados na Tabela C.1 geraram os dados de transferências/perdas, para os sistemas modelo, apresentados na Tabela 4.4. As variações dos parâmetros acidez e teor de água no solvente foram recomendadas pelo planejamento experimental 2^2.

Tabela C.1 – Dados de equilíbrio líquido-líquido para o sistema óleo de farelo de arroz refinado (1) + Ácido oléico comercial (2) + solvente etanol aquoso [etanol (3) + água (4)] + γ-orizanol (5) a (298.2±0.1) K

Sistema	Ponto de mistura				Fase Oleosa (FO)				Fase Alcoólica (FA)			
	$100w_1$	$100w_2$	$100w_{3+4}$	$100w_5$	$100w_1$	$100w_2$	$100w_{3+4}$	$100w_5$	$100w_1$	$100w_2$	$100w_{3+4}$	$100w_5$
Pontos Centrais	44.21	5.03	50.01	0.76	83.39	5.06	10.21	1.34	1.60	5.01	93.24	0.16
	44.19	5.03	50.01	0.76	83.47	5.18	10.06	1.29	1.77	4.84	93.24	0.15
	44.18	5.03	50.03	0.76	83.35	5.16	10.25	1.24	1.76	4.91	93.19	0.15
	44.19	5.03	50.02	0.76	83.18	5.15	10.38	1.30	1.77	5.04	93.04	0.15
2^2	46.67	2.50	50.02	0.82	86.12	2.44	10.07	1.37	2.19	2.56	95.10	0.15
	46.68	2.50	50.00	0.82	88.43	3.02	7.09	1.46	0.93	1.94	97.06	0.08
	41.76	7.50	50.02	0.73	77.99	7.25	13.51	1.25	3.08	7.76	88.96	0.20
	41.74	7.50	50.04	0.73	80.10	8.74	9.87	1.29	0.99	6.17	92.71	0.13
Pontos Axiais	47.68	1.46	50.00	0.86	88.81	1.32	8.40	1.47	1.06	1.66	97.17	0.12
	40.72	8.53	50.02	0.74	76.97	8.87	12.89	1.27	2.20	8.19	89.42	0.19
	44.21	5.00	49.99	0.80	81.41	4.67	12.63	1.29	3.80	5.34	90.65	0.21
	44.13	5.06	50.02	0.80	84.11	5.98	8.60	1.32	0.55	4.06	95.33	0.06

Anexo D

D.1. Composição das correntes referentes aos experimentos de desacidificação em PRDC (Capítulo 5).

Tabela D.1 – Composição das correntes nos experimentos em PRDC

		Fase Refinado			Fase Extrato		
Exp.	**tempo**	**Ácido**	**Solvente**	**Óleo**	**Ácido**	**Solvente**	**Óleo**
1	0 *	5,5300	0,0000	94,4700	0,0000	100,0000	0,0000
	60	1,7800	8,6100	89,6100	3,0600	94,7600	2,1800
	75	2,0600	8,8500	89,0900	3,2000	94,6100	2,1900
	90	2,0800	9,1000	88,8200	3,5200	94,5500	1,9300
	105	2,0700	8,9300	89,0000	3,3900	94,3100	2,3000
	120	2,2800	8,9100	88,8100	3,4200	94,3700	2,2100
2	0	5,5300	0,0000	94,4700	0,0000	100,0000	0,0000
	60	1,9000	8,9900	89,1100	3,1200	94,4600	2,4200
	75	1,8900	8,7000	89,4100	3,2000	94,4100	2,3900
	90	1,7700	8,7800	89,4500	3,0600	94,6200	2,3200
	105	1,8000	8,5800	89,6200	3,0800	94,5900	2,3300
	120	1,8700	8,8000	89,3300	3,0300	94,5900	2,3800
3	0	5,5300	0,0000	94,4700	0,0000	100,0000	0,0000
	60	1,7000	8,5400	89,7600	2,8600	94,7300	2,4100
	75	1,8300	8,7300	89,4400	2,9300	94,6800	2,3900
	90	1,6600	7,9000	90,4400	2,8600	94,5700	2,5700
	105	1,6000	8,0800	90,3200	2,9600	94,7300	2,3100
	120	1,6000	8,1000	90,3000	2,8800	94,7300	2,3900
4	0	5,5300	0,0000	94,4700	0,0000	100,0000	0,0000
	60	1,5000	8,6991	89,8009	2,7100	95,2908	1,9992
	75	1,5400	8,6667	89,7933	2,6300	95,2538	2,1162
	90	1,4700	8,5200	90,0100	2,6500	95,3205	2,0295
	105	1,5400	8,5083	89,9517	2,6800	95,2203	2,0997
	120	1,3700	8,4392	90,1908	2,5200	95,0112	2,4688
5	0	6,4200	0,0000	93,5800	0,0000	100,0000	0,0000
	120	2,9300	7,7700	89,3000	3,5400	94,8600	1,6000
	135	3,0200	7,9000	89,0800	3,3300	94,8200	1,8500
	150	3,1800	7,8400	88,9800	3,5900	94,6400	1,7700
	165	3,2100	7,7400	89,0500	3,3400	94,5300	2,1300
	180	3,1200	8,0400	88,8400	3,4900	94,6000	1,9100
	média	3,1600	7,6600	89,1800	3,4400	94,6900	1,8700

* O tempo 0 (zero) corresponde à alimentação.

Tabela D.1 – Composição das correntes nos experimentos em PRDC (cont.)

		Fase Refinado			**Fase Extrato**		
Exp.	**tempo**	**Ácido**	**Solvente**	**Óleo**	**Ácido**	**Solvente**	**Óleo**
6	0 *	6,4200	0,0000	93,5800	0,0000	100,0000	0,0000
	105	2,5900	8,8500	88,5600	4,1600	94,6100	1,2300
	120	2,6500	9,1000	88,2500	4,0000	94,5500	1,4500
	135	2,6000	8,9300	88,4700	3,9900	94,3100	1,7000
	150	2,6000	8,9100	88,4900	4,2800	94,3700	1,3500
	média	2,7300	7,5400	89,7300	4,0900	93,7800	2,1300
		Fase Refinado			**Fase Extrato**		
Exp.	**tempo**	**Ácido**	**Solvente**	**Óleo**	**Ácido**	**Solvente**	**Óleo**
7	0 *	11,6700	0,0000	88,3300	0,0000	100,0000	0,0000
	120	3,4200	9,8100	86,7700	8,1200	86,9800	4,9000
	135	3,4600	10,6500	85,8900	8,2700	86,7800	4,9500
	150	3,2300	10,6400	86,1300	8,4600	78,3900	13,1500
	165	3,3700	10,8200	85,8100	8,6900	74,4200	16,8900
	180	3,2900	10,7400	85,9700	8,8100	69,7200	21,4700
	média	3,9700	10,6400	85,3900	8,6400	80,1700	11,1900

* O tempo 0 (zero) corresponde à alimentação.

Tabela D.1 – Composição das correntes nos experimentos em PRDC (cont.)

		Fase Refinado				Fase Extrato			
Exp.	**Tempo (min.)**	**Ácido**	**Solvente**	**Óleo**	**Orizanol**	**Ácido**	**Solvente**	**Óleo**	**Orizanol**
8	0 *	7,6600	0,0000	90,4800	1,8600	0,0000	100,0000	0,0000	0,0000
	70	1,8800	27,5700	69,1142	1,4358	3,3300	94,2500	2,2042	0,2158
	120	2,0400	27,5700	69,0745	1,3155	3,7900	94,2500	1,7002	0,2598
	150	1,8500	21,5000	75,2561	1,3939	3,7000	94,0100	2,0337	0,2563
	180	1,8400	26,8600	70,0400	1,2600	3,7100	93,9800	2,0539	0,2561
	média	1,9700	22,5600	74,0986	1,3714	3,8700	94,1200	1,7676	0,2424

* O tempo 0 (zero) corresponde à alimentação.

Tabela D.1 – Composição das correntes nos experimentos em PRDC (cont.)

		Fase Refinado				Fase Extrato			
Exp.	**Tempo (min.)**	**Ácido**	**Solvente**	**Óleo**	**Orizanol**	**Ácido**	**Solvente**	**Óleo**	**Orizanol**
9	0 *	8,5200	0,0000	89,5900	1,8900	0,0000	100,0000	0,0000	0,0000
Etapa1	média	2,7700	10,4244	85,3556	1,4500	5,1900	90,9700	3,8400	0,2500
	0 *	2,7700	0,0000	86,8056	1,4500	0,0000	100,0000	0,0000	0,0000
Etapa 2	média	1,4100	12,2175	85,0425	1,3300	1,5200	97,2100	1,2700	0,1800
	0 *	1,4100	0,0000	85,0425	1,3300	0,0000	100,0000	0,0000	0,0000
Etapa 3	média	0,5900	14,1918	84,0182	1,2000	0,7600	97,8500	1,3900	0,1300
	0 *	0,5900	0,0000	84,0182	1,2000	0,0000	100,0000	0,0000	0,0000
Etapa 4	média	0,3000	15,3017	83,2083	1,1900	0,6500	97,8900	1,4600	0,1100
	0 *	0,3000	0,0000	83,2083	1,1900	0,0000	100,0000	0,0000	0,0000
Etapa 5	média	0,1400	16,1143	82,5957	1,1500	0,2706	98,7302	0,9992	0,0900

* O tempo 0 (zero) corresponde à alimentação.

D.2. Figuras referentes aos experimentos de desacidificação em coluna de discos rotativos perfurados (Capítulo 5).

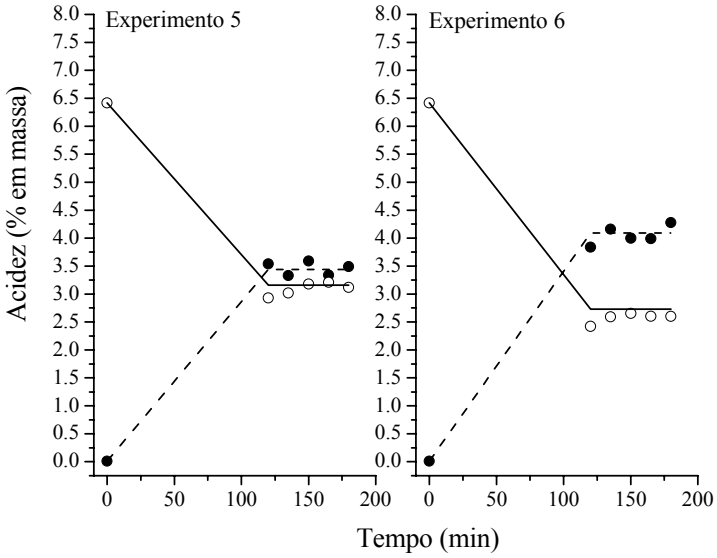

Figura D.1. Variação da concentração de ácido oléico no extrato e no refindo com o tempo em PRDC; (○) concentração pontual no refinado; (●) concentração pontual no extrato; (—) concentração média no refinado; (- - -) concentração média no extrato.

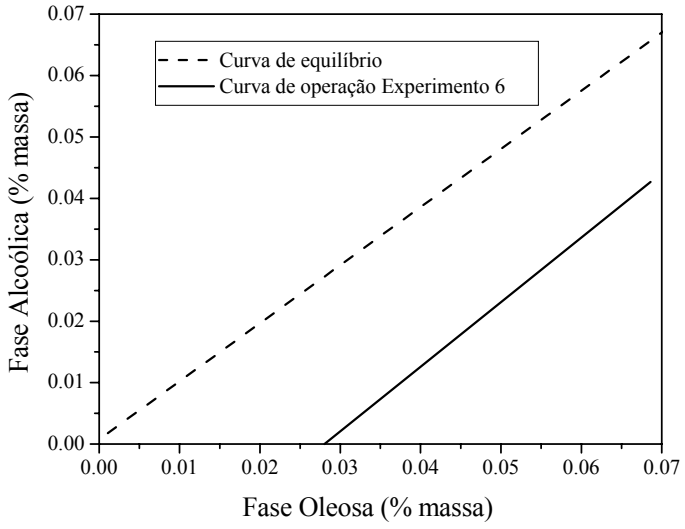

Figura D.2. Curva de equilíbrio e linha de operação para o experimento 6.

D.3. Balanço de Massa

Com os resultados das análises de acidez (ácidos graxos livres) e teor de solvente, calculou-se o balanço de massa para cada processo, a fim de se monitorar as transferências de ácido, óleo e solvente.

As equações do balanço de massa são:

Balanço de massa global:

$$M_{(1+2)}^{alim} + M_{(3+4)}^{alim} = M^{ref} + M^{ext} \quad \text{(D.1)}$$

Balanço de massa por componente:

(componente óleo)

$$M_{(1+2)} \cdot w_1^{\,alim} = M^{ref} \cdot w_1^{\,ref} + M^{ext} \cdot w_1^{\,ext} \quad \text{(D.2)}$$

(componente ácido)

$$M_{(1+2)} \cdot w_2^{\,alim} + M_{(3+4)} \cdot w_2^{\,alim} = M^{ref} \cdot w_2^{\,ref} + M^{ext} \cdot w_2^{\,ext} \quad \text{(D.3)}$$

(componente solvente)

$$M_{(3+4)}^{alim} = M^{ref} \cdot w_{(3+4)}^{\,ref} + M^{ext} \cdot w_{(3+4)}^{\,ext} \quad \text{(D.4)}$$

D.4. Cálculo do número de estágios teóricos.

Foi realizada uma análise de sensibilidade para a equação 5.1 considerando um desvio de 0,01% na composição do ácido. Além disso, o desvio padrão do valor do N calculado foi estimado pelo emprego de propagação de erro na equação 5.1. A Tabela D.2 apresenta os resultados obtidos.

Tabela D.2 Teste de sensibilidade e desvio padrão para Número de estágios teóricos (N)

Etapa de refino	Valor estimado	Teste de sensibilidade	Desvio Padrão
1	1,64	1,57 < N < 1,70	0,020
2	1,09	0,79 < N < 1,49	0,082
3	1,07	0,54 < N < 2,14	0,10
4	3,40	2,76 < N < 4,21	0,18
5	2,97	1,91 < N < 4,78	0,84

D.5. Erro relativo no balanço de massa.

Através do balanço de massa, foram calculados os erros relativos para o ácido, óleo e solvente, por meio da comparação das massas destes componentes nas correntes de saída e entrada. Este dado avaliou a eficiência de controle de processo da coluna.

Os erros foram calculados segundo as equações:

$$\%\,Erro\,\acute{a}cido = \frac{\left(\left|M_{\acute{a}cido}^{alim} - M_{\acute{a}cido}^{ext+ref}\right|\right)\times 100}{M_{\acute{a}cido}^{alim}} \quad \text{(D.5)}$$

$$\%\,Erro\,\acute{o}leo = \frac{\left(\left|M_{\acute{o}leo}^{alim} - M_{\acute{o}leo}^{ext+ref}\right|\right)\times 100}{M_{\acute{o}leo}^{alim}} \quad \text{(D.6)}$$

$$\%\,Erro\,solvente = \frac{\left(\left|M_{solvente}^{alim} - M_{solvente}^{ext+ref}\right|\right)\times 100}{M_{solvente}^{alim}} \quad \text{(D.7)}$$

Com relação aos balanços de massa, observa-se que os maiores erros são obtidos para o balanço de ácido graxo. O motivo para isso é a baixa vazão e concentração deste componente no equipamento. Já para o solvente e o óleo, componentes com altas vazões no sistema, os erros são menores. No caso do balanço de ácidos graxos observa-se a seguinte distribuição de erros: em 25 % dos experimentos realizados (total 8) este erro foi menor que 5 %, em 50 % dos casos ficou entre 5 % e 10 %, em 12,5 % dos casos esteve entre 10 e 15 %, e somente para 12,5 % dos experimentos foi maior que 15 %. Deve-se observar que os erros no balanço de massa expressam erros obtidos em diversas medidas experimentais, a saber, as vazões de entrada e saída, assim como as composições de todas as correntes. O impacto deste acúmulo de erros é mais expressivo para o componente presente em menor quantidade, no caso o ácido graxo. Mas na grande maioria dos experimentos (87,5%) este erro manteve-se dentro de limites aceitáveis, abaixo de 15 %. A Tabela D.3 apresenta os valores dos erros no balanço de massa para alguns dos experimentos realizados.

Tabela D.3 Erros no Balanço de massa para alguns dos experimentos realizados

Experimento	Rotação (rpm)	Erro no Balanço de massa (% mássica)		
		óleo	**ácido**	**solvente**
1	150	1,80	21,05	-2,03
2	200	0,49	3,44	-11,26
3	250	-3,21	5,05	1,92
4	300	-8,08	4,21	-7,04
5	50	3,70	-6,50	-5,35
6	150	-3,21	-8,91	-6,19
7	200	-19,05	-12,34	-9,39
9	150	8,37	-9,04	-21,13

Anexo E

E.1. Estrutura molecular de um Triacilglicerol

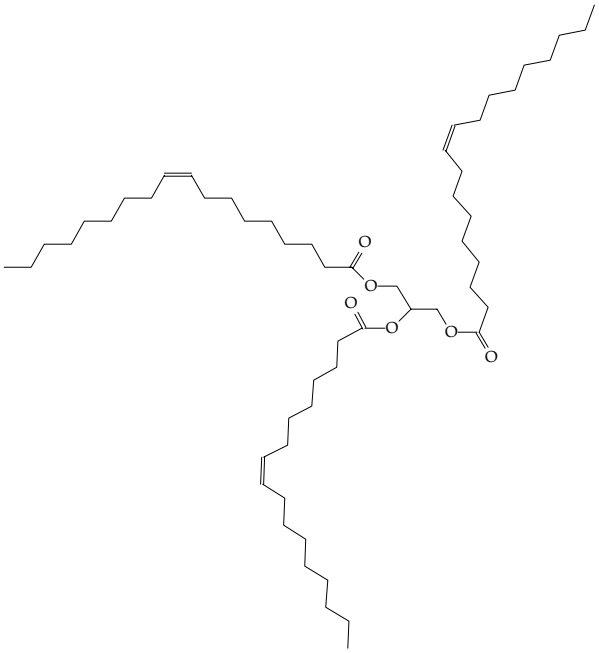

Figura E.1. Estrutura Molecular da Trioleína.

E.2. Estrutura molecular de um Ácido Graxo

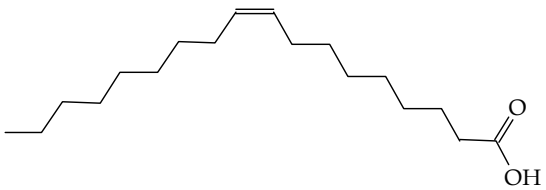

Figura E.2. Estrutura Molecular do Ácido Oléico.

E.3. Estrutura molecular do Orizanol

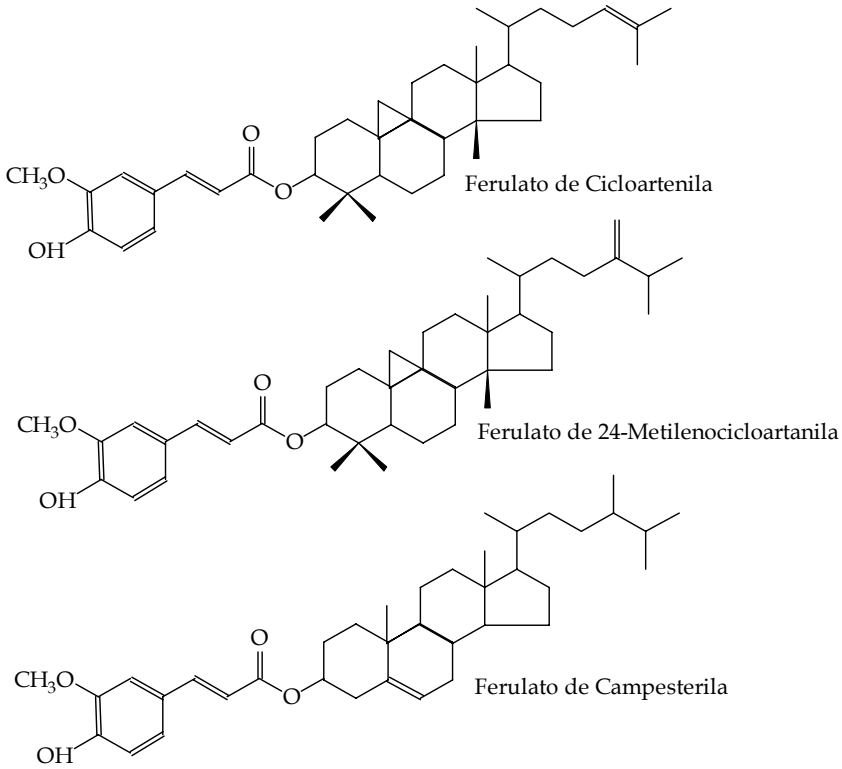

Figura E.3. Estruturas Moleculares de alguns dos componentes do γ-Orizanol.

E.4. Estruturas moleculares dos Tocoferóis e Tocotrienóis

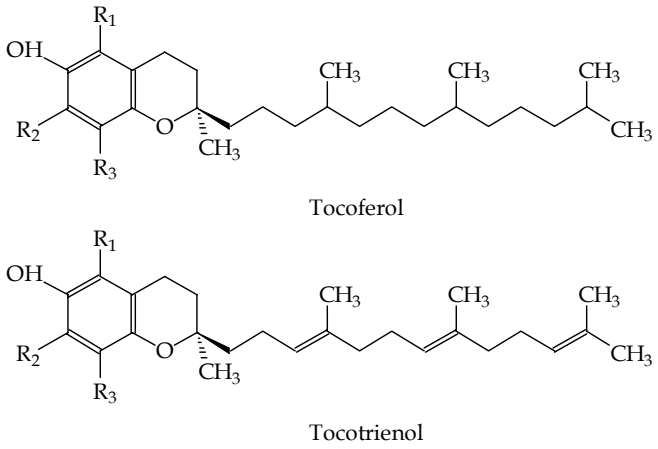

Figura E.4. Estruturas Moleculares do Tocoferol e Tocotrienol.
(Os substituintes dos anéis aromáticos (R_1, R_2, R_3) estão especificados na Tabela E.1).

Tabela E.1 - Substituintes dos anéis aromáticos nas famílias de Tocoferóis e Tocotrienóis

	R_1	**R_2**	**R_3**
α	Me[a]	Me	Me
β	Me	H	Me
γ	H[b]	Me	Me
δ	H	H	Me

[a] Me: metil; [b] H: hidrogênio.