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**FORMULATION OF INTRAVENOUS LIPID EMULSION CONTAINING  
OIL-SOLUBLE VITAMINS**



**Miss Nuntana Candido**

**A Thesis Submitted in Partial Fulfillment of the Requirements  
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Program in Pharmaceutical Technology (International)**

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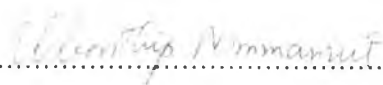
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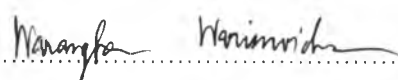
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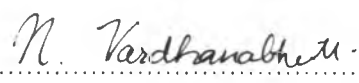
  
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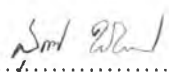
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ฉันทนา คันดิโต : การตั้งตำรับอิมัลชันเสริมวิตามินชนิดละลายในไขมันเพื่อให้ทางหลอดเลือดดำ (FORMULATION OF INTRAVENOUS LIPID EMULSION CONTAINING OIL-SOLUBLE VITAMINS) อ. ที่ปรึกษา: อ. ดร. วรางคณา วารีสน้อยเจริญ, อ. ที่ปรึกษาร่วม: ดร. นงนุช พงศ์เจริญเกียรติ. 254 หน้า. ISBN 974-17-0728-2

การบริหารยาทางหลอดเลือดของวิตามินชนิดละลายในไขมันสามารถทำได้ในรูปของอิมัลชันชนิดน้ำมันในน้ำ ปัจจัยที่ส่งผลกระทบต่อคุณสมบัติทางเคมีฟิสิกส์ของอิมัลชันไขมันได้ถูกศึกษา อันได้แก่ จำนวนรอบในการปั่นผสม, ส่วนประกอบของอิมัลชัน รวมทั้งวิธีการทำให้ปราศจากเชื้อ ส่วนผสมของไตรกลีเซอไรด์ชนิดสายโมเลกุลปานกลาง (โมกลีออล 812) และไตรกลีเซอไรด์ชนิดสายโมเลกุลยาว (น้ำมันถั่วเหลือง) หรือน้ำมันถั่วเหลืองเพียงชนิดเดียว ถูกใช้เป็นวัฏภาคน้ำมันในการเตรียมอิมัลชันไขมันที่ความเข้มข้นร้อยละ 10 หรือ 20 สารอิมัลซิฟายเออร์หลักที่ใช้ได้แก่ เลซิทีนจากถั่วเหลือง หรือเลซิทีนจากไข่ สารอิมัลซิฟายเออร์ร่วมที่ใช้ได้แก่ ฟอสฟาติลกลีเซอรอล, สเตียร์วอเอมีน และทวิน 80 โดยใช้สารอิมัลซิฟายเออร์หลักเพียงตัวเดียวหรือ ใช้ผสมร่วมกับสารอิมัลซิฟายเออร์ร่วม หรือส่วนผสมของสารอิมัลซิฟายเออร์ร่วม จำนวนรอบของการปั่นผสมด้วยเครื่องปั่นผสมแรงดันสูงที่ใช้ในการเตรียมได้ถูกปรับเปลี่ยน (3, 5, 7 หรือ 10 รอบ) การศึกษาแสดงให้เห็นว่า การปั่นผสมจำนวน 10 รอบ จะทำให้ได้ขนาดอนุภาคที่มีขนาดเล็กที่สุด ซึ่งมีค่าประมาณ 0.2 ไมโครเมตร อิมัลชันไขมันที่เตรียมขึ้นโดยใช้น้ำมันถั่วเหลืองร้อยละ 10 โดยใช้ส่วนผสมของสารอิมัลซิฟายเออร์ คือ เลซิทีนจากไข่ (หรือเลซิทีนจากถั่วเหลือง) ทวิน 80 และ สเตียร์วอเอมีน มีความคงตัวที่ดีที่สุดเมื่อเก็บในตู้เย็นเป็นระยะเวลา 4 เดือน ขนาดอนุภาคโดยเฉลี่ยของตำรับดังกล่าวที่ผ่านการทำให้ไร้เชื้อโดยใช้หม้อนึ่งอັดไอน้ำ มีค่าประมาณ 0.2 ไมโครเมตร ความเป็นกรด-ด่างของอิมัลชันมีค่าโดยประมาณที่เป็นกลาง อิมัลชันที่เตรียมขึ้นโดยใช้สารอิมัลซิฟายเออร์ คือ เลซิทีนจากไข่ ร่วมกับทวิน 80 และสเตียร์วอเอมีนแสดงค่าความเป็นบวกของค่าต่างศักย์ที่ผิวของอนุภาคที่สูงกว่าอิมัลชันที่เตรียมขึ้นโดยใช้สารอิมัลซิฟายเออร์ คือ เลซิทีนจากถั่วเหลืองร่วมกับ ทวิน 80 และสเตียร์วอเอมีน ดังนั้นจึงถูกนำมาใช้ในการบรรจุวิตามินได้แก่ วิตามินเอ ปาล์มมิเตท, ดีสาม, อี อะซีเตท และเคหนึ่ง

อิมัลชันที่เตรียมขึ้นโดยใช้เลซิทีนจากไข่ ร่วมกับทวิน 80 สเตียร์วอเอมีน และวิตามินมีคุณสมบัติทางเคมีฟิสิกส์คล้ายกับอิมัลชันที่เตรียมโดยไม่ใส่วิตามิน หลังการเก็บเป็นเวลา 1 เดือนค่าความต่างศักย์ที่ผิวของอนุภาคของตำรับที่ถูกทำให้ไร้เชื้อโดยใช้หม้อนึ่งอັดไอน้ำและเตรียมโดยใช้เลซิทีนจากไข่ ร่วมกับทวิน 80 สเตียร์วอเอมีน และวิตามินมีค่า 14.21 มิลลิโวลท์ และมีขนาดอนุภาคเท่ากับ 0.2 ไมโครเมตร ค่าความเป็นกรด-ด่างของทุกตำรับมีค่าโดยประมาณที่เป็นกลาง ค่าออสโมแลติคของอิมัลชันมีค่าประมาณ 300 มิลลิออสโมลต่อกิโลกรัม ทำการเปรียบเทียบวิธีการทำให้ปราศจากเชื้อ คือการกรอง และการใช้หม้อนึ่งอັดไอน้ำพบว่าทั้งสองวิธีมีผลกระทบต่อปริมาณวิตามินที่เหลืออยู่ในอิมัลชันอย่างไม่มีนัยสำคัญ ( $P > 0.05$ ) พบการสูญเสียของปริมาณวิตามินชนิดละลายในไขมันในอิมัลชัน ซึ่งจะขึ้นกับชนิดของวิตามินและระยะเวลาการเก็บ วิตามินอี อะซีเตท เป็นวิตามินที่มีความคงตัวมากที่สุดเมื่อเทียบกับวิตามินตัวอื่นๆ จากการทดลองทั้งหมดสรุปได้ว่าความคงตัวของอิมัลชันที่ประกอบด้วยเลซิทีนจากไข่สามารถเพิ่มขึ้นได้โดยการใช้ส่วนผสมของสารลดแรงตึงผิวชนิดไม่มีประจุ (ทวิน 80) และ สารที่มีประจุบวก (สเตียร์วอเอมีน) โดยอิมัลชันที่เตรียมได้มีคุณสมบัติที่เหมาะสมสำหรับการนำส่งวิตามินชนิดละลายในไขมันทางหลอดเลือดดำ

ภาควิชา.....  
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ลายมือชื่ออาจารย์ที่ปรึกษาร่วม.....

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KEY WORD: LIPID EMULSION / OIL-SOLUBLE VITAMINS / PHYSICAL STABILITY /  
PHYSICOCHEMICAL PROPERTIES

NUNTANA CANDIDO: FORMULATION OF INTRAVENOUS LIPID EMULSION CONTAINING  
OIL-SOLUBLE VITAMINS. THESIS ADVISOR: DR. WARANGKANA WARISNOICHAROEN,  
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The oil-soluble vitamins could be parenterally administered via oil-in-water emulsion. Factors affecting the physicochemical properties of lipid emulsion were investigated, i.e., cycles of homogenization, emulsion compositions, including sterilization method. The combination of medium chain triglycerides (Miglyol 812) and long chain triglycerides (soybean oil) or only soybean oil was used as oil phase in lipid emulsion preparation at concentration of 10% or 20%. The main emulsifiers used were soy lecithin (SPC) or egg lecithin (EPC). The co-emulsifiers used were phosphatidylglycerol (PG), stearylamine (SA) and Tween 80 (T80). The main emulsifier was used either alone or in combination with co-emulsifier or a mixture of co-emulsifiers. The methods of preparation were varied in number of cycle times of emulsion through the high pressure homogenizer (3, 5, 7 or 10 cycles). The studies revealed that 10 cycles of homogenization provided the smallest droplet size, which was approximately 0.2  $\mu$  m. Lipid emulsions formulated using 10% soybean oil, stabilized by the combination of EPC (or SPC), T80 and SA showed the best stability upon 4-month storage in a refrigerator. The mean droplet sizes of such autoclaved formulations were approximately 0.2  $\mu$ m. The pH values of emulsions were approximately neutral. Emulsion using EPC+T80+SA exhibited higher positive zeta potential than that found in emulsion using SPC+T80+SA, hence it was used to incorporate vitamins (V) that were vitamins A palmitate, D<sub>3</sub>, E acetate and K<sub>1</sub>.

Emulsion containing EPC+T80+SA+V had the similar physicochemical properties to prepared emulsion without vitamins. After 1-month storage, zeta potential of autoclaved formulations using EPC+T80+SA+V were 14.21 mV and the particle size was 0.2  $\mu$ m. The pH of the formulation was approximately neutral and the osmolality was approximately 300 mOsm/kg. Comparison between methods of sterilization, filtration and autoclaving, was found that they insignificantly affected the amount of vitamins remaining in the emulsion ( $P > 0.05$ ). The loss of oil-soluble vitamins in the emulsion was observed and seemed to depend on the type of vitamins and storage time. Vitamin E acetate was the most stable vitamin compared to others. From the overall results, it would be concluded that stability of emulsion containing EPC could be increased using a mixture of nonionic surfactant (T80) and positively-charged agent (SA). The prepared emulsion provided a promising properties for intravenous delivery of oil-soluble vitamins.

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## LIST OF ABBREVIATIONS

BHT	=	butylated hydroxytoluene
bo	=	blended oil
°C	=	degree Celcius
CNS	=	central nervous system
d(3,2)	=	the surface weighted mean diameter
d(4, 3)	=	the volume weighted mean diameter
d(v, 0.1)	=	the diameter of particles of 10% volume percentile
d(v, 0.5)	=	the diameter of particles of 50% volume percentile
d(v, 0.9)	=	the diameter of particles of 90% volume percentile
DMSO	=	dimethyl sulfoxide
DOTMA	=	dioleyl oxypropyl trimethyl ammonium chlorine
e.g.	=	exempli gratia (for example)
EPC	=	egg phospholipid
et al.	=	et all (and others)
etc.	=	et centera (and so on)
FFA	=	free fatty acid (s)
GC	=	gas chromatography
HDL	=	high density lipoprotein
HLB	=	hydrophilic-lipophilic balance
HPLC	=	high performance liquid chromatography
i.e.	=	id est (that is)
IFCC	=	International Federation of Clinical Chemistry
IU	=	international unit
IUPAC	=	International Union of Pure and Applied Chemistry
IV	=	intravenous
KPa	=	kilopascal
LCT	=	long chain triglyceride
LD <sub>50</sub>	=	lethal dose at 50%
LDL	=	low density lipoprotein

Lot No.	=	lot number
LPC, lyso-pc	=	lysophosphatidylcholine
LPE	=	lysophosphatidylethanolamine
M	=	molality
MCT	=	medium chain triglycerides
mg	=	milligram (s)
M.I.U.	=	million international unit
ml	=	milliliter (s)
mM	=	millimolar
MmHg	=	millimeter Mercury
mOsmol/kg	=	milliosmols per kilogram water
mOsmol/l	=	milliosmols per liter water
mV	=	millivolt
N	=	normality
ND	=	not determined
nm	=	nanometer (s)
No.	=	number of sample
o/w	=	oil in water
o/w/o	=	oil in water in oil
PA	=	phosphatidic acid
PC	=	phosphatidylcholine
PCS	=	photon correlation spectroscopy
PE	=	phosphatidylethanolamine
PFEs	=	parenteral fat emulsions
PG	=	phosphatidylglycerol
PG-E <sub>1</sub>	=	prostaglandin – E
pH	=	the negative logarithm of the hydrogen ion concentration
PI	=	phosphatidylinositol
PL	=	phospholipids
PS	=	phosphatidylserine
psi	=	pound (s) per square inch
R <sup>2</sup>	=	correlation of determination

RDA	=	recommended dietary allowance
RES	=	reticuloendothelial system
SA	=	stearylamine
SD	=	standard deviation
SEM	=	scanning electron microscopic
so	=	soybean oil
SP	=	sphingomyelin
SPC	=	soy phospholipid
T80	=	Tween 80
TEM	=	transmission electron microscopic
TPN	=	total parenteral nutrition
v/v	=	volume by volume
w/o	=	water in oil
w/o/w	=	water in oil in water
w/w	=	weight by weight
w/v	=	weight by volume
$\zeta$	=	zeta potential
%	=	percentage
®	=	registered
>	=	more than
$\geq$	=	equal or more than
$\mu$ l	=	microliter (s)
$\mu$ g	=	microgram (s)
$\mu$ m	=	micrometer (s)